

2<sup>ND</sup> INTERNATIONAL CONFERENCE ON FOOD, AGRICULTURE AND ANIMAL SCIENCES 8-11 NOVEMBER 2019 Antalya, TURKEY



# 2<sup>ND</sup> INTERNATIONAL CONFERENCE ON FOOD AGRICULTURE AND ANIMAL SCIENCES PROCEEDING BOOK

ISBN: 978-605-031-683-4

2079

## INTERNATIONAL CONFERENCE ON FOOD, AGRICULTURE AND ANIMAL SCIENCES

**ICOFAAS 2019** 

ANTALYA, TURKEY

NOVEMBER 8-11, 2019

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### Dear Conference Participants,

Welcome to the 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences.

On behalf of the Organizing Committee, I am very happy to open 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences. I believe that this event, which is the fruit of an intensive and devoted teamwork, will have an invaluable contribution to the scientific world. At the end of busy schedule of nearly one year, we have now achieved to organize this conference under the name of Atatürk University.

The aim of the 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences is to bring together experts and young researchers from all over the world working in food, agricultural and animal sciences to present their researches, exchange new ideas, discuss challenging issues, foster future collaborations and interact with each other.

The main objective of the our conference is to discuss recent results in in food, agricultural and animal sciences and their applications, particularly food, agricultural and aquatic sciences, animal sciences, biodiversity, toxicity and biotechnology. We expect the participation of many prominent experts from different countries who will present best quality papers.

The conference brings together about almost 250 participants from different countries (Algeria, Jordan, Brazil, Belgium, Serbia, India, Iran, Pakistan, Turkey), out of which 124 are contributing to the meeting with oral and with 127 poster presentations, including seven keynote talks.

It is also a goal of the conference to promote collaborative and networking opportunities among senior scholars and graduate students in order to advance new perspectives. Additional emphasis at 2nd International Conference on Food, Agriculture and Animal Sciences is put on applications in related areas, as well as other science, such as natural science, economics, computer science and various engineering sciences. The papers presented in this conference will be considered in the journals listed on the conference websites.

I'd like to express my gratitude to all our authors, members of scientific committee, keynote speakers and contributing reviewers. I believe we will see the best papers of scholars in this event. My sincere thanks go to Prof. Dr. Ömer ÇOMAKLI, the rector of Atatürk University, sets the goal of being also a top-ranking university in scientific sense, for supporting and motivating us in every respect. Special thanks are also due to the organizing committee members, for completing all preparations that are necessary to organize this conference. I express my gratitude to the members of technical committee of the conference for the design and proofreading of the articles. Last but not least, my special thanks go to the our sponsors SFCAGRO LTD. Company, Kotan Biotechnology and Supersol LTD. Company, that unsparingly supports our event financially and emotionally. We wish everyone a fruitful conference and pleasant memories in Antalya, Turkey. Thank you.

> Assoc. Prof. Dr. Fatih DADAŞOĞLU Chair of ICAFOAS 2019

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## Development of Bioformulations w/o Seed Coating for Crop Production and Protection in Agriculture

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## ABSTRACT

Crop production and protection strategies combines different management practices to get maximum yield and quality in agricultural products. Extensive use of of synthetic fertilizers and chemical pesticides in Agricultural production is causing a number of environmental and human health problems in the World. Last 50 years, indiscriminate use of chemical pesticides, fertilizers and hormones in commercial agricultural products forced the scientists to develop alternatives such as sustainable (reduced chemical inputs allowed) and/or organic (no chemical intakes). Therefore, there has been a growing interest in sustainable and organic agricultural practices to alleviate detrimental effects of intensive farming currently practiced. Recent studies showed that many microorganisms called plant growth promoting rhizobacteria (PGPR) in the nature have a profitable role in agriculture by promoting the circulation of plant nutrients, and reducing the need for chemical fertilizers and pesticides. Biostimulants, biofertilizers, and biopesticides may be the alternative products to fully meet the nutrient requirements of soil and crops. Our scientific works at Yeditepe University demonstrated that the PGPRs selected based on *in vitro* and *in vivo* experiments on many agricultural crop species have a very important roles in degrading organic waste, agricultural waste, re-plant nutrient cycling, biological fixation of nitrogen from the air, dissolving phosphate from organic and mineral sources, stimulating plant growth by phytohormone production and nutrient (macro and micro) uptake in the soil, and biological control of plant pests/pathogens. Since 2014, a few commercial biofertilizers and biopesticides developed from the selected PGPR strains at Yeditepe University have been registered and launched into the local and international agricultural market. Bioformulations w/o seed coating may have great potential for crop production and protection purposes in the future.

Keywords: biofertilizer, biopesticide, PGPR, biocontrol, seed treatment, sustainable agriculture.

## Antimicrobial Resistance: Preserving the Efficacy of B-Lactam Antibiotics is a Global Challenge

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### ABSTRACT

β-lactams form a broad class of antibiotics. For over 60 years, β-lactams have been the first line of antibiotic treatments for many community and hospital-acquired infections. Gram negative bacteria have become increasingly resistant to β-lactams, despite the enormous efforts to develop new β-lactam derivatives to overcome growing bacterial resistance, so far no single molecule escapes from hydrolysis by several of the thousands of β-lactamases described [1]. Infections caused by β-lactamases producing GNB are often complicate therapy, limit the therapeutic options and cause treatment failures [2]. Therefore, Early detection of β-lactamases producing GNB is crucial for optimal treatment . However, detection of these enzymes is a great challenge in the clinical microbiology laboratory [3]. our study reveals and confirms the dissemination of carbapenemase and ESβLs-producing Gram-negative bacilli in community and Algerian hospitals. Thus, this constitutes a serious problem because carbapenems are in many cases considered as the last line of therapy.

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## **Current Status of Microbial Biopesticide Studies in Turkey**

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Abstract: Intensive agricultural practices which involves heavy use of chemical fertilisers and pesticides cause deterioration in natural resources and environmental problems threatening sustainability of production systems and human health. Thus, there is a need for new solutions to plant disease and pests problems that provide effective control while minimizing negative consequences for human health and the environment. Biological control, using microorganisms to suppress plant disease and pests, offers a powerful alternative to the use of synthetic chemicals. It is thought that biological control is also the most sustainable and environmentally acceptable control method of plant diseases and pests. Recent advances in molecular biology and biotechnology are inciting the development and application of effective plant disease and pests management programs on the basis of biological control using bacterial organisms. Numerous commercial biopesticide formulations have been prepared and applied to the world today and applied successfully. In conclusion, in our previous studies conducted many different plants species showed that many bacterial strains may be useful as plant growth promoting and/or potential biocontrol agents against some of the plant disease and/or pests. These bacterial strains can be used as new biocontrol agents or biofertilizers. Some of them have been commercialized as microbial biofertilizers (Trade name: SS-Super Root, SS-Super Pan, SS-Super Iron, SS-Super Green, BM-Mega Flu, BM- Coton Plus and BM-Root Pan) but not biopesticide. Therefore, further study is necessary to develop a long-term carrier material, to complete ecotoxicity and toxicity tests of these bacterial strains on target organisms. In this review, basic researches related to the development and production of biopesticides in Turkey are summarized.

Keywords: Bacteria, biological control, microbial biopesticide

## Estimation of Genetic Parameter for Body Measurement in Madura Cattle as Local Indonesian Breed

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## ABSTRACT

Body measurements are used to represent growth and as criteria for selection in Indonesia. Those important traits should be genetically improved. This research was conducted to estimate the heritability for body measurement at birth and weaning age in Madura cattle in the Technical Service, Pamekasan, East Java, Indonesia. Chest Girth (CG), Body Length (BL) and Wither Height (WH) were measured and recorded. Data were analysed using analysis of variance applying software GENSTAT 18. The heritability for WH at birth and weaning age were high (0.54 and 0.40, respectively). The heritability for BL showed medium heritable: 0.25 at birth and 0.22 at weaning age. While the heritability for CG was high at birth (0.59), but low at weaning age (0.07). Selection based on trait with medium to high heritability value enhance the genetic response to selection.

## ACKNOWLEDGEMENT

This work was supported by Faculty of Animal Science, Universitas Brawijaya, Malang, Indonesia.

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## Pesticides in Surface Waters – Monitoring with Passive Sampling Techniques

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## ABSTRACT

The presence of pesticides in the environment has become of great concern. Their intensive application in agricultural activity especially endangers aquatic environments. Runoff over fields, spray drift and infiltration caused by precipitations are the major causes of the presence of these agrochemicals in surface and ground waters, and as a consequence, residues of some harmful pesticides can be detected in water samples. According to the Water Framework Directive 2013/39/EU [1], priority pollutants comprise an important number of pesticides.

Due to all mentioned, it is necessary to carry out a continuous monitoring program of pesticides in water. However, most studies that report the occurrence of these contaminants in water were conducted using traditional sampling methods. Recently, passive sampling, a new innovative method for sampling of a wide range of pesticides, as well as other POPs, is applied. This contemporary technique is characterized by simple construction and handling, low price and the possibility of multiple uses. The base of the technique is the accumulation of analytes in corresponding sorption material in a device for passive sampling.

The use of passive sampler for the determination of pesticides in the aquatic environment requires a sampling rate (Rs) – a number of liters of water per day that are sampled 'through' the sampler during the exposure time, and it is specific for each compound. However, for most of the pesticides there is a lack of data on the sampling efficiency and sampling rates. The available data on Rs, obtained by laboratory or in-situ calibration of devices for passive sampling, refer to a smaller number of pesticides, mainly those from the WFD list of priority pollutants. Thus, it is of great importance to calibrate passive samplers in water-flow calibration systems in order to obtain Rs values for the analysis of a large number of pesticides.

Keywords: water, pesticides, passive sampling, monitoring

## ACKNOWLEDGEMENT

This work was supported by Ministry of Education, Science and Technological Development of the Republic of Serbia, No. III43005.

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## Influence of Type of Sex and Livestock Breeding System on Features of Carcass and non-Carcass of Dorper Crossbred

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## Abstract

The aim of the present study was to analyze LW, CW, DP and non-carcass components by sex and livestock breeding system The evaluated a total of 20 crossbred ( $\frac{1}{2}$  Dorper +  $\frac{1}{2}$  Santa Inês), with 7 ewe raised free in the pasture, 7 ewe kept in feedlot and 6 mutton kept in feedlot. The results were submitted to Pearson and Spearman correlation analysis. The sex and livestock breeding system (feedlot or grazing) were considered as fixed effects. Thus, for each sheep, the fixed effects were compared on each analyzed characteristic, using the *t* test at 5% probability. Thus, the variables carcass weight, carcass yield, lung weight, liver weight, tallow weight and N-car were not significant (*P* >0.001). The highest value for Pearson correlation was between blood weight and liver weight (0.9741), with a positive value. In general, all characteristics had correlation values greater than 0.7. There was no statistical difference between sex or livestock breeding system (grazing or feedlot) by t-test at 5% probability.

Keywords: Live weight, sex, viscera

**Abbreviations:** LW = Live Weight before slaughter, CW = carcass weight, DP = dressing percentage, G = grazing, F = feedlot, N-car = non-carcass components

### **1.Introduction**

Organs and viscera compared to other parts of the animal body, has different growth speed and are mainly influenced by the chemical composition of the diet and its energy level [1]. The ruminant diet may also influence the development of non-carcass components (N-car), mainly of those more related to digestion, the more fibrous the sheep diet the more the non-enzymatic part (rumen, reticulum and omaso) of the ruminant stomach develops [2]. The importance of non-carcass components is not only related to the possibility of increasing the economic return at the time of commercialization of sheep farming products [3]. In study above effects of sex, slaughter weight and carcass weight on carcass characteristics and meat quality traits were evaluated using 100 Segurena lambs in Spain, with the management of all lambs was similar prior to slaughter at 19-25 kg. Slaughtered animals with a hot carcass weight below 20 kg were assigned to class B, and those greater than 22 kg to class C. Carcass weight had a significant influence on "non-carcass" components, percentage (DP), subjective dressing carcass conformation, fat deposits, carcass fatness, bone and most carcass measurements [4]. Sex had a significant effect on age at slaughter, "non-carcass" components, rib measurements, dressing percentage, fat deposits, and neck and shoulder percentage. The males had carcass heavier than the females [5]. However, female carcasses have a higher proportion of fat [6]. As the weight increase, the carcass measurements also increase. While improve the conformation indices of the carcass, leg and dressing percentages, neither the commercial cuts of the animal nor tissue composition was significantly affected. Sex primarily affected the quantity of all types of fat deposits [4]. The difference in the body weight between males and females also interfered in the weights of the rumen/reticulum, small intestine and blood and males were heavier [7].

When compared different breeds with slaughter data from 304 Menz and 153 Horro ram lambs were analyzed to determine between-breed differences in yields of carcass and edible non-carcass components. Average age at slaughter was 515 days. Total edible non-carcass component yield was obtained as the sum of weights of blood, lungs (with trachea), liver (with gall bladder). heart, kidneys. reticulorumen, omaso-abomasum, hind gut and abdominal and kidney fats. Total yield of usable products was calculated as the sum of hot carcass weight and non-carcass component. The difference between breeds in non-carcass component was statistically significant (p0.01). Additionally, the Menz dressed higher (p0.01) than the Horro [8]. A study on carcass composition and meat quality compared in yearling sheep and goats. The yearling sheep had higher (P < 0.05) pre slaughter weight, hot carcass weight and DP than the goats. The muscular development as indicated by loin eye area was significantly (P < 0.01) greater in sheep than goats [9]. Dorper crossbred sheep are widely used for crossbreeding which results in fast growing animals and satisfactory daily weight gain [10]. The aim of this study is the present study was to analyze live weight (LW), carcass weight (CW), DP and non-carcass components by sex and livestock breeding system of Dorper crossbred.

#### 2. Method

#### 2.1 Experimental site

The experiment was conducted in Fazenda Rancho Alegre, Posse city, Góias State, Brazil (average altitude of 811 m, at 14°5′34″S, 46°22′8″W). The climate of the region is classified as type Aw by the Köppen classification, with an average annual rainfall of 1200-1500 mm.

### 2.2 Animal management

The evaluated a total of 20 crossbred ( $\frac{1}{2}$  Dorper +  $\frac{1}{2}$  Santa Inês), with 7 ewe raised free in the pasture, 7 ewe kept in feedlot and 6 mutton kept in feedlot.

The animals raised in Coastcross (*Cynodon dactylon*) pastures, with free access to mineral salt. Feedstuff used in feedlot (**Table 1**). Diets were formulated according to the Cornell Net Carbohydrate and Protein System software [11]. The LW was measured with restrict the animals of feed and water for 12h.

**Table 1**. Percentage composition of the diets on a DM basis

Ingredients	Diet used in feedlot (%)	
Coast Cross hay	60.2	
Corn	12	
Soybean meal	18.1	
Urea	6	
Sulfur	1.2	
Mineral mix <sup>a</sup>	2.4	

<sup>a</sup>Composition per kilogram: P = 60 g; Ca = 180 g; Mg = 5 g; S = 17 g; Na = 135 g; Cu = 650 mg; Mn = 500 mg; Zn = 2400 mg; I = 48 mg; Co = 38 mg; Se = 12 mg.

Characteristics evaluated:

LW = Live weight (kg) = before slaughter, obtained the last time that the animals were weighed at the farm, after a 14-h solid food fasting.

CW= carcass weight (kg) = after slaughter. DP = Dressing Percentage (%) = (Carcass Weight / Live Weight) x 100. Dressed carcasses of animals were weighed to obtain a hot carcass weight and then chilled and the cold carcass weight was recorded the day after slaughter.

N-car = Non-carcass components (kg) = The weight of non-carcass components was obtained from the sum of edible components (blood, lung + trachea, heart, liver, kidneys, empty gastrointestinal tract, tallow, except head and feet).

The carcasses were divided into right and left carcass, after performing cuts in 5 anatomical regions (shoulder, leg, neck, loin and ribs) according to methodologies described by [12].

#### 2.3 Statistical analysis

The data were submitted to the normality test (Shapiro-Wilk). After verifying if the data follow a normal distribution N ( $\mu$ ,  $\sigma^2$ ), the results were submitted to Pearson and Spearman correlation analysis.

Pearson correlation coefficient is a measure of the linear correlation between two variables *X* and *Y*. It has a value between +1 and -1.

where:

 $\rho = 1$  is total positive linear correlation;

 $\rho = 0$  is no linear correlation;

 $\rho = -1$  is total negative linear correlation.

$$\rho = \frac{\sum (x - \overline{x})(y - \overline{y})}{\sqrt{\sum (x - \overline{x})^2 \sum (y - \overline{y})^2}}$$

The Spearman correlation coefficient is defined as the Pearson correlation coefficient between the ranked variables, according recommendation by [13].

$$\rho = 1 - \frac{6\sum_{i=1}^{n} d_i^2}{n^3 - n},$$

where:

 $d_i$  = (positions of  $x_i$  between the values of x) - (positions of  $y_i$  between the values of y). If the positions of x are exactly equal to the positions of y, then all  $d_i$  will be zero and  $\rho$ will have value 1.

*n* is the number of pairs  $(x_i, y_i)$  and

The sex and livestock breeding system (feedlot or grazing). were considered as fixed effects. Thus, for each sheep, the fixed effects were compared on each analyzed characteristic, using the t test at 5% probability. All statistical analyzes were submitted to statistical software SAS, 9.0 software [14].

#### 3. Results

For the analysis of the Pearson and Spearman correlations it is necessary that the data have a normal distribution. Thus, the test of Shapiro-Wilks was used. Among the characteristics analyzed, the variables carcass weight (CW), carcass yield (CY), lung weight, liver weight, tallow weight and N-car weight were not significant (P>0.001). Thus, the normality of the errors was assumed to analyze the variables (**Table 2**).

**Table 2.** Correlations of Pearson (above diagonal) and Spearman (below diagonal) for the characteristics analyzed.

Item	LW	CW	DP	Blood	Heart	Lung	Liver	Kidney	Tallow	N-car
LW		0.9278	-0.4922	0.8191	0.6660	0.7030	0.7864	0.7105	0.5321	0.8156
CW	0.9251		-0.1461	0.7380	0.6125	0.6371	0.7382	0.6756	0.6757	0.8172
DP	-0.3864	-0.0993		-0.5093	-0.4115	-0.4434	-0.4380	-0.3644	0.1270	-0.3215
Blood	0.7634	0.7168	-0.3475		0.8948	0.9439	0.9741	0.8740	0.4103	0.9206
Heart	0.7517	0.7104	-0.3646	0.9684		0.9332	0.9102	0.8967	0.4136	0.8700
Lung	0.7590	0.6950	-0.4179	0.9740	0.9612		0.9704	0.8457	0.4427	0.9171
Liver	0.7917	0.7254	-0.3819	0.9910	0.9653	0.9842		0.8820	0.5020	0.9539
Kidney	0.7444	0.6945	-0.3672	0.8649	0.9039	0.8549	0.8722		0.4240	0.8469
Tallow	0.5644	0.6657	0.2430	0.4962	0.4465	0.4719	0.4815	0.3828		0.7254
N-car	0.8256	0.8015	-0.2526	0.9447	0.9220	0.9352	0.9416	0.8209	0.7191	

LW = Live Weight before slaughter, CW= carcass weight, DP = dressing percentage, N-car = non-carcass components

The highest value for Pearson correlation was between blood weight and liver weight (0.9741), with a positive value. In general, all characteristics had correlation values greater than 0.7, which is considered high, according to [15] and demonstrates the degree of linear statistical dependence between the variables. Only the DP characteristic was negatively correlated with moderate values. Variables obtained through the ratio of other variables, besides have linear dependence between the parameters, may result in negative correlation when compared to other characteristics that have a high correlation with the original characteristics used in the ratio. Thus, the data suggest that higher values for weight of each viscera separately implies lower values of DP, which is expected. However, this correlation is expected to be negative and high, when in fact it was moderate to low. In this case, the slaughter weight does not determine whether an animal will have higher DP. Neither the viscera weight is determinant for higher DP.

The tallow weight characteristic has a positive mean correlation between all other characteristics, except for carcass yield, which does not have the same behavior when compared to the other variables.

Spearman's correlation performed similarly to the estimated Pearson correlation values. The highest positive and high correlation was also between liver weight and blood weight. Correlation values in this paper are important and can be used to estimate viscera weight values. This implies a future possibility of selection of animals for viscera size, as an improvement strategy. Normally, animals submitted to selection have higher viscera values, which may compromise the carcass yield, which was not observed in this study. In Figure 1 shows the Distribution to non-carcass components (kg), which facilitates the understanding and graphic visualization of the sum of viscera with the exception of the sheep's head and feet. And in Figure 2. Shows the Probability to non-carcass components after statistical analysis.

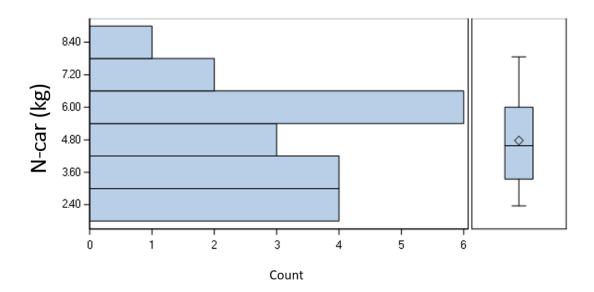


Figure 1. Distribution to non-carcass components

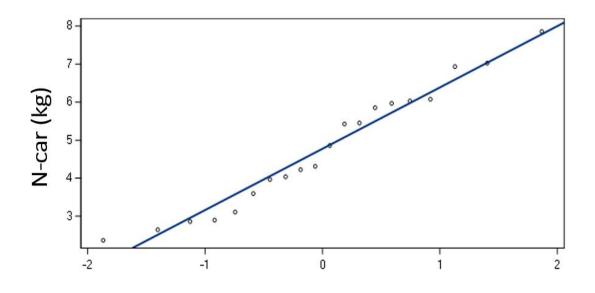


Figure 2. Probability to non-carcass components

The values obtained in **Table 3** were through the least squares, where the fixed effect of sex and livestock breeding system was considered. There was no statistical difference between sex by t-test at 5% probability. There was no statistical difference between livestock breeding system (grazing or feedlot) by t-test at 5% probability. Although there is no difference, higher values for LW of females can be explained by the weight of reproductive organs, which in females are larger, however, the same cannot be observed for the other characteristics.

**Table 3**. Means and standard deviation of the characteristics analyzed shared by sex and livestock breeding system obtained by the least squares methods.

Effect		LW*	CW*	DP*	Blood*	Heart*	Lung*	Liver*	Kidney*	Tallow*	N-car*
Sex	Male	37.74±4.8	17.10±2.0	45.90±2.1	1.76±0.3	0.21±0.0	0.53±0.1	0.98±0.1	0.16±0.0	0.55±0.2	4.20±0.8
	Female	45.38±2.6	19.47±1.1	43.41±1.9	1.91±0.1	0.20±0.0	0.55±0.0	1.06±0.0	0.16±0.0	1.17±0.1	5.05±0.4
Sys	G	45.80±4.6	19.40±2.0	42.86±2.1	2.05±0.3	0.21±0.0	0.57±0.0	1.08±0.1	0.17±0.0	0.62±0.2	4.69±0.7
	F	37.32±2.7	17.17±1.2	46.45±1.2	1.62±0.1	0.20±0.0	0.52±0.0	0.97±0.0	0.16±0.0	1.10±0.1	4.56±0.4

Effect Sex = Sex, Effect System = Sys, LW = Live Weight before slaughter, CW= carcass weight, DP = dressing percentage, G = grazing, F = feedlot, N-car = non-carcass components

Means followed by \* do not differ from each other within the same column, by *Student's* t test at 5% probability.

### Discussion

Weight at slaughter had a significant influence on intermuscular fat and bone, which corresponded to their allometric growth. The quantity of subcutaneous fat also increased with slaughtering weight. Given the great variability among animals, the difference was not statistically significant [4]. Similar result to [16] evaluated the characteristics of carcass and commercial cuts of Santa Ines lambs of different biotypes (traditional and modern) slaughtered at 32 kg and 34 kg. The biotypes were on average 180 days old and weighed 16 kg initially. The traditional biotype influenced significantly (P<0.05) the carcass yields, conformation, and marbling, compared to the modern biotype. The biotype did not influence (P>0.05) carcass morphometric measurements and commercial cuts. On the other hand, the 34-kg slaughtering weight affected most of carcass characteristics. The traditional biotype displayed higher yield, better conformation and greater amount of intramuscular fat in the carcass. According to [17] fat deposition varies with the nutritional status, age, slaughtering weight, and genotype. However, it is likely that the diet with more concentrate has caused an increased deposition of fat on the carcass. Thus, increased the concentrate level of the diet supplied to Dorper x Santa Ines crossbred sheep and observed an increase of the amount of fat deposited on the carcass.

Weight class had influence mainly by slaughter weight, and carcass fatness varied according to sex. Therefore, the producer could increase the returns from breeding and fattening by determining the appropriate time for slaughter [4]. The higher slaughtering weight influenced carcass characteristics and the amount of fat. Generally, the carcasses presented high muscle proportion and fat amount appropriate to the consumer [16]. The conformation and finish are highly correlated with subcutaneous adipose tissue and the accumulation of cover fat occurs simultaneously to the deposition of pelvic and kidney fat in hair lambs [18].

Carcass grades in the present study were significantly better (p < 0.01) for Dorper than for Red Maasai. The scores for muscle formation were also slightly higher for Dorper. As this study, which comprised a limited number of purebred lambs regarding comparison of carcass traits of Red Maasai and Dorper sheep, and since carcass weights

unexpectedly did not differ between the breeds, further studies on slaughter lambs of Red Maasai and Dorper lambs are warranted [19].

Sex affected the proportion of commercial cuts only in the neck and back, which were more developed in the males. The carcass contained 22% fat, 54% muscle and 20% bone. Carcass muscle content was approximately 54.5% for carcasses of less than 10 kg and 53.9% for those heavier than 10 kg; the bone percentage varied between 20.3 and 19.7%, and that of fat 21-23% [4].

In a study on weights of empty body, hot and cold carcasses, total edible offal and total usable products were highly correlated with the slaughter weight and within each other (p<0.001) to ram lambs. Compared correlation coefficients among slaughter weight and carcass traits, although the associations between the dressing percentage and the slaughter weight or the total edible offal. This difference could be attributed the fed with maize stover form that had higher values of slaughter weight and of the non edible offal relative to the empty body weight and to the weights of hot and cold carcasses, leading to the decrease of the dressing percentage. The highly significant and strong positive correlations of the slaughter weights with the empty body weights or with the weights of carcasses confirm this fact [20].

In general, total non-carcass fat contents were more in sheep than goats. Similarly, the dissected total fat of half carcass was also more (P < 0.01) in sheep than the goats. Neck and shoulder portion was heavier (P < 0.01) in goats than the sheep. Shear force value was greater (P < 0.01) in goats (7.42 kg/cm<sup>2</sup>) than sheep (3.74 kg/cm<sup>2</sup>). Goat meat had more (P < 0.01) moisture and less fat than mutton. In sensory evaluation both the species were rated almost equal in overall palatability scores. The current study showed that dressing yield was higher in sheep than goats. But goat yielded leaner carcass which is desirable for the calorie concern consumers. It was also revealed that meat from goat carcasses was tougher than mutton [9].

The factors that influence the non-carcass components of lambs are variable and contradictory, which requires further research to encourage their use and add value to sheep systems for meat production [21].

## Conclusion

Feeding had no influence on sheep carcass yield. However, animals of both sex in feedlot system had higher numerical value for carcass yield and tallow weight when compared to grazing animals, but without statistical difference, and the economic analysis for financial viability should be made in each case specific.

The non-components of sheep carcass are variable and unstable due to the weight of each organ, which resulted in inaccurate data, which reinforces the need for further research for a more robust conclusion for sheep meat production.

## Acknowledgment

The research is financed by IFGoiano and FAPEG Go state, Brazil.

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## The Potential Use of Biochar to Enhance Soil Properties and Plant Growth Performance under Arid Environment

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#### Abstract

This study was conducted to determine the effect of biochar on plant growth and soil properties. Two experiments were done: pots experiment under greenhouse condition at the Faculty of Agriculture, Mutah University on tomato and bell pepper plants using biochar produced from the broccoli residues in five levels (control, 0.5, 1.0, 2.5 and 5.0% by weight), and field experiment which was conducted at Ghor Al-Safi, NCARE research station in the Jordan Valley using five levels of biochar produced from the olive pruning residues (control, 8, 16, 30, 40 ton ha<sup>-1</sup>). The pots experiment showed that addition of biochar resulted in an increase of tomato and bell pepper plants nutrients content with biochar application. In addition, the soil pH and EC values increased with biochar application. The results of the field experiment showed that leaf N and P content of tomato and bell pepper increased with biochar application. Moreover, the addition of biochar increased soil pH and EC. Thus, this present study suggest that biochar can be used as a soil amendment; however, still several studies are needed to investigate the effect of biochar feedstock, pyrolysis conditions on different soil, climate and plants.

Keywords: Pyrolysis, sustainable waste recycling, soil fertility, water availability, biochar.

#### Introduction

Soil degradation may cause a significant loss in food production and rural livelihood, mainly in poor and densely inhabited areas in much of the world. Appropriate policies are needed to motivate land-improving investments and better land management to sustainable meets the food needs (Mohawesh, 2016). The potential uses of soil amendments to increase soil water and nutrient holding capacity become an important issue over time, especially in regions of reduced water availability and fragile soil ecosystem such as most of the Middle East region (Mohawesh and Durner, 2017). Biochar is a recent soil amendment that being tested and suggested to enhance carbon sequestration in soils and boost soil properties. Improved soil carbon sequestration can enhance soil chemical, biological, and physical indices due to carbon effects on soil process (Laird, 2008). Biochar is a byproduct of the pyrolysis processing of organic feedstocks (Novak et al., 2014). Biochar is a carbon-rich solid obtained by heating or charred the organic biomass at low levels or in the absence of

oxygen (pyrolysis or charring) under different conditions and feedstocks, resulting in products of varying properties (Novak et al., 2009; Sohi, 2012). Biochar can persist in soils and sediment for a long time (Downie et al., 2011), and can possibly enhance agronomic production when applied as a soil amendment (Lehmann and Rondon, 2006).

Biochar applied to soil has been indicated to have numerous agricultural advantages, i.e. increase soil holding capacity, decrease nutrient loss by surface and groundwater runoff, and a continuous arrival of nutrients to the growing plant (Laird, 2008). This indicates that soil amendment with biochar may enhance crop productivity by holding more water from the rainfall in arid and semi-arid regions and reducing the recurrence or quantity of irrigation water in the irrigated areas. Biochar addition has shown certain positive increases in cation exchange capacity (CEC), pH (Topoliantz et al., 2002) and enhanced the nutrients retention of soils (Verheijen et al., 2010).

Growing vegetable crops in arid and semi-arid regions such as Jordan faces different kinds of challenges, which include both biotic and abiotic issues. These problems are poor soils fertility, low water holding capacity, and low organic matter (Al-Karaki and Al-Omoush, 2002; Ouda and Mahadeen, 2008). To overcome low soil fertility, farmers usually apply high amounts of chemical fertilizers to enhance crop growth and productivity (Al-Karaki, 2002). Glaser et al. (2002) and Lehmann et al. (2003) had stated that biochar can be advantageous as a soil amendment for improving the quality of the agricultural soils. Hammond (2009) reported that the different types of biochar in terms of the source and production systems affects differently depending on the type of soil, climate, and different crops. Most of the researchs about the beneficial effects of the biochar were conducted on the tropical regions such as USA, East Asia, and Europe. Therefore, the current study aimed to investigate the effects of different application levels of biochar as a soil amendment on soil properties and plant growth performance under arid and semi-arid environment.

## **Material and Methods**

## Pot Experiment

The experiment was conducted at the Faculty of Agriculture, Mutah University, Al-Karak, Jordan, (Elevation 920 m a.s.l, Longitude  $35^{\circ}45$  E and Latitude  $31^{\circ}16^{\circ}$  N), at a greenhouse during the period from June to November, 2015. The experiment was repeated two times. Each experiment was terminated after 75 days from transplanting date. Four-week-old seedlings of tomato and bell pepper were transplanted into 20 cm diameter plastic pots, containing about 5 kg of sandy loam soil (Clay 14%, Silt 18 %, Sand 68 %, EC 0.48 dS m<sup>-1</sup>, pH 7.3, organic matter 1.09 %, total nitrogen 980 ppm, CaCO<sub>3</sub> 47%, available potassium (K) 42 ppm, available phosphorous (P) 30 ppm) with five treatments based on dry weight bases (0.0, 0.5, 1.0, 2.5 and 5.0% by weight) in triplicates. All pots were received an equal amount of irrigation. The irrigation was initiated when 50% of available water (readily available water, RAW = 50% of available water, AW) was depleted. Soil samples were collected from each pot at two times (Mid and at the end of the experimental period) for pH and EC measurements in a 1:1 (soil: distilled water) ratio. The soil samples were dried, ground and then sieved through a 2 mm sieve. Soil texture was determined using the

Proceedings of the 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences (ICOFAAS 2019) Antalya, Turkey, 8-11 November 2019 hydrometer analysis (Klute, 1986). Soil chemical properties were analyzed according to the standard procedures of the United States Salinity Laboratory Staff.

#### Biochar production and characterization

The biochar was produced using a pyrolysis apparatus. The broccoli crop residues were used for biochar production as feedstock. The collected samples were piled inside a greenhouse to dry (moisture content less than 10%). The biochar was obtained from a slow pyrolysis process at 350-400 °C. The produced biochar was grinded (2-3 mm) prior to experimental use. The pH and EC of the biochar were determined in a 1:10 (biochar: distilled water) ratio (Cheng and Lehmann, 2009). Scanning electron microscope (SEM) with energy dispersive spectroscopy (EDS) were used to determine the chemical composition and micrographs of the biochar.

## Plant measurements and analysis

Leaf samples were collected at the end of the experiment to determine chloroplast pigments, proline content, leaf nitrogen (N), phosphorus (P) and potassium (K) contents in triplicate. Leaf samples were stored in plastic bags with wet tissue paper at 4 °C. Measurements were made using all leaflets within 4-6 h of cutting to estimate leaf chlorophyll content (Vemmos, 1994). The method of Bates et al. (1973) was used to measure leaf proline content on dry basis (mg g<sup>-1</sup>). Each leaf sample was dried at 75 °C for one week, ground, and then the samples were used for N, P and K determination. Available P was determined by the vanadate-molybdate method (Olsen, 1954) using a UV/visible spectrophotometer (UV-1601PC, Shimadzu, Japan). Available K was measured using atomic absorption (Perkin Elmer AAnalyst 300, USA) (Knudsen et al., 1982). Total N was determined in samples by the Kjeldahl method (Chapman and Pratt, 1982). The Cation Exchange Capacity (CEC) was determined using ammonium acetate method (Chapmen, 1965). Finally, the plants were removed from the pots and the roots were washed to remove soil particles. Plant, shoot and root fresh and dry weights were measured. The plants, shoots and roots were dried at 75 °C for 4 days.

## Field Experiment

The experiment was conducted at NCARE Research Station, Ghor Al-Safi, Jordan Valley (Elevation -387 m b.s.l, longitude  $30^{\circ}50^{\circ}$  E and latitude  $30^{\circ}20^{\circ}$  N ) from September 2016 to February 2017 at two locations. Tomato and bell pepper plants seedlings were used. The soil was prepared for planting, disking and leveling. Drip irrigation was used according to the common agricultural practices in the area. Recommended agricultural practices were followed in the experimental field throughout the growing season. The biochar was applied for each treatment. Biochar application levels were 0, 8, 16, 30 and 40 ton ha<sup>-1</sup>. Physical and chemical properties of soil are shown in Table (1). Each plot area was  $3.2 \text{ m}^2$  (3.2 m length and 1m width).

Table 1: Physical and chemic	al properties of field e	experiment soil.

рН	7.7
EC (dS $m^{-1}$ )	0.65
$CEC(m_{eq}/100g)$	22.7
N (%)	0.098
P (ppm)	10
K (ppm)	23
Texture class	Sand clay loam

Biochar production and characterization

The same pyrolysis process was used as the pot experiment biochar; however, the feedstock was olives trees pruning residues.

## Plant measurements and analysis

Leaf chlorophyll content, RWC, plant biomass, fruit weight and total yield were measured. Soil pH and EC were recorded at the mid and the end of the experiment. At the end of the experiment, leaves and fruits were analyzed for N, K and P.

## Experimental design and statistical analysis

The experimental design of pot and field experiments was randomized complete block design (RCBD) in triplicates. SPSS version 16 was used for statistical analyzes of the data (SPSS, Inc., Chicago, IL, USA). Means separation were done using the least significant difference (LSD) test and t-test at 0.05 level of significance.

## **Results and discussion**

## **Biochar characterization**

The produced biochar characterization from broccoli residues and olive trees pruning residues are shown in Table (2). Broccoli residues gave higher values of EC, N, K, and P contents, while the biochar produced from olives trees' pruning residues had a higher CEC, C, O, and Ca contents. The biochar is alkaline for both sources. The CEC of previous biochar sources were (29.6 and 33.9  $m_{eq}/100$  g soil), which is lower than the typical CEC of humified organic matter which is around 200-300  $m_{eq}/100$  g soil. Zhao et al. (2013) showed that CEC varied greatly among biochar produced from different feedstocks. This may be because CEC is related to cations (e.g., K and Ca) present in biochar, which vary greatly with feedstock.

The elemental composition of the final material is essentially dependent on the feedstock type and the pyrolysis temperature. Scanning Electron Microscope (SEM) of biochar produced after charring broccoli and olives trees pruning residues are shown in Figures (1 and 2), respectively. Scanning electron microscope is an effective way to study structural characteristics of biochar. The microscopic physical structure of biochar is one of the key properties related to its effect on soil properties. The surface area of the charred source material can be increased several thousand fold and greatly enhance its water holding capacity (Mohawesh and Durner, 2017).

The physical structure of biochar, such as pore volume, and average pore size are typically related to its sorption and water holding capacity that, in turn, relates to its effect on soil structure, contaminant mobility, and microbial interactions (Zhao et al., 2013). A few researchers considered that specific essential properties of the derived biochar are specifically attributed to the properties of the type of feedstock. For example, Hamer et al. (2004) showed that biochar from corn was fine, friable, and easily broken down. Conversely, biochar from oak was hard and more resistant to external pressure. This corresponds to the biochar produced from broccoli residues and olives trees pruning residues, respectively. Figures (3 and 4) are shown the energy dispersive spectroscopy (EDS) for biochar produced from broccoli and olive trees pruning residues.

Table 2: Characterization of blochar produced from two different sources.							
Biochar/Broccoli residues	Biochar/Olives pruning residues						
9.5	9.5						
8.8	6.5						
29.6	33.9						
3.5	1.4						
1.9	0.69						
17.01	2.55						
59.85	76.43						
6.76	10.08						
4.82	9.94						
	Biochar/Broccoli residues 9.5 8.8 29.6 3.5 1.9 17.01 59.85 6.76						

Table 2: Characterization of biochar produced from two different sources.

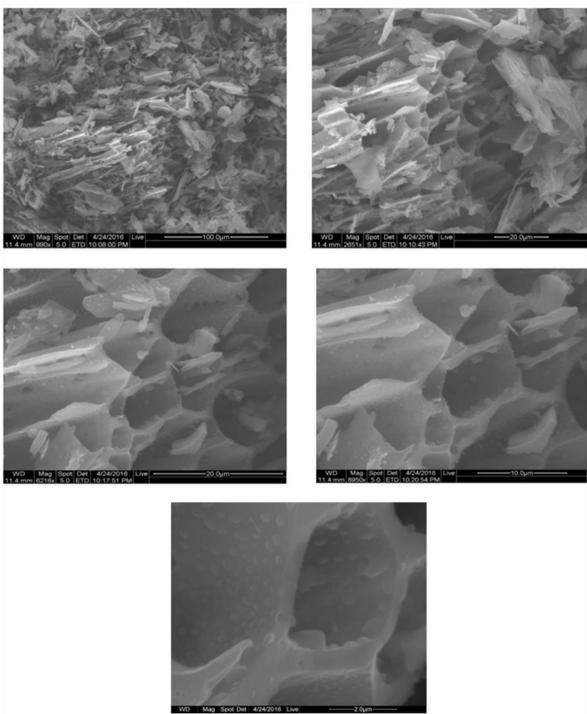


Figure 1. Scanning electron microscope of the produced biochar from broccoli crop residues.

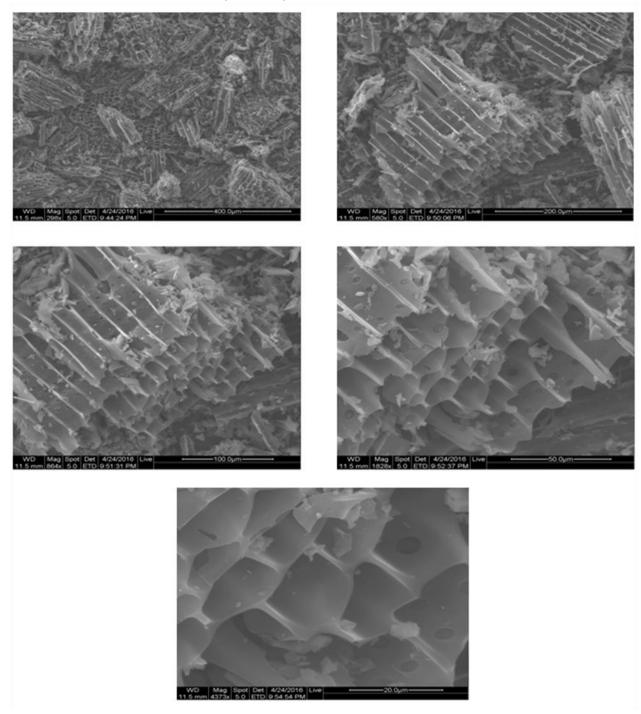


Figure 2. Scanning electron microscope of produced biochar from olives pruning residues.

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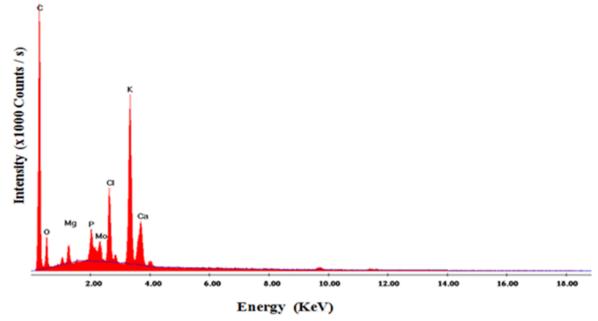


Figure 3. EDS of produced biochar from broccoli crop residues.

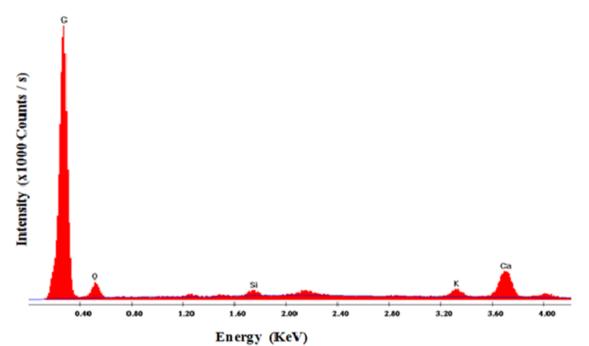


Figure 4. EDS of produced biochar prepared from olives pruning residues.

Biochar (%)	SFW (g)	SDW (g)	RFW (g)	RDW (g)
0	137.83 a	14.99 a	12.41 a	3.63 a
0.5	141.11 a	14.33 ab	10.43 a	2.56 b
1	140.83 a	14.98 a	10.68 a	2.44 b
2.5	119.17 b	13.10 ab	11.58 a	2.97 ab
5	115.83 b	12.64 b	9.35 a	2.10 b

Pots experiment

Table 3: Effect of biochar levels on shoot and root fresh and dry weight of tomato.

The effect of biochar treatments on tomato growth is shown in Table (3). The shoot fresh and shoot dry weight were significantly decreased at 5% of biochar level. Root fresh weight was decreased with biochar addition; however, root dry weight was not affected with biochar addition. At 0.5 and 1% levels, there was an increase in shoot fresh and dry weight. Table (4) shows the effect of biochar on shoot fresh weight, shoot dry weight, root fresh weight, and root dry weight of bell pepper. The 0.5% and 1.0% levels showed an increase in the measured parameters, however at 2.5% and 5% exhibited a decrease in the measured parameter compared to control.

The data presented in Table (5) shows the effect of different levels of biochar on the minerals concentration on tomato and bell pepper leaves. The biochar increased the shoots concentration of the measured minerals on tomato at the lower levels of biochar. For bell pepper plant, the biochar increased shoots nutrients concentrations.

It is clear that the adding of biochar led to a significant increase of pH values at the mid and at the end of the experiments. The 5% level of biochar gave a significant increase compared to control (Table 6). As for the soil EC, there was an increase of soil EC with increasing the biochar levels. At the end of experiment, the EC was increased significantly at 2.5 and 5% biochar levels.

 Biochar (%)	SFW (g)	SDW (g)	RFW (g)	RDW (g)
 0	34.06 ab	6.54 ab	10.18 ab	3.97 b
0.5	38.69 a	9.32 a	12.41 a	5.74 ab
1	38.36 a	8.75 a	11.91 a	5.95 a
2.5	29.58 b	7.76 a	9.36 b	4.66 ab
 5	16.21 c	3.87 b	7.13 c	1.52 c

Table 4: Effect of biochar levels on shoot and root fresh and weight of bell pepper.

Biochar	Ν	Р	Κ	Ca	Na	Ν	Р	Κ	Ca	Na
(%)			(%)					(%)		
Tomato shoots							Bell	pepper s	hoots	
0	2.69a	0.30b	1.39ab	0.99ab	0.09a	3.7ab	0.26ac	1.17b	1.55ab	0.09bc
0.5	2.79a	0.46a	1.66a	0.84ab	0.10a	3.7ab	0.24ac	1.53ab	1.29ab	0.10bc
1	2.65a	0.42ac	1.53ab	1.18a	0.12a	3.9 a	0.21bc	1.49ab	1.00b	0.08c
2.5	2.88a	0.35bc	1.40ab	0.97ab	0.11a	3.8 a	0.29a	1.69a	1.81a	0.13ab
5	2.74a	0.36bc	1.27b	0.73b	0.10a	3.5 b	0.27ac	1.43ab	1.27ab	0.15a

Table 5: Effect of biochar levels on N, P, K, Ca, and Na concentrations in tomato and bell pepper plants shoot.

Table 6: Effect of biochar levels on soil pH and EC at the middle and end of the experiment.

Biochar (%)	pH <sub>mid</sub> (-)	pHend (-)	$EC_{mid}$ (dS m <sup>-1</sup> )	EC <sub>end</sub> (dS m <sup>-1</sup> )
0	7.25 e	7.37 e	0.53 c	0.60 c
0.5	7.39 d	7.49 d	0.72 bc	0.96 c
1	7.46 c	7.56 c	0.61 c	0.87 c
2.5	7.55 b	7.61 b	0.90 b	1.27 b
5	7.65 a	7.77 a	1.55 a	1.48 a

## **Field experiment**

Tables 7 and 8 show the effect of biochar application on tomato and bell pepper plants yield, respectively. The biochar affected tomato plant yield, an increase in tomato yield was observed at 8 tons ha<sup>-1</sup> and a decrease with other biochar levels. However, the plant bell pepper yield was increased with the increasing of biochar level compared to control. The increase may be due to the high concentrations of available nutrients found in the biochar. Tomato fruit yield increased with application of biochar (Yilangai et al., 2014). Biochar also increased vegetable yield by 4.7-25.5% as compared to farmers' practice (Vinh et al., 2014). Biochar positively influenced growth and yield of French bean (Saxena et al., 2013). On the other hand, biochar didn't increase annual yield of winter wheat and summer maize but the cumulative yield over four growing seasons significantly increased in a calcareous soil (Liang et al., 2014). In a study conducted on the response of dry matter production of radish using green wastes, it was established that a yield increases were only found at biochar application rates greater than 50 ton ha<sup>-1</sup> (Chan et al., 2007).

The effect of biochar on nutrients contents of tomato and bell pepper leaves are shown in Table (8). There was a significant increase of N and P content with biochar application. The significant increase of N, K, and P were found at 40, 8, 30 tons ha<sup>-1</sup>, respectively. Zeelie (2012) indicated that different soil-biochar mixtures had no significant effect on the leaf N content. However, it is likely that the reduced growth was not due to a lack of N

uptake as the leaf N content was high. Furthermore, the differences in leaf N content in bell pepper plant was possibly due to the size of the overall biomass production of the plant. The addition of biochar to soils increased plant K tissue concentration compared to control (Biederman and Harpole, 2013). This increase was due to the high concentrations of available P and exchangeable K found in the biochar respectively (Chan et al., 2007). According to Salim (2016), all biochar treatments enhance the leaf concentrations of N, P and K of wheat plants.

The effect of biochar levels on fruit nutrient content of tomato plant, showed a significant increase in N content in tomato fruit at 8 and 16 tons ha<sup>-1</sup>. The K content was significantly increased at 16 and 30 tons ha<sup>-1</sup>, while the 8, 30, and 40 tons ha<sup>-1</sup> gave a significant increase of P content compared to control (Table 9). The bell pepper content of N, K, and P are shown in Table (9). Biochar addition did not significantly affect N content, but K was significantly increased at 16, 30, 30, and 40 tons ha<sup>-1</sup>. At 8 tons ha<sup>-1</sup>, the P was significantly increased.

The measured soil pH at mid and at the end of the growing season (after five months) is shown in Table (10). There was a significant increase in soil pH with biochar application. Also, the pH at end of season was significantly increased at the high level of biochar. Novak et al. (2009) explained through their study that pH increased from 4.8 to 6.3 with the addition of 2% biochar. Van Zwieten et al. (2010) tested two biochars produced from slow pyrolysis of paper mill waste for two agricultural soils; they found that they differed slightly in their liming values by 33% and 29%, respectively.

The effect of biochar levels on soil EC at mid and at the end of the season are shown in Table (10). There was a significant increase of EC at 40 tons ha<sup>-1</sup> at the mid of season. At the end of the season, the biochar addition was increased the EC. This increase may be due to the high EC of biochar, which produced from olives trees pruning residues. Chan et al. (2008a) found significantly higher EC at higher rates (>50 tons ha<sup>-1</sup>) of green waste biochar in alfisol soil. Similar results were observed also, when poultry litter biochar was tested (Chan et al., 2008b).

Biochar (ton ha <sup>-1</sup> )	Plant yield (kg m <sup>-2</sup> )	Plant yield (kg m <sup>-2</sup> )
	Tomato	Bell pepper
0	8.1 ab	1.5 a
8	9.1 a	1.8 a
16	5.5 b	1.76 a
30	6.7 ab	1.96 a
40	7.6 ab	2.1 a

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Biochar (ton ha <sup>-1</sup> )	N (%)	P (%)	K (%)	N (%)	P (%)	K (%)
		Tomato			Bell peppe	er
0	2.99 c	0.15 b	1.15 ab	4.01 ab	0.21 a	1.12 a
8	3.20 bc	0.19 ab	1.35 a	3.82 b	0.26 a	1.21 a
16	3.78 ab	0.20 ab	1.08 b	4.84 a	0.30 a	1.40 a
30	3.54 abc	0.29 a	1.16 ab	4.01 ab	0.31 a	1.08 a
40	3.92 a	0.21 ab	1.03 b	4.08 ab	0.25 a	1.30 a

Table 8: Effect of biochar levels on leaf nutrients content of tomato and bell pepper.

Table 9: Effect of biochar levels on fruit nutrient content of tomato and bell pepper.

Biochar (ton ha <sup>-1</sup> )	N (%)	P (%)	K (%)	N (%)	P (%)	K (%)
		Tomato			Bell peppe	er
0	1.30 b	0.24 b	3.19 b	2.03 a	0.21 b	1.93 b
8	1.40 a	0.44 a	3.91 b	2.24 a	0.37 a	2.74 ab
16	2.42 a	0.24 b	5.43 a	2.46 a	0.21 b	3.82 a
30	1.47 ab	0.38 a	5.09 a	2.31 a	0.23 b	4.22 a
40	2.03 a	0.38 a	3.24 b	1.79 a	0.23 b	3.18 a

Table 10: Effect of biochar levels on soil pH and EC at the middle and at the end of the experiment.

Biochar (ton ha <sup>-1</sup> )	$pH_{mid}(-)$	pH <sub>end</sub> (-)	EC <sub>mid</sub> (dS m <sup>-1</sup> )	$EC_{end}$ (dS m <sup>-1</sup> )
0	7.69 c	7.68 c	0.57 b	0.57 b
8	7.88 c	7.96 d	0.64 b	0.84 a
16	7.97 b	8.02 bd	0.74 ab	0.96 a
30	8.06 b	8.09 b	0.72 ab	0.98 a
40	8.15 a	8.24 a	0.90 a	1.09 a

## Summary and Recommendation

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- Pot experiment: The addition of biochar resulted in an increasing of shoot fresh weight and shoot dry weight at low levels of biochar then the values were significantly decreased at high levels of biochar. The concentration of the most nutrients in the plant leaves was increased with biochar application. Soil pH and EC were significantly increased with increasing of biochar levels.

- Field experiment: The leaves mineral and fruits content of tomato and bell pepper plants increased with biochar application. The tomato yield increase at the lower biochar level and decreased at higher levels. On the country, bell pepper yield increased with increasing biochar level. Soil pH and EC were increased with the increasing the biochar levels.
- To our knowledge, the research on the application of biochar as a soil amendment has not been investigated in Jordan. Thus, this present study suggest that biochar can be used as a soil amendment; however, still several studies are needed to investigate the effect of biochar feedstock, pyrolysis conditions on different soil, climate and plants.

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# The Effects of Copper Sulphate Pentahydrate on the Enzyme Activities of Rainbow Trout Gills

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#### ABSTRACT

Copper sulphate is too toxic in aquatic environments. this chemical may accumulate in aquatic food chain, especially in fish. The aim of this study was to determine the copper sulphate pentahydrate (CuSO<sub>4</sub>.5H<sub>2</sub>O) toxicity mechanism on superoxide dismutase (SOD), glutathione peroxidase (GPx) and catalase (CAT) and malondialdehyde (MDA) levels in gill tissues of the rainbow trout (*Oncorhynchus mykiss*). After fish were exposed to two different concentrations with control group (0.175 mg/L and 0.350 mg/L) of CuSO<sub>4</sub>.5H<sub>2</sub>O for 28 days, the levels of SOD, CAT and GPx activities of rainbow trout gill tissues decreased but MDA level increased (p<0.05).

Copper sulphate pentahydrate showed biochemical effects in fish gill by inhibiting the enzymes according to the findings of this research. It has also been determined that enzyme activity and MDA level can be used as a biomarker in determining the toxic mechanism of action in fish.

**Keywords:** Rainbow trout, copper sulphate pentahydrate, gill, enzyme.

# Effect of Feeding Astaxanthin on Common Carp (*Cyprinus carpio* L.) Fillet Quality

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## ABSTRACT

Astaxanthin has a vital role in stabilizing fish fillet quality, including proximate composition of carp, TBA values by impacting the oxidative stability of lipids. This study was taken to evaluate roles of astaxanthin in quality of common carp fillets through experiment that ran for 60 days, fish were divided into to 4 treatments T1 (control), T2 (2.5% astaxanthin), T3(5% astaxanthin) and T4 (7.5% astaxanthin). Significant differences (p > 0.05) were observed for proximate composition such as (moisture, protein and fat) except ash that was insignificant. For TBA, water holding capacity, cooking loss and Ph tests were significant differences were seen among treatments. All criteria's in sensory evaluation were above rejection rate and overall were accepted. As expected, feeding common carp with astaxanthin has significantly affected fillets quality and composition.

## ACKNOWLEDGEMENTS

We acknowledge that our research was not funded by any organization.

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# Investigation of the Inhibitory Effect on β-lactamase Activity in Some Seaweeds from Algerian Coast

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## ABSTRACT

The high prevalence of B-Lactmases producing isolates wordwide is distressing because it constitutes a serious health problem. Hence, new and potential β-lactamases inhibitors is an urgent need. It is well known that, over the past decades, seaweeds have been attracting attention in the search for bioactive compounds to develop new drugs and healthy foods.We investigated the inhibitory effect of the methanolic extract of three seaweeds (Corallina Cystoseira *tamariscifolia*) elongata, Ulva lactuca, and on β-lactamase activity. The methanolic extracts of seaweeds were subjected for phytochemical analysis before the inhibition tests. The total polyphenol, flavonoids and vitamin E contents varied between the methanolic extracts of the three seaweeds extract tested. However, the kenitic data, categorizing the inhibition of the seaweed extracts as mixed inhibition type.

## ACKNOWLEDGEMENT

This work was supported by the Algerian Ministry of Higher Education.

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# *In Vitro* Cytotoxicity of Combined Mycotoxins Enniatin-A and Fusaric Acid on HeLa and MCF-7 Cells

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#### ABSTRACT

The contamination of food by natural mycotoxins is a serious problem worldwide for food and agricultural industries, as well as human and animal health. Enniatin-A (EN-A) and Fusaric acid (FA) are species of *Fusarium* mycotoxins that frequently contaminate cereals especially in maize. Fusarium spp. have been described to have immunotoxic, genotoxic and hepatotoxic effects [1]. Additionally, previous studies have shown that individual mycotoxins EN-A and FA have cytotoxic effect on human cervix carcinoma (HeLa) [2, 3] and human umbilical vein endothelial (HUVEC) cell lines [4]. However, in the case of cooccurrence of several mycotoxins in a single food product, additive or synergic toxic effects can be expected [5]. The data on combined toxic effects of mycotoxins are not well-known. Thus, the main goal of this study was to assess the cytotoxic effects of combinations of mycotoxin EN-A and FA on the cell viability of human cervix carcinoma (HeLa) and human breast cancer (MCF-7) cell lines with 3-(4,5-dimethylthiazolyl-2)-2,5 diphenyltetrazolium bromide (MTT) assay. Cells were grown to confluence at 37°C under 5% CO<sub>2</sub> in flasks with Dulbecco's Modified Eagle Serum including 10% fetal bovine serum, 1 (%) penicillin/streptomycin and 2 mM L-glutamine. Cells were treated with nine different combinations of EN-A and FA (between 0.78 EN-A+100 FA and 3.125 EN-A+400 FA µg/mL) for 24 h. A solvent [DMSO, 0.5% (v/v) of the culture medium] and a negative control was also maintained. As a result, the combinations of EN-A+FA treatment significantly decreased the cell viability (%) at all concentrations (except at 0.78 EN-A+200 FA µg/mL on MCF-7 cell) in HeLa and MCF-7 cells compare to control. In addition, the cell viability was determined as 20.02 (%) on HeLa cell and as 32.86 (%) on MCF-7 cell at the highest concentration of combination (3.125 EN-A+400 FA µg/mL). It was observed that the combinations of EN-A+FA had more cytotoxic effect on HeLa cells than MCF-7 cells at all treatment (except at 3.125 EN-A+100 FA µg/mL). This result obtained that combinations of EN-A and FA have strong cytotoxic effect in both cell line. This cytotoxic effect may lead to slow down the cell cycle and cell proliferation thus causing impaired DNA replication.

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## Potentials of Three Essential Oils as Postharvest Insecticides in the Management of the Flour Moth, *Ephestia kuehniella*

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#### ABSTRACT

The use of essential oils, as a bio-insecticide, in the control of insect pests of stored grains has shown specificity and variation in the potentiality of their mode of action. In the present study, three essential oils extracted from three aromatic plants of different families, Armoise (Artemisia herba alba, Asteraceae), Oregano (Origanum vulgare, Lamiaceae) and Rue (Ruta montana, Rutaceae), were evaluated for their repellent and fumigant toxic potential against the flour moth larvae, Ephestia kuehniella (Lepidoptera, Pyralidae), under laboratory conditions. The essential oils extraction was done by hydrodistillation method. The repellent activity of each plant was carried in Petri dishes using a filter paper treated with different oil dilutions (25, 75, 100, 120, 130, 150µl/ml). The fumigant toxicity was determined on three concentrations (50, 130, 150µl/l air). Two plants were shown to be repellent against the E. kuehniella larvae. Origanum oil was the most repellent with 67% of repellency rate followed by Artemisia oil (46%) at 120µl/ml after 2 hours of exposure. The oil of *R. montana* had an attractant activity against the larvae and was the most toxic with 56.67% of larval mortality at the first 24hours. The LC50 recorded were 11.64, 175.40 and 1100 µl/l air for the plant oils R. montana, O. vulgare and A. herba alba, respectively. The knowledge of potential insecticide of various plants enables to select the specific essential oils to the target insect and lead to the best stored product protection and could be integrated in insect pest management program.

**Keywords:** stored grains, *Ephestia kuehniella*, essential oils, repellent activity, fumigant toxicity.

# Effect of Treating Maize Cobs with Urea and Wood Ash on Chemical Composition and Gas Production Substrates

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## ABSTRACT

Effect of treating maize cobs with a combination of urea and wood ash (WA) on chemical composition and *in vitro* gas production substrates were examined. The treatments were: 100U (100% urea +0% wood ash), 75U25WA (75% urea +25% wood ash), 50U50WA (50% urea +50% wood ash), 25U75WA (25 % urea + 75% wood ash) and 0U0WA (Untreated maize cobs). Samples were analyzed for dry matter (DM), organic matter (OM), ash, crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL). They were incubated in vitro for 6, 12, 24, 48 and 72 hrs; gas production substrates were also determined and their DM degradation kinetics described using the equation  $y = a + b (1 - e^{-ct})$ . It was observed that the chemical compositions of treatment 25U75WA were improved significantly compared to the rest of the treatments. Treatment 25U75WA had the highest gas production (73.5ml), truly degraded substrate (TDS) (70.53%), microbial mass protein (MMP) (53.55 mg), efficiency of microbial mass protein (EMMP) (23.68) and partitioning factor (PF) (2.88). Treatment 0U0WA had the least TDS, MMP, EMMP and PF. It was concluded that combinations of 25% urea and 75% WA in the treatment of maize cobs could improve its nutritive value compared to treating it with urea alone. This strategy will reduce cost of treatment and also incorporate minerals present in ash.

## ACKNOWLEDGEMENT

Tertiary Education Trust Fund (TETFUND) Abuja, Nigeria is acknowledged for its sponsorship.

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# Contribution of Wild Boar Ticks (Sus Scrofa Algira) in the Maintenance of the Wild Cycle of Rickettsioses in the Far Northeastern Part of Algeria

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#### ABSTRACT

The study carried out on wild boar Ixodidae (Sus scrofa algira) shows that currently the latter is considered among the animals most parasitized by ticks in Algeria with a load of 29 ticks / animal. (Zeroual et al., 2014). These ticks have been the subject of research on their pathogenic role to verify their contribution to the maintenance of the wild cycle of rickettsioses in the far north-east of Algeria and thus their impact on public health. Of 188 randomly selected ticks that were subject to real-time PCR-based Rickettsia spp. DNA targeting a 25-bp fragment of the Citrate synthetase gene from Rickettsia spp. 76 ticks were carriers of this type of coccobacilli, a positive control R. montanensis is used to ensure the proper functioning of the manipulation in case of negativity. DNA samples from positive tick species showed after sequencing the following results: 55/76 of D. marginatus ticks were R. slovaca positive (GenBank: HM161787.1); 17/92 ticks of Rhipicephalus turanicus were positive to R. massiliae (GenBank: U43793.1); 03/10 of Hyalomma marginatum ticks were positive for R. aeschlimannii (GenBank: HQ335158.1); and in late 1/1010 ticks of Ixodes ricinus were positive for R. monacensis (GenBank: FJ919640.1). These results clearly show the contribution of wild boar ticks (Sus scrofa algira) in maintaining the wild cycle of rickettsioses, which increases the risk of transmission of these pathogens to humans.

**Keywords**: wild boar (Sus scrofa algira), *R. slovaca, R. massiliae, R. aeschlimannii, R. monacensis*, extreme northeastern Algeria.

# Evaluation of the Hematological Parameters, Erythrocyte Fragility and Antioxidant Status of Chinar (*Platanus orientalis*) Leaf Infusion Against Ethanol Toxicity in Rats

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## ABSTRACT

In this study the protective effects of the leaf infusion obtained from the chinar (Platanus orientalis L.) plant were evaluated on hematological parameters and erythrocyte fragility and its antioxidant role against ethanol-induced oxidative stress in rats. For this purpose, after a toxicity test thirty male Wistar rats were divided into 5 groups: Control, Ethanol 20%, Ethanol 20% + Silymarin (10 mg/kg), Ethanol 20% + PO-20 mg/mL infusion, and Ethanol 20% + PO-60 mg/mL infusion. The PO-leaf infusion doses were given ad libitum during 28 days to test the studied parameters. According to results, in the Ethanol group, Red Blood Cells counts, Red cells Distribution, Platelet crit, Platelet and Lymphocyte levels decreased significantly compared to Control, while PO-60 dose-fed group showed significant increase in Haematocrit and Haemoglobin values compared to Ethanol. There were significant changes in erythrocyte fragility of Ethanol and Ethanol-treatment groups at different NaCl concentrations of 0.3, 0.6 and 0.7 (only in Ethanol and Ethanol + PO-20 group) according to Control group. It was observed that PO Leaf infusion reduced the hemolysis caused by ethanol at a concentration of 0.3% NaCl, thus reducing the values to the control values. In addition, PO leaf infusion caused a significant increase in total antioxidant status (TAS) against ethanol toxicity and a significant decrease in total oxidative status (TOS) and oxidative stress index (OSI). As a result, it was concluded that PO leaf infusion may have antihematotoxic effect, reducing erythrocyte fragility and increase antioxidant capacity against ethanol toxicity.

**KEY WORDS:** *Platanus orientalis,* Hematological parameters, Erythrocyte fragility, Antioxidant, Ethanol, Rat.

#### ACKNOWLEDGEMENT

This work was supported by the Van Yuzuncu Yil University Scientific Research Project Commission (Grant No: TAP-2019-8183).

# Partly Regeneration / Repairing Impact of Lyophilized Ethanolic Extract of Achillea arabica Kotschy. in the Islets of Langerhans Against Streptozotocin-induced Diabetic Rats

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#### ABSTRACT

Diabetes mellitus (DM) is a serious metabolic disorder characterized by hyperglycemia resulting from a deficiency in insulin secretion or action. The present study was undertaken to evaluate protective effect of lyophilized ethanolic extract of *Achillea arabica* Kotschy. on pancreatic islet of Langerhans in streptozotocin (STZ)-induced diabetic rats. The acute toxicity of the extract on animals were evaluated giving increasing dosages of 25, 50, 100, 250, 500, 1000 ve 2000 mg/kg by oral gavage. The rats were randomly divided into five groups: Group I (normal control, NC) received citrate buffer at 1 ml/kg given by intraperitoneal (i.p.) way; Group II (DM) were treated with a single dose of freshly prepared STZ in citrate buffer (50 mg/kg body weight (bw)) by i.p. way; Group III (A. arabica, Aa) was received daily a single dose (400 mg/kg bw) of extract of A. arabica; Group IV (DM + Aa): extract of Aa was given to the diabetic animals. Group V (DM + Glibenclamide (Gli)): Gli (2 mg/kg bw) was daily given to the diabetic animals. The extract and Gli were applied to animals during 21 days by oral gavage. Next, histopathology, morphometric analyses and  $\beta$ -cell immunohistochemistry in the Langerhans islets, and blood insulin and C-peptide levels were conducted. STZ treatment caused histopathological changes in the islet and lowered significantly islet diameter / area,  $\beta$ -cell index values and, blood insulin and C-peptide levels. Administration of lyophilized ethanolic extract of A. arabica improved of the islet histology and significantly increased islet diameter / area and  $\beta$ -cell index values and caused significant increases in insulin and C-peptide levels comparing to diabetic rats. Our findings suggest that the A. arabica extract possesses protective ability on the pancreatic islets due to its partly repairing or regeneration impact on  $\beta$ -cells.

#### ACKNOWLEDGEMENT

The present study was partly supported by the Scientific Research Project Coordination Unit of the Van Yuzuncu Yil University, under project number: FAP-2019-8503.

## Molecular Characterization of Thermophilic Bacteria and Investigation of Their Potential to Produce Biotechnologically Important Enzymes

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#### ABSTRACT

Thermophilic organisms whose optimum growth temperatures may range from  $55^{\circ}$ C to  $105^{\circ}$ C are commonly known as extremophiles within the group of eubacteria. These organisms will be naturally found in hot springs, tropical soils, dung heaps, feces and litters.1 Thermophiles, which are used effectively in various industrial applications and can be functional under extreme conditions, would produce various enzymes and enzyme groups. Enzymes obtained from microorganisms which would be resistant to extreme conditions are more preferable than other enzymes since they are used for a longer period of time due to their high catalytic activity, they form less by-products, they are more stable and cheap and they can be produced in high amounts and high purity in the production process. Within the scope of the study, primarily, bacteria were isolated from 11 samples obtained from thermal plants located in different regions of Turkey. 16 S rRNA gene region of the isolated bacteria was cloned and species were determined on molecular level. Afterwards, lipase, amylase and protease enzyme activities of the microorganisms identified were designated by disc diffusion method. As a consequence of analyses performed, it was detected that isolated bacteria would belong to Aeribacillus pallidus (4), Bacillus pumilus (3), Anoxybacillus gonensis (1), Bacillus licheniformis (1), Anoxybacillus kaynarcensis (1) and Bacillus thermoamylovorans species.

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# Molecular Characterization of Lactic Acid Bacteria from Cheese Samples

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#### ABSTRACT

Lactic acid bacteria (LAB) are highly valuable microorganisms producing lactic acid which is one of the main fermentation products of carbohydrate metabolism. LAB are prevalently found in nutrient-rich environments, particularly in dairy products, meat and vegetables. Based on this information, white cheese samples were collected from many different regions, chiefly in Erzurum, and isolation, identification and molecular characterization of lactic acid bacteria were performed. Ultimately, bacterial isolates belonging to *L. brevis, L. casei, L. paracasei, Pediococcus acidilactici, L. paraplantarum, L. buchneri, L. plantarum* and *Enterococcus* faecium were obtained. For this purpose, prelusively, temperature, pH and salt concentration ranges at which bacteria developed at 30-40 °C, pH 4-7 and 2-6 % salt concentration values. Afterwards, the test isolates were examined using 16S rRNA gene analysis method. According to 16S rRNA sequence analysis data, it was concluded that AB1 strain was similar to *L. buchneri* with the rate of 98 % and AB5 strain have similarity with *E. faecium* at the same rate and these two isolates are likely to be new species

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# Quantitative Inventory of Ectoparasites of young Barn Swallows (*Delichon urbica*) from Mouldi Achouri colony of the City of Tebessa, Northeast of Algeria

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## ABSTRACT

During the wintering season (2018-2019), the collection and quantification of ectoparasites was carried out for the first time on the young barn swallows "Delichon urbica" from 18 nests in Mouldi Achouri colony of the city of Tebessa. The results showed that fleas (Ceratophyllus gallinae, Ceratophyllus hirundinis, Callopsylla sp., Cimex hirundinidus, Xenopsylla trispinis, Ornithomya sp) are the most abundant. They represent 73,06% of the overall total of the sample, followed by mites (Ornithomya sp, Ixodes frontalis) with 21,14% and finally, lice (Menacanthus sp) with 5,76%.

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## Role of wild and honey bees (Hymenoptera: Apoidea) in the pollination of *Vicia faba* L. var. *minor* (Fabaceae) in Constantine area (Algeria)

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#### ABSTRACT

Pollinating insects of spring fababean, *Vicia faba* var. minor were determined during the flowering period of 2019 in a private plot located in the locality of Ibn Ziad (North of Constantine). Four orders of insects : Hymenoptera, Lepidoptera, Diptera and Coleoptera were encountered on the flowers. The Hymenoptera (93% of recorded visits) were the most abundant visitors. The honeybee was the dominant forager (58% of visits and 16 individuals/100 flowers), followed by *Eucera numida* Lepeletier (20% of visits and 5 ind /100flrs). Only 42% of honey bee visits are positive (contact with the stigma) while *Eucera numida* visits are all potentially fertilizing. The two bees have the same frequency of visits (5 and 6 flowers/min). The presence of pollinating insects significantly improves the yield of the plant ; the percentage of pods formed (55%) was higher in free pollination (FP) compared with self-pollination (SP) (29%). Similarly, the average number of seeds /pod (3,09 ± 0,71) and the average weight of 10 seeds (10,16 ± 0,94) were higher in FP than in SP (2,00 ± 0 64 ; 6,29 ± 1,11).

## Contribution to the Study of Ovarian Cysts in Cattle

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## ABSTRACT

A clinical study was performed to evaluate the diagnostic methods for ovarian cysts (OC), and to determine the metabolic profiles of animals with OC in the region of Mitidja in the North of Algeria. A total of 504 non-pregnant lactating cows were used in this study. Ultrasonography was performed by EXAGO scanner and was combined with assessment of serum P4. Biochemical serum parameters were assayed by spectrophotometry and insulin and cortisol serum measurement was performed by electrochemiluminescence. The results showed an overall incidence of 11.9% of OC. The incidence of OC was higher among cows in third lactation. Holstein breed was the most affected by OC compared with other breeds (P<0.001). There were no effects of average BCS (Body Condition Scoring) and milk production on the incidence of OC (P>0.05). OC were single in 91% of cases. They were found mainly on the right ovary (66.66%). Seasonality had a significant influence on incidence rate of OC with higher incidence rates during winter and spring (71.66%); while, 28.33% of OC were detected during the summer and autumn (P<0.05). OC were associated with low serum concentrations of glucose, insulin and urea as well as high levels of cortisol. Ultrasound examination and progesterone assays were proposed as the most effective diagnostic combination to diagnose OC. In conclusion, in addition to hormonal imbalances, metabolic disorders are involved in the formation and/or persistence of OC. Therefore, the use of metabolic indicators in understanding and exploration of OC is of great interest.

Keywords: Cow, Ovarian cyst, Metabolic profile, Ultrasonography

## Coccidiostatic Effect of Artemisia herba-alba Asso in Chicken

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## ABSTRACT

This study was conducted as part of the search for new alternatives to anticoccidial drugs. The coccidiostatic effect of an Artemisia species, widespread in Algeria, was studied during an experimental infection with Eimeria tenella in chicken. Three groups of 30 chicks each were formed: Infected Untreated Group (IUG), Infected Treated with Monensin Group (ITMG); Infected Treated with Artemisia herba-alba Asso Group (ITAG). Animals received individually by gavage 105 sporulated oocysts of Eimeria tenella. No mortality was recorded in ITAG animals receiving the studied plant in their diet. Oocysts excretion was significantly (p<0.05) reduced in ITAG animals, treated with Artemisia herba-alba Asso at the 6th and the 8th days Post-Infection. Results show that the studied plant presents a good coccidiostatic effect through the prevention of mortality, and reduction of oocysts excretion during caecal coccidiosis.

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# Occurrence of Aflatoxin M1 in Raw Milk Collected in the East of Algeria

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#### ABSTRACT

In order to assess the mycological contamination of cattle feed, and the search for aflatoxin M1 in milk consumed in Algeria, we realized this work in eastern Algeria zone. Mycological results showed the presence of 247 fungal strains belonging to 9 genera (Aspergillus, Penicillium, Alternaria, Mucor, Rhizopus, Fusarium, Geotricum, Helminthosporium, Cladosporium), with a dominance of Aspergillus, followed by Penicillium. Among the foods analyzed, the concentrate appears to be the most contaminated substrate. The mycotoxicologique study by HPLC, and involving a total of 22 raw milk samples, revealed only one contaminated sample without exceeding the standard described by European regulations (11ng / L).

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## Fruit Flesh Browning Susceptibility of 'Amasya' Apple Cultivar and Its Two Selected Sibs

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#### ABSTRACT

Post-cut internal flesh browning of apples is one of among the most common consumer complaints due to the unattractive appearance and consequential off-flavour. Therefore the browning of apples plays a critical role in fruit breeding programs as well as fruit researches. Non-browning cultivars are also expected to be in demand for fresh fruit and processed products to gain the needed consumer attention. In this study, we aimed to evaluate and 'Amasya' apple cultivar and two superior sibs (named '109' and '120'), of which female parent is 'Amasya', for internal flesh browning susceptibility both visually and objectively. Colour changes of apple fruit after slicing were determined using different colour parameters. Measurements were taken every ten minutes during the first thirty minutes after slicing to achieve the browning curve. The most rapid initial browning of fruit flesh occurred within the first ten minutes after the disruption in both sibs and 'Amasya'. The total colour change during browning showed significant difference among genotypes. It was observed that 'Amasya' undergone significant colour change first, followed by '120' in all the studied colour parameters, whereas '109' did not brown significantly. Hence, it can be concluded that '109' could be useful in the fruit processing industry thanks to the tremendous post-cut anti-browning properties.

Keywords: Enzymatic browning, polyphenol oxidase, breeding, fruit quality

# Eggshell Supplementation in Feed for Laying Hens: Effects on Egg Quality

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## ABSTRACT

Eggshell solidity is a very important and very necessary criteria in the sector of laying eggs for consumption. It allows to limit the number of broken eggs during egg laying or handling and thus reduce economic losses. This criteria is closely associated to the quality of the food, more specifically its calcium and phosphorus content. The main objective of this study was to valorize eggshell supplementation in the feed of laying hens as a source of calcium and to determine its effects on egg quality. After experimentation, the results obtained showed that egg shell supplementation in the diet of laying hen improved the external qualities of the eggs (egg weight [+3g], shell weight [+1.3g], shell proportions, the average thickness of the shell [0.41g], the shape index [p=0.01]).

## ACKNOWLEDGEMENT

This work was supported by Poultry complex of Bali group of El Hachimia (Bouira)

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# **Toxicity and Sublethal Effect of Citrus Peels Essential Oils Formulations to Aphidophagous Coccinellid Predators**

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#### ABSTRACT

Plant essential oils are among the most potential botanicals for alternative bioinsecticides in pest management. Essential oils (EO) exhibit biological action on a wide range of plant pests and they may affect their behaviour but also that of their beneficial ennemis. Coccinellid beetles are among the most important aphid predators. The goal of the present study was to evaluate the toxicity and the sublethal effect of two citrus essential oils against two common aphidophagous coccinellid predators, Coccinella algerica K. and Hippodamia variegata G. Formulated EO solutions extracted from lemon peels Citrus lemon (var. Eureka) and orange peels Citrus sinensis (var. Valencia Late) fruits collected in untreated orchards situated in southeast of Blida region (Algeria), were tested at different concentrations exposure on adults beetles in laboratory conditions. Pure essential oils cause a total lethal effect after an 1 hour exposure to the bioproduct. The orange EO formulated at 25% as well as the lemon EO formulated at 5% were repulsive on the adults of Adonia variegata goeze at the dose of 100%. Lemon zest 1% EO showed a moderate effect at (100%), while lemon EO 3% formulation resulted in a 50% of repellency. The orange peel EO formulations at 5% and 1.14% did not affect the growth of C. algerica third instar larvae, but it caused disturbances in C. algerica and H. variegata fertility. The alternatives for the utililization of essential oils in aphids control management are discussed regarding present results.

#### ACKNOWLEDGEMENT

The authors are grateful to Saïdal group and CRAPC Bousmaïl Algiers for providing facilities.

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# Effects of Humic Acid and Zinc Applications on Yield and Agronomic Features of Oat Genotypes Sown in Winter or Spring

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#### ABSTRACT

The demand for cultivation of oats increases day by day due to its use of human nutrition, industry, green feed, and artificial grazing. Oats are generally sown in spring due to their sensitivity to winter cold. However, oat will have a longer growth cycle in the case of sowing in winter will be maturing earlier so it will have higher yields than those grown in summer. It is known that sufficient organic substance and zinc content in soil have a positive effect on plants regarding cold tolerance and yield. On the other hand, in Central Anatolian soils, zinc deficiency (Çakmak et al., 1999) is one of the leading causes for limited oat production. Under normal conditions, zinc fertilization in the soil promotes plant growth and development (Muneta et al., 2017). It is common to be cold related plant death in seedlings that develop from seeds with low zinc content (Cakmak et al., 1999). Humic substances (fulvic acids and humin) speeds up the growth of root development in the early stages of plant growth, facilitates the germination of the seeds and seedling development in addition to improving the soil structure, cation exchange capacity and microbic activity (Mora et al., 2010).

This study was conducted in Agricultural faculty research area, Eskişehir Osmangazi University in the 2017/2018 growing season in order to determine the effects of zinc and humic acid applications on the winter and spring planting of oats, their winter tolerance, and some yield features. Experiment was established randomized complete block design in split plot arrangements with three replications by using three oat genotypes at two sowing time (October and March). Humic acid applications (60 l/ha) were placed in main parcels, whereas zinc rates (0, 23 and 46 kg/ha) were placed in sub-parcels, and then oat genotypes (Local-1, Local-2, and Albatros) were put in sub-sub-parcels. In this study, humic acid and zinc rates were applied to the soil before sowing. Chlorophyll content was measured at heading stage, while at harvest, plant height, panicle height, panicle weight, number of grain per panicle, weight of grain per panicle, harvest index, grain yield, and protein content were determined.

Results indicated that the effects of humic acid and zinc applications varied based on genotypes and sowing time. Similar values were found in the winter and spring sowing of oat genotypes. According to the results of this study, humic acid and zinc applications may increase the winter tolerance of oat plants, and oat can also be grown in winter under certain climate conditions. These results show that humic acid and zinc applications can be a

benefical model for sensitive plant species to cold stress and contribute to the country's economy.

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# Effects of Sugar (sucrose) on Physical Quality, Fermentation Profile, Nutritive Value of Alfalfa Silage at Different Fermentation Time

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#### ABSTRACT

The present study was carried out to investigate the effects of sugar (sucrose) on physical quality, fermentation profile, nutritive value of alfalfa silage at different fermentation period. After harvesting, fresh alfalfa samples were chopped into small pieces and the sugar were manually applied (3% of fresh weight) to fresh alfalfa in a plastic basin. Five silos from each group were opened for the analysis of physical quality (Smell, color, structure of alfalfa silage), fermentation quality (pH, Flieg point, Ammonia-nitrogen, organic acids), nutritive value (NDF, ADF, DDM, DMI, RFV) on 7, 14, 30, and 60 days of ensiling. Physical quality analysis was assessed by using DLG scoring system (1). Flieg point (2) was calculated by using pH and dry matter values of silage at different days of ensiling. Digestible dry matter (DDM) was determine by using ADF content of silage, dry matter intake (DMI) was measured by using NDF content of silage and Relative feed value (RFV) was calculated (3) by using DDM and DMI. Silage pH value was determined with glass electrode pH meter from filtrate. Fresh silage NH3-N/TN analysis were measured according to the Kjeldahl method (4). Physical quality of silages revealed that good and excellent quality silages were obtained regardless of the days. At d 7, 14, 30 and 60, silage pH was low and Flieg point was high in sugar group compared to control group. Sugartreated alfalfa silage had low NH3-N/TN content compared to control group during the study. Acetic acid and lactic acid was linearly increased and propionic acid and butyric acid were decreased during the weeks. To establish a base-point to better determine where RFV's rate on the quality scale, it can be noted that an ADF of 41% and an NDF of 53% would relate to an RFV of 100 (5). As compared to control group, sugar addition decreased the NDF and ADF content, while DDM, DMI and RFV were linearly increased by the sugar addition. As a result, addition of sugar improved silage quality at different fermentation time

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# The Effect of Calcium on Ion Accumulation to Pepper Plant under Salt Stress

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#### ABSTRACT

As with all plants, climate characteristics have a limiting effect on pepper cultivation. Under natural conditions, plants are often exposed to environmental stressors. Some environmental factors such as air temperature create short-term stress; stresses caused by other effects such as water content in the soil can last for days. Therefore, plants may be exposed to many biotic and abiotic stress factors at the same or different times during their lifetime. Stress in crop production; It can be defined as one or more environmental factors affecting plant, slowing growth and low yield. Factors that cause stress in the plant, disease-forming and pests, such as live origin; salinity, drought, low and high temperatures, radiation and deficiencies or excess nutrients. The study was carried out in order to understand whether the degree of damage caused by salt stress would be reduced by eliminating the calcium (Ca) element deficiency occurring in pepper plants under salt stress and whether it would affect the intake of other nutrients. The study was carried out under controlled conditions in 16/8 hour light / dark photoperiod, 25 0C and 70% humid climate room.

Some ion amounts (Na, K, Ca, Cl) were determined in the study. Increased levels of calcium in the saline environment generally had a positive effect on the amount of macro elements in the plant, reducing and / or eliminating the harmful effect of NaCl. It was determined that the accumulation of Na and Cl ions decreased in root, stem and leaves as Ca dose increased. As a result of the measurements and analyzes, it can be said that increasing doses of Ca to pepper seedlings under salt stress is effective, albeit partially, in reducing the negative effect of salt.

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# Some Reproductive Characteristics of Hair Goat Flocks Under Extensive Conditions in Turkey

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#### ABSTRACT

This study has been carried out to determine vitality of kids and some reproductive characteristics according to kidding and growth of hair goats in rural farm conditions in Turkey. The reproductive performances related to two kidding seasons of the flocks in Kavsit village, Çine, Aydın, in South-West Anatolia were recorded. A total of 840 goats and 772 kids in 6 rural farms were used in this study. The kidding results, fecundity, litter size, infertility rate and twinning rate were found to be 0.95, 1.09, 13.4 % and 9.21%, respectively. The age of the dam and the farm were significant sources of variation for the fecundity and the infertility rate of the animals. The vitality rates (%) of the kids until 7, 30, 60, 90 and weanning/marketing age were found as 94.11, 85.81, 81.82, 80.65 and 79.90 for two years. The age of the dam and the farm were significant sources of variation for the number of weaned or marketed kids per served goat. The farm was only found to be a significant variable for the number of weaned or marketed kids per kidded goat.. The results of this study

have shown significant problems related to fertility and vitality in hair goat flocks under extensive conditions.

Keywords: hair goat, reproductive charachteristics, vitality, rural conditions.

#### ACKNOWLEDGEMENT

This work was supported by BAP (The Scientific Research Projects Coordination Unit).

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# The Current State Of Agricultural Cooperatives in Turkey, İssues And Comparison With The Cooperatives in Agriculturally Developed Countries

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#### ABSTRACT

When the agricultural infrastructure of developed countries is examined; it is clear that manufacturers are organized in order to have an effective role in supplying inputs and to have an active role in the market. Thereby the producers are also industrialists and with this method, rural development can be achieved. The most effective way to determine the policies to be applied in agriculture by affecting political mechanisms as in developed economies is the organization of producers In this context, cooperatives are considered to be the most effective organization model in the development of the agricultural sector. The purposes of this study are; to reveal the current status and problems of agricultural cooperatives in Turkey, to compare with cooperatives in agriculturally developed countries and to offer some advice that will contribute to more effective operation of agricultural cooperatives in Turkey.

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# Evaluation of Growth Parameters of Rainbow Trout (*Oncorhynchus mykiss*) Fed with Boron Additive Feed

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#### ABSTRACT

Additives in aqua feed are nutritive/nonnutritive ingredients that are supplemented in small amounts (alone or in combination) for a specific purpose, such as to improve the growth or quality of fish, to preserve the physical and chemical quality of the diet. The commercial trout feed (Aquamaks, Extruded) is lightly moistened with pure water and milled thoroughly with the aid of a stirrer. The boric acid was mixed to trout feed in six different rates as 500 mg/kg (B1) - 1 g/kg (B2) - 10 g/kg (B3) - 25 g/kg (B4) - 50 g/kg (B5), and 100 g/kg (B6). Then boric acid + feed mixtures compounded equally in the dough machine. After the proportional distribution, the feeds were passed through the pellet machine and brought to the level that the feed size could take by fish. In order to prevent the feed from harming and deteriorating, feed was dried. Growth parameters, gonadosomatic index (GSI) and hepatosomatic (HSI) index values of rainbow trout (Oncorhynchus mykiss) were examined after 90 days feeding period. When the results were evaluated, the highest weight gain was obtained in B6 (100 g / kg) group (p<0.05). The best feed evaluation rate was 1.40  $\pm$  0.02 in the B2 (1g / kg) group, the highest protein evaluation ratio was 1.91  $\pm$  0.04 and the specific growth rate was observed in the B3(10 g/kg) group with  $0.92 \pm 0.14$  (p<0.05). Although alterations in terms of conditional factor, GSI and HSI values were observed among the groups, these differences were not statistically significant (p>0.05).

Keywords: Boric acid, Rainbow Trout, Growth Parameters

#### ACKNOWLEDGEMENT

This work was supported by Atatürk University with Project number: 2016/223.

# Toxic Effects of Lead and Cadmium in Ewes Following Subchronic and Sublethal Exposure

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#### ABSTRACT

Lead (Pb) and Cadmium (Cd) are widely distributed in the environment. These heavy metals are known for their toxic effects in humans and animals, for their cumulative properties and their co-existence. We have designed our experimental conditions in order to simulate a repeated low oral exposure and to highlight the toxic effects, after lead and lead-cadmium exposure for 9 weeks in ewes. The experiment was conducted using "Ouled Djellal" ewes during two periods: before exposure and during exposure. Ten ewes were randomly divided in two groups of five; the lead group received lead nitrate at 2.5 mg.Pb/kg/day and the lead-cadmium group received lead nitrate at 2.5 mg.Pb/kg/day + cadmium chloride at 2 mg Cd/kg/day. Both groups were tested for their blood lead levels and hematological and biochemical parameters before and after receiving the treatment. Clinical signs were taken into account. Before exposure, blood lead levels were below the detection limit of 4  $\mu$ g/l. During the exposure, the levels varied from 135±57 $\mu$ g/l to  $356\pm147\mu g/l$  for the lead group and from  $192\pm75\mu g/l$  to  $445\pm294\mu g/l$  for the co-exposed group. Mean blood lead levels of co-exposed group were more elevated than the ones of the lead group. The transaminases (ALT, AST) and total proteins are high for the Pb-Cd group during the two last weeks of exposure. The ratio albumin/globulin is low. The rates of hematocrit and hemoglobin decreased for the Pb-Cd group to reach a value of 24% and 7.9±0.6mg/100ml, respectively. The co-administration of Pb and Cd resulted a low increase of serum creatinine and a significant reduction in zinc and copper plasma contents. Toxicokinetic analysis showed a greater systemic exposure. Concentrations of lead and cadmium were determined in organs. Histopathologic lesions occurred in liver and kidney. ANOVA was used for statistical analysis.

Keywords: Cadmium - Lead - Ewe - Subchronic oral toxicity - Co-exposure

#### ACKNOWLEDGMENT

As part of National Research Program (PNR), this study was supported by The ANDRU (Agence nationale pour le développement de la recherche universitaire) under the auspices of DGRS-DT (Direction générale de la recherche scientifique et du développement technologique -Alger).

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#### The Nutritional Interest of Atriplex halimus in Arid Areas of Algeria

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#### ABSTRACT

In arid area of Algeria, climate variation leads to drought of soil that becomes a constraint for the plant biodiversity. The new conditions, drought and soil salinity, have modified the stability of the ecosystem and some species are disappeared. The utilization of halophytic plants in saline soils for pasture and fodder production is considered the only economic solution presently available. These groups of plants not only tolerate high level of salinity but reveal optimal growth in saline conditions. The area of El Haouche in South-East of Algeria is considered as an extremely arid zone. Halophytic plants, succulent and perennial species are the mainly vegetation grazed specially by small ruminants and dromedary in this region. The physiognomical map of the study area vegetation reveals the predominance of Atriplex halimus. It is a perennial shrub tolerates well harsh conditions: drought, salinity, water stress and cold. The bioclimatic data shows that the study area is characterized by one long period of estival dryness varying from 6 to 7 months. The pedological approach shows a silt-clay texture, characterized by high salinity; soil pH varied from 7.5 to 8.2. Chemical composition in stems and leaves of Atriplex halimus reveals that leaves are particularly rich in ash, fat and nitrogen, whereas the stems were high in the organic matter and the fiber. There is a lack of information about the mineral profile of this halophyte in the arid regions of Southeastern of Algeria and its relation to the soil characteristics on which this forage plant grows. The knowledge of the level of minerals concentration present in this shrub may suggest minerals supplementation strategies to get better growth and reproductive efficiency of the ruminants The aim of this study was to evaluate and compare during one year, seasonally contents of the macro (Ca, P, Mg, Na and K) and microminerals (Cu, Zn, Mn and Fe) in soil and A. halimus that grow in El Haouch area, as indicators of minerals contribution, deficiencies or excess each season for enhanced nutrition of grazing small ruminants in this area.

Keywords: Atriplex halimus, macro-mineral, trace-mineral, arid zone, drought, salinity

# The Effect of Different Commercial Fertilizer Applications on the Pomological Properties and the Amount of Vitamin C, Water Soluble Dry Matter of Pepper

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#### ABSTRACT

Fertilizer is one of the most important inputs in agricultural production. If it is not applied sufficiently, it causes significant losses in yield and quality, but if it is applied more, it causes pollution of bottom and surface waters, especially with washing of nitrogen and phosphorus fertilizer, nitrogen oxide (NO, N<sub>2</sub>O, NO<sub>2</sub>) emission and air pollution. In order to obtain the expected benefit from the applied fertilizer, it is necessary to know the data such as the time, method, plant type, soil character, climate and amount of plant nutrients in the soil. In the Study Demre pepper variety was used. The study was carried out in the Physiology Laboratory, Faculty of Agriculture, Van Yuzuncu Yıl University, in a climate room where normal atmosphere was provided. The main purpose of the normal atmosphere in the experiment was to ensure that salt stress effects occur under normal conditions. . The study was carried out under controlled conditions in a light / dark photoperiod of 16/8 hours. climate room of 25°C and 70% humidity. The study aimed to investigate the on some kinds of-quality criteria of some commercial fertilizer having a big market share in the world and Turkey. To this end, nine different commercial fertilizer applications were made to the pepper plants. Base fertilizers were applied to the groups to be applied to the soil before planting seedlings. Other fertilizer applications were applied in 10-15 days intervals according to the instructions of the company. In addition, the group watered with distilled water only (control) and second groups with only Hoagland nutrient solution of irrigation. fertilizer application constituted control of these commercial applications. The fertilization plan was made in accordance with the size of the company said. In this study; fruit stalk length, fruit diameter, fruit weight, fruit length, and vitamin C contents, the amount of Water Soluble Dry Matter were examined. When the data obtained were evaluated in terms of fruit feature; It was remarkable that Bestline fertilizer had the highest values in terms of all the developmental parameters examined except for the plant nodes distance. The highest values in terms of fruit diameter and weight were measured from 13-24-12 + 10 (SO3) + Fe + Zn application and the lowest values were measured from 10-30-10 application.Potassium and phosphorus-weighted fertilizers (TSP, Potasmag, Bestline, 10-30-10) were determined to cause a decrease in vitamin C. According to the control applications, 13-24-12 + 10 (SO3) + Fe + Zn, Potasmag, 20-20-20, 10-5-40 fertilizers were also found to have significant decreases in Water Soluble Dry Matter amounts.

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# Determination of Relationship Udder Morphological Traits and Udder Type in Morkaraman and Awassi

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#### ABSTRACT

In this study, it was aimed that determination of relationship udder morphological traits and udder type in Morkaraman and Awassi and the correlation between the results. Cylindrical udder, udder teats are upward and lateral udder type was determined to be 86% and 84% for Morkaraman and Ivesi respectively. In Morkaraman, there was a significant negative correlation between udder type and distance between teats (p < 0.05; r = -0.473) and teat length (p < 0.05; r = -0.440). Also there was a positive correlation between udder type and teat diameter (p < 0.05; r = 0.452). On the other hand, negative correlation was found between udder type and udder length (p < 0.05; r = -0.626) and rump at height (p < 0.05; r = -0.523). It is recommended to carry out new studies on the suitability of machine milking with udder measurements.

# Evaluation of Genotoxic and Antigenotoxic Effects of Pullulan in *in vitro* Human Lymphocytes Using MN and MN-FISH Assays

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#### ABSTRACT

Pullulan produced in aerobic medium from Aureobasidium pullulans and it is a watersoluble, viscous polysaccharide, an alpha-glucan, consisting of glucose units (1). Pullulan is frequently used as an additive in the pharmaceutical and food industries. On the other hand, it can also be used for controlled and targeted drug release (2, 3). The purpose of this study performed to determine genotoxic and antigenotoxic effects of pullulan against Mitomycin C (MMC) using in vitro Micronucleus (MN) and Micronucleus-Fluorescent in situ hybridisation (MN-FISH) assays. For this purpose, these assays were performed on human peripheral lymphocytes from three donors, two women and a man. Five different (250, 500, 1000, 1500 and 2000 µg/mL) concentrations of pullulan were used alone and in combination with 0.20 µg/mL MMC. Administration of these concentrations with MMC was performed in 2 different ways: 1) pre-treatment (antioxidant addition 1 hour before MMC treatment), 2) simultaneous treatment (antioxidant addition at the same time as MMC treatment). As a results, It was determined that pullulan did not significantly affect MN frequency and nuclear division index in human lymphocytes. In contrast, it was found that Pullulan decreased the MN frequency statistically in all studied concentrations (except pretreatment, 1500 µg/mL concentration) in simultaneous and pretreatment, compared to the positive control. In MN-FISH assay, no significant change was observed in the frequency of the centromere-positive (C+) or centromere-negative (C-) MNs compared to the positive control. Besides, pullulan (1000 and 2000 µg/mL) significantly decreased the C + MN frequency compared to the control. The obtained results showed that exopolysaccharide Pullulan is not genotoxic in human lymphocytes, but may have antigenotoxic effect.

#### ACKNOWLEDGEMENT

This study was supported by Gazi University Research Fund under the project number 05/2017-14.

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### Contamination of Zn,Pb,Cd and Hg in Roasted Coffee Powder

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#### ABSTRACT

One of the most popular beverages which is daily consumed by millions of people and the second largest Commertial and industrial commodity after petroleum is Coffee [1, 2]. Today coffee that has been consumed for over 1,000 years and is the one of the most favorite drink in the world (more than 400 billion cups yearly)[3]. Wide activities of mankind in the word has been caused a vast dispersion of heavy metals in the environments and as a result human food contamination [4, 5]. As some of the heavy metals are very toxic for human health, the investigation of metal content in food is considerable [6, 7, and 8]. As mentioned before world trade has been influenced greatly by coffee culture [1, 2]. The sensory characteristics and other social and economic factors makes coffee as a popular drink. Worldwide coffee consumption from 2012/13 to 2015/16 (in million 60kg bags) has been reported respectively 146.98, 149.04, 151.82, 155.71[9, 10]. According to the high consumption of coffee, the levels of Zn, Pb, Cd, and Hg in coffee are studied.

In this study, some of the most consumped brands of coffee with different batch numbers, were selected randomly from imported commodity in market of Iran. Heavy metals (Zn, Pb, Cd, and Hg) in the samples were determined by ICP-OES. Mineralization of coffee infusions: an infusion was filtered through a Whatman paper. 10 mL of the filtrate was mineralized in tubes with 1 mL HNO3 and 0.1 mL HClO4 at 140°C for 5 h. Concentrations of the heavy metals Zn, Pb, Cd, and Hg were determined by ICP-OES. Each metal concentration was estimated 3 times from each sample of coffee. The changes were calculated by one way Anova and for analysis of the role of multiple factors univariate analysis was used by SPSS 21. Probability values of <0.05 were considered significant.

The metal contents in the coffee infusions respectively were as follow: In all samples the amount of Pb and Hg respectively were reported <0/05 mg/kg and <0/1 mg/kg. the amount of Zn in the most samples were similar between  $8.24\pm0.5798$  to  $11.65\pm0.1483$  mg/kg. Analyses shows that Zn in 2 samples of coffee were not detected and in the one sample the content of Zn was about  $19.13\pm0.1$  mg/kg. The results of the analysis indicated that the Cd contents were less than 0/05 mg/kg in all samples just like lead. According to variance analyses of data, heavy metal concentrations in roasted coffee samples were significantly

affected by company and storage time. The data obtained in the present study could be very useful for consumers, suppliers and all researchers who would like to pursue this subject.

Keywords: Food Safety, Coffee, Heavy Metals, ICP-OES

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# The Antioxidant Functions of Achillea millefolium Found in Iran

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#### ABSTRACT

Achillea millefolium is under the group of Asteraceae family. Approximately, 19 species have been found in Iran where can be mostly exist in west, northwest, north, northeast and the center [1]. This study was carried out to investigate the antioxidant impacts of A. millefolium which is endemic plant in Iran. Plants collected from medical plant garden of Islamic Azad University, Shoushtar branch. Clevenger device was used to extract the essential oil and then Gas Chromatography was utilized to identify the components [2]. Antioxidant properties were determined in 2 different test systems with different mechanisms of action, including antioxidant activity by scavenging abilities on DPPH (2,2diphenyl-1- picrylhydrazyl radical) and ABTS (2,2'-azinobis-3-ethylbenzotiazoline-6sulfonic acid radical), reducing power test,  $\beta$ -carotene bleaching and oxidative stability by the Rancimat method. Content and profile of potential antioxidant in these vegetables are also examined. The antioxidant impacts of A. millefolium can be contributed to some components like  $\alpha$ -pinen,  $\beta$ -pinen [1]. Based on anecdotal evidence, some diseases were rarely found among the ancestors like cardiovascular ones because of using herbal medicines. The main goal of current study was to identify the constituents of antioxidant activity from endemic plant and also to determine the chemical composition Achillea millefolium which can be used as drug after toxicity studies and also taking into consideration of high antioxidant effects of A. millefolium, it is recommended that it be used in daily foodstuff to prevent some modern life style diseases.

#### ACKNOWLEDGMENTS

We greatly appreciate Dr. Parisa Ziarati for helping us to conduct the research.

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# Determination of *Origanum acutidens* (Hand.-Mazz.) Ietswaart in Erzincan Province

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#### ABSTRACT

This study was carried out in order to determine the existence of *Origanum acutidens* (Hand.-Mazz.) Ietswaart (endemically grown in turkey) and to reveal the morphological parameters and volatile oil ratio of *Origanum acutidens* populations in Erzincan province. For this purpose, field trips were organized to all districts within the borders of Erzincan province between July 15 and August 15 and nine genotypes with different morphology were determined from three districts with the presence of *Origanum acutidens*. Fresh herba yield, dry herba yield, dry leaf yield, leaf stem ratio, side branch number, canopy diameter, plant height, chlorophyll content and volatile oil ratios were determined in these genotypes. There were found significant differences (p<0.01) in all parameters except essential oil ratio and morphological variation was determined as high among *Origanum acutidens* populations in this province.

**Keywords:** Essantial oil, medicinal plants, endemic plants, morphological characterization

# Ecological Study on the Stéppic Spontanéous Plants in West Part of the Région of Biskra -Algéria

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#### ABSTRACT

We realised a floring sampling on field in order to studying spontaneous plants écosystèm , in tow stations (Doucen and Foughala), during tow periods (2016-2018), and within two sampling times (Décember and January) , in the west part of the region of Biskra.

The first campaign in Doucen show that the occurrence frequency of species : *Cleome Arabica* (74,87%), belongs has the category : (Constant species), with a presence of rare species, such as : *Ferula vesceritensis* (0,48%). The station of Fougala present the species : *Anabasis articulata* (30,76%), as very dominant species in this station, in the category of: (secondary species), in the other side the second period shows a dominance of the secondary species: *Cynodon dactylon* (44,8%).

The measure of the biodiversity during the two campaigns, has reveals a stable mat of spontaneous plants, continuous and diversify jn endemic genera and species (persistants), the most Important are :

The station of Doucen shows in period 1 that : *Cleome arabica* is very dominant in number : 850 plants/100 m<sup>2</sup>, with an abundance of *Anabasis articulata* (100 plants/100 m<sup>2</sup>). campaign The period 2 shows a certain dominance and adaptability of the species and genera : *Cynodon dactylon* (Fo=44,8%) and *Thymelaea microphylla* (Fo=21%).

That indicates that the tow selected stations are belonging in a same healthy steppic ecosystem in the West area of the region of Biskra.

Keywords: Spontaneous plants, stéppic ecosystem, *Cleome Arabica*, *Anabasis articulata*, Biskra.

#### AKNOWLEDGMENT

In this study, we have intersted in all population and species of steppics plants in arid region in the south of Algeria (Biskra), and their écology.

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# Antimicrobial Activity of Tomato Seed Oil Against Lactobacillus casei

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#### ABSTRACT

Probiotic bacteria such as *Lactobacillus casei* are microorganisms that improves intestinal microflora balance and strengthens host defense systems. Tomato seeds are an important waste by-product which contains about 2% vegetable oil and and is obtained from the processing of tomatoes into products. The aim of this study was to determine the in-vitro antimicrobial activity of Tomato seed oil against a probiotic *Lactobacillus (Lactobacillus casei)*. Antibacterial as a minimum inhibitory concentration of the growth and minimum bactericidal dilution method using bacteria, Against the *Lactobacillus casei* was done. The results showed this oil had antimicrobial effect on the *Lactobacillus casei*. Inhibitory zone diameter for *Lactobacillus casei* (in a tomato seed oil dilution of 1.2) was 2.910 cm. The amount of minimum bactericidal concentration (MBC) and minimum inhibitory concentration (MIC) for *Lactobacillus casei* was 0.28+0.5 mg/ml. Tomato seed oil has antimicrobial effect due to its phenolic compounds and it should be taken into consideration in the design and formulation of probiotic products.

#### ACKNOWLEDGEMENT

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# Maize Stover Silage adding Molasses with Different Time Incubation on Physical Quality, pH and Nutritive Value

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#### ABSTRACT

The objectives was to determined the effect of different time incubation of maize stover silage adding molasses treatment on physical quality, pH and nutritive value. The research methode was an experiment by using Factorial Randomized Complete Design (RCD) was used with 2 factors (maize stover = A and maize stover + 10% molasses = B), 3 incubation (0, 7, 14 days) and 4 replication. The treatments consist of  $(A_0 = \text{maize stover without})$ incubation,  $A_1$  = maize stover incubated for 7 days,  $A_2$  = maize stover incubated for 14 days) and ( $B_0$  = maize stover + 10% molasses without incubation,  $B_1$  = maize stover + 10 % molasses incubated for 7 days, and  $B_2$  = maize stover + 10 % molasses incubated for 14 days). The variable was measured physical quality, pH and nutritive value. The results showed that incubation time of A and B gave significant effect on physical quality and pH silage, maize stover silage without and with 10 % molasses incubated 14 days gave the best result of physical quality and pH (A = 4.00 and B = 3.89) silage. The result showed that incubation time of A and B gave highly significant effect (P<0.01) on Dry Matter (DM) and Ether Extract (EE), however it did not give significant effect (P>0.05) on Organic Matter (OM). It can be concluded that maize stover silage with 10% molasses incubated for 14 days could increased physical quality, decreased pH and maintain nutritive value.

Keywords: Maize stover, molasses, physical quality, pH and nutritive value.

# Potentials of Beef Cattle Development in Wet Peatland in Central Kalimantan

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#### ABSTRACT

This research aims to study the potential of beef cattle development on wet peatlands in Central Kalimantan, Indonesia. This research can be used as a guide to develop of cattle farming in peatlands area. This research was carried out in Pulang Pisau district with the consideration that the district has the most wet peatlands in Central Kalimantan, has the potential for the development of cattle farming, namely: having a large area for cattle cultivation and for growing animal feed.

#### ACKNOWLEDGEMENT

This work was supported by LIVESTOCK SERVICE of Central Kalimantan.

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# Inhibition Effect of *Matricaria recutita* Dried Flowers Aqueous Extract on Serine Betalactamase of *Pseudomonas aeruginosa*

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#### ABSTRACT

β-Lactamases are bacterial enzymes that confer resistance to β-lactam antibiotics, by hydrolyzing the amide bond of the β-lactam ring and thus bear significant clinical relevance. In efforts to find new bioactive beta-lactamase inhibitor, we report here the inhibitory effect of the aqueous extract of *Matricaria recutita* dried flowers on Serine beta-Lactamases (SBLs) of *P.aeruginosa*. An enzyme assay was performed by determining the IC<sub>50</sub> in comparaison with clavulanic acid, sulbactam and tazobactam. Effect of the three positive control and *M.recutita* extract produced from dried flowers, on serine betalactamases extract activity was studied *in vitro* by using nitrocefin as a substrate at 25°. Total phenolic contents of aqueous extracts of *M.recutita* dried flowers mesured by Folin-Ciocalteu, were reported  $87\pm1.203$  (µg GAE/mg E). The IC<sub>50</sub> values, determined by linear computerized regression analysis after logit/log transformation, of aqueous extract of *M.recutita* is 79.81±0.86 µM. In comparison with original inhibitors, clavulanic acid, sulbactam and tazobactam, results reflect a weak inhibitory effect, which caused by sterically hindered. However, aqueous extract of *M.recutita* tested is bioactive and exert its inhibitory effect on SBLs in a dose-dependent manner.

**Keywords:**  $\beta$ -Lactamases, *Matricaria recutita*, clavulanic acid, sulbactam and tazobactam inhibition.

#### ACKNOWLEDGEMENT

This work was supported by the Algerian Ministry of Higher Education.

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# A New Colorimetric Method From Qualitative to Quantitative

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#### ABSTRACT

In this study, the conversion of a qualitative method based on color reactions of carbohydrates to quantitative method has been shown. This qualitative method, called the Seliwanoff reaction, has been used for many years in the qualitative determination of monosaccharides, especially fructose. In the qualitatif original Seliwanoff test, the formation of red color is based on the reaction of resorcinol with HMF which is formed by converting fructose in acidic medium (Roe, 1934; Sánchez-Viesca & Gómez, 2018). If fructose is converted to HMF and then reacts with resorcinol to give a red color, can this test be used directly for colorimetric detection of hidroxymethylfurfural (HMF)?. This was our research question. In order to investigate this question, the following studies were performed: the effect of reaction parameters was examined; optimum values of them were determined; a new spectrophotometric method was developed and validated (ICH, 2005). With proposed method, quantitative determination of HMF in foods can be carried out colorimetrically.

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# Effect of Punica granatum L. Peel Extract on Phytopathogenic Bacteria

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#### ABSTRACT

The direct yield losses of crop caused by pathogens, animals, and weeds are altogether responsible for losses ranging between 20 and 40% of global agricultural yield. There have been many researches focused on minimize these product losses and improve product yield. Especially, the synthetic agents and antibiotics have been used for a long time to against to plant pathogenic bacteria. But, because of undesirable effects on the environment in the long run, the scientists have been researching alternative natural compounds. For this purpose, in this study, the pomegranate peels extract was prepared and tested in different concentrations (600-100  $\mu$ g/ml) with 96% ethanol. The effect of prepared extract was evaluated against plant pathogenic bacteria (*Agrobacterium tumefaciens, Pectobacterium atroseptica*). According to the results of this study, the antibacterial effect of pomegranate peel extract was observed for *Agrobacterium tumefaciens*, *P. mediterranea* and *P. carotovorum* subsp. *carotovorum*. Therefore, it is possible to conclude that pomegranate peel extract is highly effective against some phytopathogenic bacteria.

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# Bioremediation and Electricity Generation through Anaerobic Biodegradation of Vinegar Production Plant Waste with shewanella Isolated from Coastal Mud-slims of Qeshm Island in a Microbial Fuel Cell with Optimized Nafion

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#### ABSTRACT

Nowadays, bioremediation of wastewater from factories in the food industry has attracted a lot of attention and developed countries are investing heavily on the studies related to this problem. Using microbial fuel cell for wastewater treatment is regarded as a new fundamental technology. In this regard, the use of new sources for energy without producing carbon dioxide is considered as one of the major concerns among industrialized countries. The Beach Mud of Qeshm Island contains many microorganisms which can be used in these processes, and have turned into an ecological problem. The present study aimed to use this biomass for a creative exploitation of isolating bacterial nanowires. In addition, bio augmentation technique implemented for producing vinegar from Shewaenlla with a sludge isolated was regarded as another innovation used to design this research. To this aim, double-volume microbial fuel cell made of plexiglass was selected, along with a volume of 500 ml for each compartment. Electrodes were made of silver chloride. The distance among cathode and anode was 4.4 cm. Nafion was optimized with polypyrrole for better efficiency. The nylon used by polymerization conductor of polypyrrole was coated with polymerization method using FeCl<sub>3</sub> as an oxidizing agent and paratholene sulfonate (PTSNa) on the surface of sulfur particles. Iron chloride oxidized polypyrrole monomers to cationic radicals, and then they are formed together with these polyoxyethylene polymer cationic radicals. In the anode chamber, anaerobic effluent of vinegar manufacturing plant, 50% effluent and sludge, and a mixture of effluent and Shewaenlla isolated from sludge were used as inoculum, along with glucose. Regarding the cathodic chamber, a constant potassium phosphate buffer solution was used to maintain the pH of the catholyte. In order to isolate single black colonies of Shewanella, Kligler agar culture medium was used by reducing sulfate and producing H<sub>2</sub>S production. This test indicated that the studied sludge contains Shewanella and provides the required bacteria. Shewaenlla was added to the anaerobic effluent with suitable amount and was kept at  $22^{0C}$ during the experiments. Based on the results, the studied sludge contained Shewanella and provided the required bacteria. Further, polarity technique was used to evaluate the electrical performance. Thus, by comparing the results, the bioavailability of effluent with Shewanella significantly increased the efficiency of the inoculum. In this reactor, the maximum generated voltage was generated on the first day of its commissioning, which gradually decreased from the voltage value. The highest flow rate was  $\sim 351 + 9 \text{mW/m}^2$ produced by the cathodic base. Finally, brightening the waste color during the ten days of the experiment could clarify the effluent biomass.

# Morphometric and Meristic Characters of Five Cyprinid Species from Tercan Dam Lake, District Erzincan, Turkey

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#### ABSTRACT

Morphometric and meristic characters of a fish species has major role to ensure whether there is any variety between same species of different geographic region. This study was carried out to determine morphometric and meristic characters of five cyprinid species (Cyprinus carpio Linnaeus, 1758, Luciobarbus capito Güldenstädt, 1773, Capoeta trutta Heckel, 1843, Squalius cephalus Linnaeus, 1758 and Capoeta umbla Heckel, 1843) from Tercan Dam Lake on May 2017. A total of 51 specimens of the five cyprinid species of family Cyprinidae were collected using gill net by fishermen. The morphometric measurements (thirty four) and meristic count (eight) were recorded for each species. The descriptive statistical parameters and correlation coefficient (r) were analysis with independent variable (total length) and dependent variable (other morphometric and meristic characters). The results of the present study revealed that most relationships between these selected morphometric and meristic characters and the total length (TL) were found to be moderate correlation (r>0.50) and statistically not significant correlation (t-test; p>0.05). The present study gives information to fishery biologists about morphometric and meristic characters of fish species from Tercan Dam Lake. This will help to plan further conservation strategy for this fish.

# The Investigation of Biological Control Oppurtunities Against Gray Mold Causing *Botrytis cinerea* Pers:Fr in Eggplant

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#### ABSTRACT

The factor of gray mold, within so many economically-important vegetables, cause some important losses of productivity. This factor is especially encountered in productivity areas, which have insufficient ventilaton, in foggy and dewy places at flowering times, and in depots. That gray mold factor is able to survive for a long time in soil and large hostplant range of the factor make difficult to control this factor. Inadequacy of cultural precautions and the negative impacts of chemicals, which are used for struggling verbal diseases, on human health and nature, especially the fact that pathogens create endurance against chemicals; in spite of these, the situation that there is no effective and adequate method of struggling with disease of gray mold, turn the studies of alternative methods that also involves biological strrugle into prior subjects. In this study, it is used 12 bio-agent bacteria strains (2x Bacillus megaterium, 2x Bacillus pumilis, 2x Bacillus subtilis, 2x Pantoea agglomerans, 1x Pseudomonas fluorescens, 1x Paenibacillus polymxa, 1x Bacilius thuringiensis, 1x Bacilius cereus). These strains are defined in some previous studies according to oil acid esters, which are isolated by the under or upper parts of the soil's wild or cultivated plants. This study is performed under an *in vitro* and conrolled setting, aiming to determine the impact of these bio-agents on B. cinerea, which cause the losses of productivity of eggplant seedlings. Five bacteria strains, which give the best results under in vitro setting, have been chosen for stem practices and; effectiveness of these bacterias have been measured under controlled setting.

In this study, it is determined that one strain – which pertain to *Pseudomonas chlororaphis* supsp. *aurofaciens*, and one *Bacillus amyloliquefaciens* strain are also effective *in vitro* setting to struggle with *B.cinerea*. As a result, it is determined that the most two effective bacteria strains can be used as a biocontrol agent for controlling the *B.cinerea* in eggplant farming.

Keywords: Bacteria, Biological control, Botrytis cinerea, Eggplant

# An Investigation of Biological Control Possibilities against *Fusarium* proliferatum (Matsush.) Nirenberg ex Gerlach & Nirenberg, (1976) on Conic Red Pepper

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#### ABSTRACT

The chemicals used in the fight against disease factors are a problem due to phytotoxicity, residues, environmental pollution and harmful effects on human health. In recent years, the use of environmentally friendly preparations has increased due to the development of environmental and human health awareness and the search for alternatives to plant protection products that cause residual problems in plant products. In this study, it was aimed to determine the bacterial strains and fungus isolates which could be the biological control agent against Fusarium proliferatum which is a root rot (collapsing) disease in pepper from Solanacae (Eggplant) family which is an important plant in our country and in the world. In the survey studies, 4 F. prolifetarum isolates were obtained from diseased pepper fruits. The most virulent isolate was determined by applying Koch postulate method to these isolates and this isolate was used in the other stages of the study. As a bioagent, in previous studies; 52 bacteria strains isolated from the root rhizosphere and aboveground parts of various wild and cultivated plants from different geographical areas in our country were identified according to the Microbial Identification System (MIS). A total of 2 fungi isolates of Tricoderma harzianum were used. The study was carried out in two different stages, *in vitro* and *in vivo*. Of the 52 bacterial strains used *in vitro* in dual culture tests, the 5 most effective strains were tested *in vivo* and 3 yielded successful results. 2 fungal isolates were found to be effective in both in vitro and in vivo conditions. The in vivo stage of the study was carried out using 5 bacterial strains and 2 fungal isolates which were found to be effective. In vivo experiments were performed with 3 replications and 1 plant per pot. At the end of the study, it was observed that 3 bacteria strains and 2 fungi isolates are effective in the treatment of F. proliferatum under in vivo conditions. As a result, it was determined that Bacillus subtilis, Pseudomonas chlororaphis, Bacillus sp. and T. harzianum can be used as biological control agent in the control of F. proliferatum which causing diseases on peppers

**Keywords:** bioagent bacteria, biological control, bioagent fungi, *fusarium proliferatum*, kapya pepper, root rot

# Dissipation Dynamics of Imidacloprid Insecticide in Pepper Fruits Grown in a Greenhouse

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#### ABSTRACT

Peppers are grown extensively in greenhouses, however, humidity and temperature prevailing in these conditions encourage the development of harmful organisms, such as aphids. For the control of leaf aphids, a neonicotinoid insecticide imidacloprid is used. Since 2018., all outdoor uses of the imidacloprid are banned and only the use in permanent greenhouses remains possible [1]. In this study, behavior and dissipation dynamics of imidacloprid in pepper fruits were evaluated. Imidacloprid (700 a.i. g/kg) was applied at the manufacturer's recommended rate (0.1 kg/ha) for the control of Aphididae in pepper, due to the pest biology and phenophase of the crop. The experiment was carried out during 2019 in Vojvodina Province (Serbia) and designed according to EPPO methods. During the experiment, average temperatures ranged from 25.9-30.8 °C, while the humidity was 31-45%. Samples were collected randomly, from various places of the experimental plots, on a daily basis during seven days. For the extraction of imidacloprid from pepper fruits QuEChERS based method was used, while the analysis of imidacloprid residues was done with the HPLC-DAD system (Agilent Technologies 1100 Series) and Zorbax Eclipse XDB-C18 column (50 mm  $\times$  4.6 mm, 1.8  $\mu$ m). Method validation was performed through linearity (0.998), recovery (91-101.2%), precision (RSD=0.29%), matrix effect (99%), limits of detection (0.01 mg/kg) and quantification (0.05 mg/kg). Obtained results completely fulfilled the European Union guidelines SANTE/11813/2017 criteria [2]. After application, the highest concentration of imidacloprid was found immediately after the drving deposit (2.32 mg/kg). The content of imidacloprid residues in pepper dissipated moderately and four days after the application was below EU MRL (1.0 mg/kg). Based on these results, half-life  $(DT_{50})$  of imidacloprid in pepper fruits is 3.65 days and the appropriate pre-harvest interval is 7 days.

#### ACKNOWLEDGEMENT

This work was supported by Ministry of Agriculture, Forestry and Water management of the Republic of Serbia, No. 321-01-00881/2018-11.

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# The Effects of Different Salinity Levels on Germination and Seedling Development of Ulkem Forage Cowpea Variety (Vigna Unguiculata L. Walp)

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#### ABSTRACT

One of the most important factors affecting productivity in agricultural production is stress factors. Among these factors, salt stress is very limited in arid and semi-arid areas. Cowpea, a legume plant, is important because it is used as an animal feed in addition to human nutrition. In this study, it was tried to determine the salinity tolerance of Ülkem feed cowpea cultivar used as feeder. In the study, different doses of NaCl doses were applied and the experiment was established with 10 replications according to the randomized plot design. As a result, germination rate (%), average germination time, radicle and plumule length (cm), radicle and plumule fresh weight, radicle and plumule dry weight (gr) factors were evaluated. Specifically, feed cowpea was found to be resistant to salt doses below 90 Mm

Keywords: Stress physiology, NaCl, Forage crop

# Proceedings of the 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences (ICOFAAS 2019) Antalya, Turkey, 8-11 November 2019 Effects of n-butanol on Cardiac Activity and Heart Contraction Power in Zebrafish Larvae (Danio Rerio)

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#### ABSTRACT

The n-butanol, which is widely used in the food industry, is the most toxic of the butanol group alcohols. Although cardiotoxic effects of many alcohol groups have been determined, no studies of butanols have been performed. The purpose of this study is to determine the effects of n-butanol, that is becoming increasingly common, on cardiac activity in larvae of zebrafish. n-Butanol, at different concentrations (10, 250, 500, 750 and 1000 mg / L) was started to be administered at 4th hour after fertilization and cardiac activities of zebrafish larvae at 96th hour were examined. As a result, n-butanol has been shown to have decreased cardiac activity and heart contraction power due to dose increase.

Keywords: n-Butanol, Zebrafish, Cardiac activity, Heart contraction power

# Influence of Several Physical Treatments on the Improvement of Some Quality Parameters of Locally Produced Olive Oil

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#### ABSTRACT

Virgin olive oil is the oil obtained from the olive fruit through physical procedures without refining or being mixed with other oils or any substances. The level of peroxide value (PV), free fatty acids (FFA), vitamin E, and total phenols in olive oil is are some of the olive oil quality characteristics that affect, its keeping ability and health significance. The aim of this study was to improve the overall quality characteristics of locally produced olives oil and in particular, reducing PV and FFA contents, to enhance its keeping and to enhance its health promoting effect. Fresh olive oil was exposed to oxidation, then eluted separately through a glass column ( $10 \times 75$  cm) loaded with an adsorbent of one of the following materials: activated charcoal, calcium chloride, aluminum oxide, bentonite, pectin, clay, sweileh sand, acacia powder, acacia, arabic gum, and silica gel. The oxidized oil eluted through some adsorbent exhibited an excellent oil quality improvement significantly as indicated from the use of silica gel that improved the PV from 32.39 meq/kg to 25.06 meq/kg, also reduced the FFA from 1.675% to 0.915 %, arabic gum could reduce the PV from 32.39 meq/kg to 17.79 meq/kg, while was reduced the FFA from 1.675% to1.05%, acacia powder reduced the PV from 32.39 to 21.98 meg/kg and FFA from 1.675% to 1.005%. Calcium chloride reducing the PV from 32.39 to 24.11 and FFA from 1.675% to 1.145%. While other adsorbent are effective in lowering significantly FFA but not effective in lowering PV. Clay and activated charcoal were not effective in reducing both PV and FFA. On other hand, all treatments exhibited a negative impact on the total phenolic and vitamin E contents in the tested samples. Different responses on specific absorption coefficient at 232, and 270 nm were obtained from the eluted oxidized olive oil through several adsorbent.

#### ACKNOWLEDGEMENT

This work was supported by Deanship of Scientific Research at the University of Jordan

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# Proceedings of the 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences (ICOFAAS 2019) Antalya, Turkey, 8-11 November 2019 Biocontrol Organims Against *Pectobacterium carotovorum* on Peper Stem Rot

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#### ABSTRACT

Stem rot caused by *Pectobacterium carotovorum* subsp. *carotovorum* is a major constraint in the production of on pepper (*Capsicum annum* L.). Chemical control methods are applied against *Pectobacterium caratovorum* but pesticides have harmful effects on human and environment health. For this reason alternative control methods have gained importance. The objective of this study was to demonstrate that 13 bacterial biocontrol agents [(3 *Bacillus megaterium* (B60d, TV6D, TV91C), 2 *Paenibacillus polymyxa* (Ç9, TV12E), 2 *Bacillus subtilis* (TV6F, TV17C), 1 *Pantoea agglomerans* (B79), 1 *Agrobacterium radiobacter* (A16), 1 *Bacillus megaterium* (FDG161) 1 *Bacillus atrophaeus* (FD1), 1 *Bacillus pumilus* (TV3D), 1 *Pseudomonas fluorecens* (FDG37)] efficiacy against *P. c.* subsp *carotovorum* in *in vitro* conditions. In dual culture tests, the inhibition zones of bacterial biocontrol agents were determined as 50.00 -10.00 mm. The most effective bacterial biocontrol strain was B60d (50 mm). Subsequent it is greatly important to test B60d on pepper fruits.

# Investigation of the Possibilities of Using Bacterial Bioagents in Biological Control Against Carrot Lead Mold Disease (*Botrytis* sp.)

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#### ABSTRACT

Variety, harvest and sowing density due to the pathogens is effective on the yield and quality on carrot (*Daucus carota* L. var. *sativus*) cultivation. Lead mold disease (Botrytis sp.) causes a significant loss of yield among these disease factors. The intensive use of the chemical has reached a threatening level for the environment and human health in the fight against this factor. For this reason, recently, great importance has been given to biological struggle studies which are environment friendly and can be effective for a long time. The effectiveness of 67 bioagent bacteria strains which were determined to be effective against lead mold which is an important disease factor in carrot depots, were tested in in vitro conditions in this study. According to the results obtained from the experiments; *Pseudomonas fluorescens* Biotip F (FDG37), *Bacillus subtilis* (TV6F) - *Paenibacillus macerans* (RK1443 A) strains, was effective against carrot Botrytis sp, 66.22%, 65.32%, 63.51%, 57.66%, 56.76%, 52.7% respectively. As a result; it was concluded that these strains can be used successfully in biological control of carrot lead mold disease.

Keywords: carrot, lead mold disease (Botrytis sp.), biological control

# Investigation of the Possibilities Use of Some Bacterial Biopesticides in the Biological Control against *Aphis pomi* (De Geer, 1773)

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#### ABSTRACT

In this study, the usability of the bacteria biological control against Aphis morphis, an important pest of apple trees, was investigated. For this purpose, previously a total of 4 bacterial strains that are effective in biological control against different pests were determined: Bacillus thuringiensis subsp.kenvae (FDP-8), Bacillus cereus GC subgroup B (FD-63), Bacillus atrophaeus (FD-17) and Brevibacillus brevis (FD-1), culture media as a negative control was used and the red sunny insecticide as a positive control. The study was designed as two stages, in the first stage the bacteria were analyzed under laboratory conditions. Strains that were successful in *in vitro* conditions were tested in field conditions in the second stage. According to the results obtained 72 hours after *in vitro* application; FDP-8 and FD-63 of the tested bacteria strains were found to reduce the harmful population by 90% and 83%, respectively, compared to the negative control. Likewise, the positive control was 100% successful after 72 hours, but the difference between the effective bacteria was not significant when statistically compared to effective bacteria. These two effective bacteria were applied to the shoots with intense pest populations in the field conditions and live and dead pest populations in 10 cm shoots were calculated. According to the results obtained 72 hours after in-vivo application; FDP-8 and FD-63 strains tested were effective against pests with the percentage of 95% and 51%, respectively. Likewise, the positive control was 100% successful after 72 hours but the difference was not statistically significant when compared to FDP-8, one of the most effective bacteria. As a result; It is thought that biochemical bacteria used in this study can be used successfully in biological control against Aphis pomi, which is an important pest of FDP-8 strain, and it can also be an alternative to chemicals threatening human and environmental health.

Keywords: Biological Control, Biopesticides, Bacillus thuringiensis subsp. kenyae, Aphis pomi

# Thymus Fontanesii Boiss & Reut Against Human and Plant Fungi

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#### ABSTRACT

Dermatotophytes and unicellular fungi cause serious health problems for humans; and plant-pathogenic fungi are very detrimental agents to our crops, and therefore cause very significant economic losses. In order to combat these agents, we will test bioactive complexes extracted from the leaves of an Algerian plant: Thymus fontanesii boiss & Reut.

Three extracts were prepared according to different processes: hydrodistillation with Clevenger (essential oils), maceration (methanolic extract) or by infusion (aqueous extract). The components of each extract were determined by gas chromatography coupled with mass spectrometry (GC / MS) and high performance liquid chromatography (HPLC). Two human pathogenic species (*Trichophyton rubrum* and *Candida albicans*) and one plant pathogen (*Rhizopus stolonifer*) were cultured in the presence of different concentrations of plant extracts; microscopic and macroscopic observations were made to see the effect of plant extracts on the morphology and growth of hyphae and reproductive structures of the tested fungi.

According to our results, the studied thyme species is carvacrol chemotype (52, 138%), and we have identified by HPLC a variety of metabolites responsible for the fungicide activity of the plant: quercitin, myricetin, Berberine, trans cinnamic acid. The macroscopic and microscopic study of mycelia revealed a reductive, even inhibitory, effect of hyphae growth and spore production. All the extracts have affected the growth of the fungal species tested, and essential oils have been effective even at very low concentrations (0.01%). The most apparent fungicide effect was observed in *Rhizopus stolonifer* when treated with aqueous extract (no growth was observed during the incubation period with all concentrations of aqueous extract). Based on our study, the antifungal activity of the extracts depends on many factors, including the nature and concentration of the extract, as well as the fungal strain tested.

**Keywords:** *Thymus fontanesii* Boiss & Reut., fungicide effect, essential oil, methanolic extract, aqueous extract, phytopathogenic fungus, dermatophytes.

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# Cedrus atlantica M. needles Essential Oil: Active Ingredient of Dermal Cream with Pronounced Anti-Inflammatory Effect

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# ABSTRACT

The treatment of inflammation uses steroidal and nonsteroidal anti-inflammatory drugs. Although these molecules are effective, they most often have undesirable effects. So, secondary metabolites of some plants could be an alternative in anti-inflammatory therapy because of their better accessibility and less toxicity in general. This study done on the needles essential oil of Atlas cedar (*Cedrus atlantica* M.) aims to identify the different biochemical compounds, formulate a dermal cream and study the therapeutic effect of dermal cream.

The essential oil from fresh needles of *Cedrus atlantica* obtained by hydrodistillation, was analysed by GC–MS. Anti-inflammatory activity of the dermal cream formulated from essential oil was evaluated by Croton oil-induced ear swelling in mice models.

Thirteen components, with a total yield of 0.12% of the essential oil, were identified. sesquiterpenoids represented the main fraction with  $\beta$ -Himachalene (30.08%),  $\alpha$ -Himachalene (16.38%), Longifolene (14.45%) and  $\delta$  –Cadinene (4.08%, 4.55%) as major constituents. Application of the dermal cream of Atlas cedar needles essential oil reduced inflammation significantly (P < 0.01) in croton oil ear swelling and present an important anti-inflammatory effect compared to the reference product (Soduim Diclofenac gel at 1%) with an edema reduction of 45,55%.

Dermal cream formulated with needles essential oil has very pronounced anti-inflammatory effect similar to that of Soduim Diclofenac gel at 1%.

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# Antioxidant Activity of Phenolic Compounds of *Thymus fontanesii* Boiss. and *Reut*.

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# ABSTRACT

Free radicals or reactive oxygen species are the direct cause of various pathological state such as cell ageing and cancer. Plants are a Precious source of many molecules with biological activities of interest to humans, pharmacological or nutritional. This work aims to determine the phenolic and flavonoid content in Thymus fontanesii Boiss. & Reut. Leaves ; by spectral methods, and assess the antioxidant power of various extracts (methanolic extract, aqueous extract, essential oil) of the plant (in vitro test) using three chemical techniques: the reduction of iron ions (reducing power), the trapping of the free radical 2,2-diphenyl-1-picryl-hydrazyl (DPPH) and the scavenger power of H<sub>2</sub>O<sub>2</sub>. The obtained results showed the richness of the studied plant in phenolic compounds (26 .2899  $\pm$  1,4603 mg Eq GA/g dry leaves) and flavonoids (8.423  $\pm$  0,652 mg/g). The study of antioxidant activity has shown that the plant extracts have very interesting antiradical properties, and i twas found that the essential oil expressed the highest antioxidant properties compared to the other two extracts but which remains lower than that of the positive control.

**Keywords**: *Thymus fontanesii* Boiss & Reut., phenolic compounds, flavonoids, antioxidant activity, essential oil, methanolic extract, aqueous extract.

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# Salt Tolerance of Eleven Durum Wheat (*Triticum durum* Desf.) Genotypes during Seedling Growth

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# ABSTRACT

Salinity is an important source of abiotic stress, limiting crop performance in most arid and semi-arid areas of the world. This research was conducted at the National Center for seeds and plants Control and Certification (CNCC), regional laboratory of Setif (Algeria). It aims to assess the effect of salt stress induced by sodium chloride (NaCl) at different concentration levels (0, 50, 100 and 150 mM) on 11 durum wheat (Triticum durum Desf.) varieties at germination and early seedling stages. There were statistically assured significant differences among the genotypes for all salt concentrations and all observed parameters. The average effect 'genotype' indicates that the minimum and maximum extreme values are depending on the trait and genotype. No variety had the best values for all traits. The average effect 'salinity' explains almost all of the total variation observed. Values of the measured traits were reduced as the stress intensity increased. Under high salt concentration (150 mM), these reductions were, relatively to the control, 7.3, 58.2, 30.9, 38.3, 70.6 and 70.2%, respectively for germination rate, root length, roots number, coleoptile length, root fresh weight and seedling fresh weight. The 'genotype x salinity' interaction indicates that the response to salt stress is a function of variety, stress intensity and organ type. The genotype Sitifis was the most sensitive to salinity-induced stress. Waha, Megress and GTA dur were the most tolerant genotypes that could be used in breeding programs and molecular physiological studies for development of high-yielding salt-tolerant durum wheat varieties.

Keywords: Triticum durum, NaCl, Genotypic variation, Selection, Tolerance.

# ACKNOWLEDGEMENT

Thee authors would like to express special thanks to the personnel of CNCC for facilities and help in conducting the experiment.

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# Bio-Ecology of Leptocybe Invasa on *Eucalyptus globulus* in the North-East of Algeria

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# ABSTRACT

The Eucalyptus species are endemic to Australia, although a few were introduced into Algeria by the Frensh colonization at the 19th century. In the last 10 years, an invasive insect have been discovered in the Algerian Eucalyptus leaves (North-East region). *Leptocybe invasa* Fisher & La Salle 2004 (Hymenoptera: Eulophidae) appears to be a widespread species in Australia, ranging from northern Queensland to Victoria and Western Australiahas, as it has been detected from Algeria in 2010, The dynamics of *L. invasa* and its field infestation rate were followed since 2017, in *Eucalyptus globulus* plantation in the North-East Region in Algeria(Msila, Setif and Beni Aziz). Some bio-ecological parameters and control strategies of thes new pest is reviewed and discussed.

Keywords : Eucalyptus, parasitic insects, parasitoids, biological control.

# Contribution to the Study of Harmful Insects on *Eucalyptus* gomphocephala (Myrtaceae) in Algeria

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# ABSTRACT

*Eucalyptus gomphocephala* is an essence of the family Myrtaceae. It resists a certain salt content and the sea wind. Despite that, it subject to many factors of degradation as parasitic and harmful insects. Field trips (Batna, Ras El Oued and Jijel in the North Est of Algeria) from September 2017 to September 2018. The results made it possible evidence the presence of 2 harmful species: *Leptocybe invasa, Ophelimus maskelli* (gallicolous insects, Hymenoptera, Eulophidae). A procession of other species has been studied highlighting the presence of parasitoids such *Closterocerus chamaeleon* (Hymenoptera) parasitoid of *Ophelimus maskelli*. To these pests of other insects live on this essence permanently or temporarily. In all 5 orders divided into 12 families comprising 14 species that have been determined. Three main orders, Hymenoptera, Coleoptera, and Collemboles alone represent 12 species, while Lepidoptera and Hemiptera, are represented by only one species.

# ACKNOWLEDGEMENT

This work was made by Laboratory ADPVA

# Antimicrobial Activity of an Algerian Medicinal Plant: Inula viscosa

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# ABSTRACT

Objective: To investigate antibacterial activity, total phenolic and flavonoid content of ethanol (E) extracts of Inula viscosa root, stem and leaf parts selected from *Kherrata* regions of Algeries. Methods: The antimicrobial activity of different extracts (aqueous and ethanol fractions) from *Inula viscosa* effect with standard antibiotic (Gentamicin) were evaluated using the disc diffusion method. The total phenolic and total flavonoid content of these extracts was determined according to the Folin-Ciocalteu procedure and Aluminum chloride colorimetric assay respectively. Total phenolic content was measured by Folin Ciocalteu reagent. Results: All the extracts showed significant antimicrobial activities and contained important levels of phenols. The ethanol extract from *Inula viscosa* showed the greatest antibacterial capacity in the three systems of assay, which was probably due to its high content of polyphenols. Total flavonoid content was found equal for all extracts. Conclusions: Our results of antibacterial and antifungal assays were justified and partially supported the popular usage of the tested plants. The high antimicrobial activity found in the plant in this region suggested that *Inula viscosa* is a good source of natural antimicrobial compounds which might have benefits for health.

Keywords: Inula viscosa, Folk medicine, Flavonoids contents, Polyphenols, antibacterial activity

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# Needles Essential Oil of Algerian Aleppo pine (Pinus halepensis) Against Microbial Germs

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#### ABSTRACT

When antibiotic therapy was practiced for the first-time, the man thought won the battle against the pathogenic bacteria. But like any drug, antibiotics are not completely harmless to the body and can cause, in addition to their antibacterial action, a number of adverse effects. In order to identify new natural bioactive molecules from the aromatic plants of the Algerian flora, this study was conducted on Aleppo pine (*Pinus halepensis*) growing in Algiers (Algeria). Essential oil extracted by hydro-distillation from the needles of *Pinus halepensis* showed a yield of 0.47%. The GC-MS analysis identified 12 constituents. The major compounds are:  $\alpha$ -humulene (9.696%),  $\alpha$ -pinene (5.961%) and terpinolene (5.903%). Essential oil of *Pinus halepensis* presents a remarkable antimicrobial activity against *Pseudomonas aerogenosa* and *Saccharomyces cereviciae*. These antibacterial and antifungal effects are more important than those of tested antibiotics and fungicide.

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# Antibacterial Activity and Anatomical Study of *Eucalyptus globulus* L. bill

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# ABSTRACT

The essential oils from *Eucalyptus globulus* L. bill. was obtained by hydrodistillation method with a Clevenger apparatus with a yield of 0.20%. The extraction produced yellowish essential oils with a very strong odor. Essential oils of this plant exhibited an antibacterial effect on *Staphylococcus aureus* ATCC 25923 (40 mm) and *Shigella sonnei*(20 mm) while it present weak activity on *Escherichia Coli* ATCC 25922 and *Pseudomonas aeruginosa* ATCC 27853 (12mm), with inhibition zone of 12mm. The anatomical study of this plant shows three different types of glandular trichomes on leaves and stems, these secretory glands are a source of essential oil present in the plant.

## ACKNOWLEDGEMENT

This work was made by Laboratory of Natural Resource Valorisation, SNV

# Efficacy of the Seeds of *Ricinus communis* in Postharvest Control Against the Flour Beetle, *Tribolium confusum*

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#### ABSTRACT

To limit the use of synthetic chemical insecticides, a study on the research of plants based insecticides, as the best alternatives, was conducted in the present work. Seeds of *Ricinus communis* (Euphorbiaceae) were evaluated for their antifeedent activity and toxic potential against the flour beetles, *Tribolium confusum* (Coleoptera, Tenebrionidae). A hydroethanolic extract of the seeds was applied on flour discs at different doses (10, 20, 40, 60 and 100%) and given as food to adult beetles, under laboratory conditions. The seeds extract was antifeedent at the lower dose, 10%, with an antifeedent activity rate of 31% to achieve at the higher dose (pure extract) a rate of 68%. The extract was toxic with 18% of mortality after the ingestion of 2 mg, i.e., 4% of treated food at the dose of 60% to reach a rate of 50% of adult mortality after the ingestion of 1.6mg (3% treated food) when the food received the pure extract. The seeds of *R. communis* have an important antifeedent properties and could be used during the storing process as grains treatment against the development of *T. confusum*.

Keywords : Ricinus communis, seeds, Tribolium confusum, antifeedent, toxicity.

# Effect of UV-C Radiation on Date Fruit Quality

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# ABSTRACT

Exposure of Deglet Nour date to UV-C caused the decrease in its water loss, decreased the enzymatic activity of polyphenoloxidase until the 4th month of storage after irradiation. This exposure enhanced polyphenols content until the first month of storage. On the other hand, the UV-C participated in the improvement of the organoleptic characteristics of Deglet-Nour by the slowdown of the browning until the end of the storage period which lasted 6 months. The antioxidant activity of the irradieted fruit, which was evaluated by two methods, proved having a significant increase in the antiradical activity in addition to the reducing effect of iron.

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# **Comparative Study of the Physico-Chemical, Microbiological and Nutritional Characteristics of the Different Milks Collected in Algeria**

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# ABSTRACT

The nutritional value of milk lies in its richness in basic nutrients (proteins, fats and carbohydrates) but also in calcium, vitamins and trace elements. It is one of the few foods that is suitable for different age groups where it can be consumed fresh or as a processed product, especially in cheese and voghurt. In order to determine the physic-chemical characteristics, microbiological quality and nutritional composition of different locally collected milks; bovine, camel and caprine. These milks were compared at the same stage of lactation. Thus, the pH, the titratable acidity, the density, the total dry extract, the fat content, fatty acid and mineral salts were determined. We have also proceeded to the microbiological quality of these different milks. The results generated show that the pH of the goat milk collected during the lactation period is slightly lower ( $pH = 6.4 \pm 0.12$ ) than boyine (6.5)  $\pm$  0.05) and camel (6.6  $\pm$  0.14) at the same stage of lactation. Its Dornic acidity is 16.45  $\pm$  0.79 ° D. It is relatively high compared to bovine milks  $17.2 \pm 0.78$  ° D and camel  $17.72 \pm 0.5$  ° D.  $29.7 \pm$ 0.5 g/l, seems to be lower than those of bovine milks  $35.07 \pm 1.9$  g/l and goat  $48.39 \pm 4.9$  g/l. The density of camel milk (1025.2  $\pm$  1.68) is lower than that of bovine milk (1028.2  $\pm$  2.08) and goat (1034  $\pm$  1.69). At the same time, the total dry matter content of this milk is equal to (112.37  $\pm$ 4.7 g / l). It appears to be weaker than those of bovine and caprine milks. The microbiological quality of the milks analyzed is relatively satisfactory, due in large part to the animal hygiene and good milking conditions. The analysis of the lipid fraction showed that long-chain fatty acids (C14-C20) are predominant (have the majority). Polyunsaturated fatty acids are in minority. The results show that camel milk contains the highest proportion of polyunsaturated fatty acids. The analysis of the mineral composition showed that camel milk is richer in major mineral elements (Ca, Mg, Na, and K) than that of cow's milk and goat's milk with a significant difference  $P \le 5\%$ . The physicochemical composition of milk varies according to the animals' breed and feed, environmental conditions and lactation period.

**Keywords:** mid-lactation, milk camel, bovine, goat, physic-chemical, microbiological, nutritional.

#### ACKNOWLEDGEMENT

We express our sincere thanks to all those who participated from near or far in the realization of this work.

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# Study of the Antibacterial and Antioxidant Powers of *Lavandulas* stoechas, Medicinal and Aromatic Plants in the Northern Algiers Region

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#### ABSTRACT

The therapy against infectious diseases relies mainly on the use of antibiotics. Their prescription at will is often inappropriate especially with regard to viral infections. Abuse and non-compliance with doses led to the emergence of resistant strains making treatment ineffective. On the other hand, synthetic preservatives added to food for consumption have been limited in several countries because of their adverse health effects. In addition, the current trend of consumers seeking a more natural diet has increased in recent decades. This has led scientists to look for other natural remedies, filling chemicals, which are not toxic to human health or our environment. In this context, research has focused on secondary compounds excreted by plants. As a result, the published results indicate that plants are endowed with several biological properties. In our experiments, we tried to evaluate, in vitro, the antioxidant activity of the Lavender essential oil, Lavandulas stoechas (Lamiaceae), in addition to its antibacterial activity on five strains of bacteria (Pseudomonacea aeruginosa, Staphylococcus aureus, Klebssiela spp, Escherichia coli and Serratia spp) with diversified properties of medical, animal and food origins. After extraction of the essential oils by hydro-distillation with a Clevenger type apparatus, the evaluation of the anti-radical activity was estimated by the DPPH (2,2-diphenyl-1picrylhydrazyl) reduction method. In addition, the antibacterial activity was demonstrated by the diffusion method on agar medium. The results obtained demonstrated a dosedependent anti-radical activity, which increases with the increase in the concentrations of essential oils in the reaction medium. However, the antibacterial activity reveals variable responses depending on the nature of the bacterial strain studied. As a result, the results showed that the essential oil of Lavandulas stoechasa causes a high sensitivity to pathogenic Staphylococci. In addition, moderate susceptibility was noted for Escherichia coli, Serratia spp and Pseudomonace aeruginosa, while Klebssiela spp was found to be resistant to this essential oil studied. Finally, we can conclude that the essential oil of Lavandulas stoechas picked in the region of El Tarf (North-east Algeria) and tested in our work, enjoys significant antioxidant and antibacterial activity and can be a good alternative to some synthetic additives.

Keywords: Lavandulas stoechas, medicinal plant, antibacterial activity, antioxidant activity.

# Artificial Neural Networks Applied to the Analysis of Water Contamination by Pesticides

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# ABSTRACT

Pesticides are considered groundwater contaminants, especially in agricultural areas. The chemical composition of these products is often harmful to human health. The degree of contamination depends on several parameters such as rainfall, the nature of the soil, the microbiological composition of the rhizosphere, the depth of the groundwater, etc. Given this situation, it is very difficult to model such a phenomenon using conventional mathematical techniques in order to determine the impact on the health of the population. In this study a technique of artificial intelligence including artificial neural networks is proposed. Because neural networks have the ability to support multiple combined variables, their application in this area is adequate. Based on field data, we have established a predictive model that associates input parameters with their effects on public health as the output variable of the system. This allows upstream action on the quality and quantity of pesticides and chemicals used in agriculture to preserve the public health of the inhabitants of these areas

Keywords: Groundwater, pesticides, public health, ANN

# In Vitro Antioxidant and In vivo Anti-Inflammatory Properties of Algerian Alkanna tinctoria Roots Extracts

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# ABSTRACT

Alkanna tinctoria is a plant widely used in traditional medicine for its therapeutic properties. The aim of this work is to evaluate the antioxidant activity and the anti-inflammatory and analgesic activities of the methanolic extract (Met. E.) and the decoction (Dec) of Alkanna tinctoria roots. Quantitative analysis of polyphenols and flavonoids showed that Met. E was the richest in polyphenols while the Dec was richer in flavonoids. The antioxidant activity was evaluated by the DPPH scavenging test and the peroxidation inhibition assay using  $\beta$ -carotene. Dec showed a strong antiradical activity toward DPPH radicals. In the  $\beta$ -carotene bleaching test, both Dec and Met. E. exhibited an important inhibitory activity of peroxidation. The acute toxicity study showed that these extracts had no toxic effect at 2 g / kg. The anti-inflammatory activity tested by the model of xylene-induced ear edema, showed a significant inhibition of inflammation for Dec with 90 %. The treatment of mice with these extracts inhibited the pain induced by acetic acid with 97.45 % and 90.90 % for Dec and Met. E. respectively. These results confirm the therapeutic characteristics of Alkanna tinctoria, which must be supplemented by further research in order to its use in therapy.

# ACKNOWLEDGEMENT

This work was supported by MESRS (The Algerian Ministry of Higher Education and scientific Research).

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# Association between the A1298C Polymorphism of MTHFR, Blood Folate and Vitamin B<sub>12</sub> Deficiency, and Elevated Serum Total Homocysteine in Young Patients with myocardial İnfarction in *Aures* region (Algeria)

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#### ABSTRACT

The aim of this molecular study was to evaluate a possible association between hyperhomocysteinemia and the different genotypes of the polymorphism A1298C of MTHFR gene, based on clinical and biochemical data, in pathological population, residing in Aures region (Northeast Algeria). The present study included 14 young patients (under 40 years) this age group is very rare, all patients were in the chronic phase of myocardial infarction (MI). Different biological methods were used to evaluate the biochemical and genetic status of our study population. The Competitive immunoassay was used for the determination of vitamins ( $B_9$  et  $B_{12}$ ), total homocysteine, CRP and BNP. By the enzymatic colorimetric method we have explored lipid profile, urea, ASAT and ALAT.The identification of this polymorphism was determined by using the Real-Time Polymerase Chain Reaction - Fluorescence Resonance Energy Transfer (Real-Time PCR -FRET) technic; it was carried out with a Light Cycler ® 480 II Instrument. The investigation of this mutation on the MTHFR gene showed a non-significant association of moderate hyperhomocysteinemia (Hhcy) with (AC) and (AA) genotypes (P=0.064). Equally, no significant differences were observed in the distribution of  $B_9$  and  $B_{12}$  (P=0.355, P=0.814, respectively). However, the levels of vitamins  $B_{12}$  and  $B_{9}$  in the moderate Hhcy groups were slightly lower than those observed in groups of normal homocysteinemia, especially in mutated genotypes (AC). It appears that there was a deleterious effect of the mutated genotype on the levels of vitamin  $B_9$  and  $B_{12}$ . However, this action remains not significant. No significant difference in the biochemical, clinical and demographic patient characteristics according to the different genotypes of MTHFR A1298C plymorphism were revealed. Our study supports the hypothesis, indicating that the polymorphism A1298C MTHFR could be considered an important risk factor for Myocardial Infarction, in the presence of moderate hyperhomocysteinemia.

**Keywords:** Myocardial infarction (MI), A1298C MTHFR polymorphism, Moderate Hyperhomocysteinemia, RT-PCR- FRET® 480 II, Aures, Algeria.

# ACKNOWLEDGEMENT

We would like to extend our gratitude and appreciation to Professor Rosa-Maria Guéant-Rodriguez, Professor Jean-Louis Guéant, all personnel of the Nutrition-Genetics-Environmental Risk Exposure Laboratory, Nancy (France) for the technical assistance to the carrying out this important work.

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# Foraging Behaviour and Pollinating Efficiency of the Honey Bee (*Apis mellifera* L.) (Hymenoptera : Apidae) on the Pear (*Pyrus communis* L.) in Constantine Area (Eastern Algeria)

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# ABSTRACT

Pollination by insects, especially domestic and wild bees, is of great agronomic and economic interest. Insects are involved in the pollination of about  $\frac{3}{4}$  of cultivated plant species including fruit trees (Vaissiere 2005) [1] and the economic value of this pollination represents about 9,5% of the value of world agricultural production (Gallai & al 2008) [2]. In this study, pollinating insects of the pear tree was studied in the region of Constantine (eastern Algeria); three mixed orchards in two localities in the region were prospected during the 2011, 2016 and 2019 flowering periods. The planted cultivar is Santa Maria (self fertile). The results obtained showed that the honeybee is the main visitor of the tree (between 91 and 99% of visits). All of its visits can be fruitful. Pollen is the most harvested floral product (more than 80% of visits) except in 2019 (16% of visits). The honeybee visited on average between 5 and 11 flowers / minute and between 10 and 22 flowers / tree. Its movements (42%) are more frequent on adjacent trees in the same row (distance = 3,5m). Yields obtained at fruit set are higher in the presence (between 34% and 85%) than in the absence of pollinators (between 8% and 44%). Flowers that received two visits from the honeybee produced more fruit (40%) than those that received only one visit (between 8 and 32%).

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# Linseed Oil (Linum usitatissimum) Toxicity and Safety of Use

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# ABSTRACT

In Algeria, the multipurpose plant species *Linum usitatissimum* has rarely been tested for its pharmacological or toxicological properties. This study aims to evaluate flaxseed extract toxicity on Artemia Salina larvae. The safety of pure linseed oil was also studied using in vivo tests of safety (ocular, anal and cutaneous), on a Californian rabbit model (06 adult animals). Based on the Artemia Salina larvae test, the lethal concentration 50 (LC<sub>50</sub>) is  $80\mu g / ml$ , which proves that flaxseed extract is not toxic to living cells. The determination of the ocular irritating potential of linseed oil was studied following a single instillation of the oil in the rabbit's eye. One hour later the Eye Irritation Index was (2.67), and on day 7, all signs of irritation disappeared, proving that flaxseed oil is not irritating to the eye. The safety test on the rabbit's anal mucosa using a single application shows no adverse effects. The evaluation of superficial skin aggression and skin irritation, on scarified or nonscarified rabbit skin, shows no oedema but a very slight erythema, which is fully transient and reversible after 48 hours of the oil's application. The primary skin irritation index of the oil is 0.75, indicating that flaxseed oil is slightly irritating to the rabbit skin. Knowing that, rabbit skin is much thinner and brittle than the human one. Knowing also, that other studies had certified that this oil favours wound healing and hair regrowth. Other clinical tests should be carried out to confirm other safety use aspects of this multi therapeutic properties oil.

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# Research and Identification of Fleas in Barn Swallows (*Delichon urbica*) Nesting in the Mouldi Achouri Colony of the City of Tebessa, Northest of Algeria

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#### ABSTRACT

The search for fleas was carried out amongst the *Delichon urbica's* young swallows' nests that are located in the Mouldi Achouri colony of the city of Tebessa in the North East of Algeria during the season of 2018-2019. In order to capture young individuals of the barn swallow, six trips were made to Mouldi Achouri colony in Tebessa between April and July 2018. To collect ectoparasites, all parts of the swallow's body were examined visually. The results have shown the existence of five genres of fleas, including *Ceratophyllus gallinae*, *Ceratophyllus hirundinis, Callopsylla sp., Cimex hirundinidus, Xenopsylla trispinis and Ornithomya sp.* And indeed, the order of Siphonaptera is the most abundant.

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# Contribution to the Evaluation of Some Reproductive Parameters in Cattle

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#### ABSTRACT

The objectives of the present study were to evaluate the reproductive parameters of 200 Holstein dairy cows, to study some factors that may influence these parameters and to characterize the metabolic profile of repeat breeder cows (RBC). The experiment was carried out on a private farm located in the North of Algeria. Fertility was estimated for each cow by calculating the parameters: Calving-First service interval (CFSI), calvingconception interval (CCI), calving-calving interval (CI), first service - conception rate (FSCR) and fertility index (FI). Parity, dairy production and BCS were used as covariate to determine their effect on CCI and FSCR dependent variables. Blood serum parameters such as glucose, total protein, triglycerides, cholesterol, urea and B-hydroxybutyrate were assayed by spectrophotometry. Insulin and progesterone serum levels were detected by immunological tests. There was a high CS1 interval. Also, 44% of animals had a prolonged CCI (>110 days) and the FSCR was estimated at 42%, below the recommended values (>60%). RBC were noted in 35% of the total, higher than the intended standard (<15%). FSCR and CCI were significantly influenced by parity and BCS (P < 0.001). No significant milk yield effect on FSCR were reported (P>0.05) but CCI was significantly related to this parameter (P <0.001). RBC were characterized by low levels of insulin and high urea concentrations compared to fertile females (P < 0.001). In the conclusion, the reproduction parameters obtained in this study were very far from the standard objectives recommended for effective management of reproduction. It can be due to bad heat detections or/and undernutrition.

Keywords: Algeria, dairy cow, Holstein, infertility, insulin, urea

# Preserving Organoleptic Properties of Harvested Date Using Modified Atmosphere

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#### ABSTRACT

We studied the impact of two modified atmospheres low in  $O_2$  on the browning process and quality of 'Deglet Nour' date fruit, during one month of storage, through analysis of changes in browning index, weight, titratable acidity, contents of water, ascorbic acid, total soluble solids and total phenols. Treatments, especially MAP<sub>2</sub> (2% O<sub>2</sub>, 5% CO<sub>2</sub>) maintained fruit quality by slowing browning and loss in water, total phenols and ascorbic acid contents.

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# Joint Use of Remote Sensing and Phytoecological Field Diagnosis to Study the Impact of Desertification on Plant Biodiversity. Case of the Wilaya of Saida, Algeria

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#### ABSTRACT

This work is a contribution to the monitoring of desertification and its impact on plant biodiversity in Saida wilaya by analyzing the evolution of vegetation and the dynamics of land use through a diachronic study of the vegetation. The objective is to develop a methodology based on the coupling of remote sensing techniques associated with applications from the G.I.S Geographical Information Systems to field observation techniques through a phytoecological diagnosis based on vegetation surveys. The approach assumes that the SAVI is an indicator of degradation to the extent that a decrease in the value of SAVI means a decrease in vegetation and therefore a degradation of the vegetation. The data used are two LANDSAT TM images from the years 1988 and 2013, transformed into a soil-adjusted vegetation index (SAVI). The qualitative assessment of the canopy dynamics was carried out in the form of a synthesis on the results of the three components under vegetation dynamics: vegetation indices, cover and phytoecological records. The combination of all these data allows the generation of several thematic maps. The results confirm the state of degradation that threatens biodiversity, characterized by a sudden change in plant groupings, from which a regressive evolution of Alfa and Armoise is accompanied by an evolutionary evolution of Spart. A map of the changes was drawn up, of which two classes of changes were established: positive change, negative change.

#### ACKNOWLEDGEMENT

This work was supported by LGEO2D (Laboratory of geomatics and sustainable development).

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# **Placental Cells in Pre-Eclampsia**

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# ABSTRACT

Pre-eclampsia is a complication of pregnancy that usually occurs during the third trimester of pregnancy. It is secondary to placental dysfunction which is responsible for the release into the maternal circulation of various substances responsible for activation or even injury to the maternal endothelium. Pre-eclampsia is a major cause of maternal morbidity, preterm birth and perinatal mortality worldwide. Its incidence varies between 3 and 14% of all pregnancies worldwide. The purpose of this work is to determine the effects of antioxidants (vitamin C and E) on placental cell function in culture, isolated from control women and women with pre-eclampsia.

The pieces of the placenta are taken just after delivery on two separate parts (maternal and fetal) and then treated with collagenase. The cells are collected and then cultured in RPMI-1640 medium at 37 ° C in an atmosphere saturated with 5% CO<sub>2</sub> for 24 hours. After culture, the MTT test, the glucose consumption, the MDA and GSH levels are analyzed.

Our results show a decrease in cell proliferation, reduced intracellular glutathione levels, and increased glucose consumption and intracellular levels of MDA in placentas of preeclamptic women compared to controls. The supplementation of the culture medium with vitamin C and E induces a correction of anomalies, particularly oxidative ones.

The management of pre-eclampsia requires hospitalization that will allow extremely regular monitoring of the future mother. This monitoring also makes it possible to measure the impact of the disease on the fetus (via the evaluation of its active movements, its cardiac activity, uterine height, etc.).

Keywords: pre-eclampsia, placental cells, vitamins C and E, reduced glutathione, MDA.

# Volatiles Constituents of Chloroform Extract of *Limoniastrum Guyonianum* and Their Antiproliferative Activity

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# ABSTRACT

*Limoniastrum species* have immense medicinal importance and are therefore used in the treatment of dysentery and gastric infections [1]. *Limoniastrum guyonianum* Durieu was described for the first time by Boissier from eastern of Algeria [2]. It is an endemic shrub from North Africa and locally known by "Zeïta" [3]. This endemic specie has been used by local communities as forage for camels [4].

This study aimed to investigate the composition of the chemical constituents from Chloroform extract of *Limoniastrum guyonianum* using gas chromatography and mass spectroscopy (GC-MS). In addition the antiproliferative activity against HeLa cell lines was examined.

The obtained chloroform extract was subjected to GC and GC-MS analysis which revealed the presence of twenty compounds and dominated by free alkanes. The main compounds were Trans-2-phenyl-1,3-diox Olane-4-methyl octadec-9,12,15-trienoate, (2-Pentanone,4-hydroxy-4-methyl and (3-Chlorophenyl) acetylene.

The antiproliferative activity of CHCl<sub>3</sub> extract (IC<sub>50</sub>:  $50.369\pm0.020\mu$ g/mL), was shown to increase of the activity depending to dose increasing against HeLa cells. The bioassay of chloroform extract showed that, it is potent against for antiproliferative activity.

# ACKNOWLEDGEMENT

This work was supported by The Ministry of High Education and Scientific Research.

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# Anti-proliferative Effect in HeLa Cell Line of *Genista Ferox* Stems Extracts

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#### ABSTRACT

Cancer is one of the most common diseases of the century. Despite numerous efforts to find medications to treat the disease, much has failed due to the undesirable side effects of the medicaments that have led to many deaths [1]. So the solution was to find a drug without undesirable side effects, and the first destination was the plants of some families known in ancient medicine. Medicinal herbs are important for cancer treatment due to the presence of multiple chemical compounds for discovering new active materials against cancer. In a continuous of our study on *Genista ferox* specie which belonging to this Fabaceae family, this later is known for its richness in various active compounds such as phenolic acids and flavonoids [2-3]. Chloroform, ethyl acetate, *n*-butanol and methanol stems extracts of *Genista ferox* were subjected to the estimation of anti-proliferative activity against HeLa cell lines using xCELLigence system. The result shows that ethyl acetate extract has the highest anti-proliferative activity at the lowest concentration tested.

Keywords: Genista ferox, Fabaceae, anti-proliferative, HeLa cell lines.

#### ACKNOWLEDGEMENT

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# In Vitro Antioxidant Activity of Extarcts from Algerian Plant (Inula)

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#### ABSTRACT

Oxidative stress is defined as the body's inability to defend against reactive oxygen species due to endogenous equilibrium disruption between the latter and oxidizing agents. This imbalance potentially leads to structural and functional damage [1].

The antioxidant activity of the extracts (Chloroform, Ethyl acetate, Methanol and Butanol) from *Inula* plant was evaluated by three methods: free radical scavenging using DPPH,  $\beta$ -carotene bleaching activity, antioxidant capacity cupric reducer (CUPRAC). BHA and BHT have been used as antioxidant standards for comparison of activities. The Ethyl acetate extract shows the most effective activity for the three methods used: DPPH,  $\beta$ -carotene and CUPRAC with IC50 equal to  $19.74 \pm 0.39 \ \mu\text{g} / \text{ml}$ ,  $1.17 \pm 0.01 \ \mu\text{g} / \text{ml}$  and  $8.59 \pm 0.17 \ \mu\text{g} / \text{ml}$  respectively, followed by the n-BuOH extract with IC50 equal to  $34.5 \pm 0.28 \ \mu\text{g} / \text{ml}$ ,  $2.45 \pm 0.08 \ \mu\text{g} / \text{ml}$  and  $12.14 \pm 0.46 \ \mu\text{g} / \text{ml}$  respectively.

**Keywords**: *Inula*, DPPH, CUPRAC, β-carotène, antioxidant activity.

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# Valorisation of Natural Resources, Evaluation of Phenolic Centents and Antioxidant Activity of an Algerian Medicinal Plant (*Santolina*)

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### ABSTRACT

The potential of plants as sources for the production of new drugs is largely untapped. The study of the biological activities of plant extracts is of great interest for the treatment of various diseases and for the promotion of traditional medicine [1]. In this perspective, our study aims to valorize *Santolina* medicinal plant through the evaluation of the antioxidant activity of its extracts: Chloroform, Ethyl acetate and Butanol. The polyphenol assay was performed using the Folin-Ciocalteu reagent, as for the antioxidant activity, it was evaluated by free radical scavenging using DPPH and cupric reductive antioxidant capacity (CUPRAC). Among the extracts evaluated, the ethyl acetate extract shows the most effective antioxidant activity at IC50 =  $9.72\pm0.12\mu g$  / ml and  $7.74\pm0.09\mu g$  / ml (for DPPH and CUPRAC tests respectively), this efficiency is due to the high content of polyphenols (764.00± 1.15µg EAG / mg) of this extract.

Keywords: polyphénols, Santolina, DPPH, CUPRAC, Plant

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# Study and Consequences of a Significant Presence of Germs Sulfite Reducers' "Clostridium Butyric" in Raw Milk on Cheese Production

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#### ABSTRACT

Cow's milk is of particular interest to all populations, as it is perfectly in line with human requirements given its high content of basic nutrients (carbohydrate; protein, fat). However, it is a very perishable product and it constitutes an excellent environment favourable to the development of microorganisms. (E. jacob and al., 2011). It has been found following analyses that the number of total flora mesophilic aerobic in registered cow's milk is  $72*10^6$ CFU/ml is slightly higher than the norm as well as the number of coliforms in cow's milk is  $1.4*10^3$  for all samples published by (JORA,2017) respectively  $10^5$  CFU/ml and  $10^3$ CFU/ml). Keywords: raw milk, sulfite reducers', economic losses, cheese swellings, ultra filtrationOur study is also based on the research in raw milk of sulfite reducers' germs which can nevertheless be responsible for serious defects in various types of cheese by producing gases that explode them and give unpleasant tastes and smells and by causing very significant economic losses ( P. Drouin and al., 2008) alto our cheese industry, knowing that milk can contain a high number of butyric clostridia's which are non-pathogenic germs for humans and are present in animal feed (forage; silage) so they are a general hygiene problem and the phenomenon affects cooked pressed cheeses and some uncooked cheeses made in our dairies. The number of sulfite reducers' germs in raw milk from the farms we studied is  $1.6*10^1$  CFU/ml, which is lower than the Algerian standard of less than 50 CFU/ml for bacteria sulfite reducer's anaerobes. (JORA, 2017). The identification of clostridium butyric by gallery Api system A was performed in raw milk. The milk contains less than 1,000 spores/l for this purpose it is of good quality and hygienically acceptable (M.Vissers and *al.*, 2006) it can be used for production. To avoid the swelling of the cheese and especially considerable economic losses, it is desirable, according to P. Kumar in 2013 in the cheese industry, to treat all raw milk by bactofugation (a process that uses centrifugal force) or ultrafiltration.

## ACKNOWLEDGEMENT

This work was supported by the fraud repression laboratory in Sour El Ghozlane Bouira

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# Halotolerant PGPR: A Hope for Cultivation of Lucerne in Saline Soils

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#### ABSTRACT

Plant growth is strongly influenced by many biotic and abiotic factors. In Algeria, soil salinity and the scarcity of water resources are major factors limiting plant productivity, particularly in the Saharan regions. Sustainable agriculture, which today incorporates biotechnological practices, has become a key concept. It involves the use of so-called "useful" microorganisms, such as Plant Growth Promoting Rhizobacteria (PGPR), which allow plant growth to be improved by different ways (biofertilization, biocontrol, bioremediation, biostimulation, etc.).In the same context, this study was carried out to investigate the effects of bio-fertilization by two plant growth promoting rhizobacteria isolated from the lucerne nodules on the salt tolerance of the same plant (Medicago sativa L.) at germination and growth stages. In absence of salt stress, the PGPR seems to have little or no effect on germination when inoculated singly. In fact, only a mixture of these PGPR showed a positive effect on germination. Inversely, all inoculations positively affect growth parameters. In opposite, under salt stress, all single inoculation and co-inoculation alleviate the adverse effects caused by salt stress on lucerne germination.Regarding to inoculated plants, salinity affected negatively growth parameters which remained better than without bacterial inoculation. Sodium content was more reduced in leaves under co-inoculated conditions than after single inoculation, this suppose that Na<sup>+</sup> ions were confined by used bacteria.With regard to others parameters, this study indicated clearly that under salt stress plants with higher leaf area, total proteins and chlorophyll contents were more tolerant to salt stress as compared to the plants with lower values for these attributes. These results indicate that all inoculations and co-inoculation with two PGPRs and in particular proportion 1/2 co-inoculation could mitigate the adverse effects of salinity on the germination and growth of alfalfa, suggesting that this biofertilization could be used as a new biotechnology that increases productivity and is a promising alternative not only to chemical fertilizers, but also to the negative effects of soil saline stress in arid and semiarid areas.

# Diversity of Bacteria Isolated from Root Nodules of Lucerne (Medicago sativa L.) Cultivated in Algeria

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#### ABSTRACT

Our work focuses on the evaluation of the biodiversity of bacteria isolated from nodules of Lucerne (Medicago sativa L.) cultivated in soils from various regions of Algeria. A total of 106 isolates were characterized on the basis of morphological, cultural and phenotypical properties. Phenotypic characteristics studied included growth rates, colony morphology, symbiotic traits, utilization of different carbon and nitrogen sources, tolerance to different abiotic stress such extremes temperature, salt, pH, drought and resistance to different antibiotics and heavy metals. For the genotypic analysis, the PCR / RFLP of the rDNA16S gene technique also called ARDRA (Amplified Ribosomal DNA Restriction Analysis) is used. The use of restriction enzymes (cfo I, Alu I, Dde I, Nde II, HaeIII, Hinf I, Msp I et Rsa I) has made it possible to ribotyping the isolates and evaluate their biodiversity. In addition, 16S rRNA gene sequencing is used for three isolates selected on the basis of the phenotypic study. The numerical analysis of the phenotypic characteristics by comparison between the strains taken in pairs was carried out by the UPGMA method. The results obtained showed that the 106 strains studied were distributed in various groups reflecting a great biological diversity. RFLP analysis of 16SrRNA genes revealed additional heterogeneity and 12 ribotypes were identified. This diversity is not clearly correlated with the geographic origin of the isolates. In addition, soil characteristics (pH and salinity) do not seem to affect this diversity. This is the first report evidencing that Lucerne nodule may be associated with strains such as Stenotrophomonas maltophila and Rhodococcus quinshengii identified by sequencing of the 16S rRNA. These results suggest that identification of the remaining strains will confirm the wide phenotypic and genotypic diversity of these alfalfa nodule-associated bacteria.

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# Effects of Exogenic Cinnamic Acid on Fusarium oxysporum f. sp. albedinis

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## ABSTRACT

Bayoud disease, or Fusariosis of date palm (*Phoenix dactylifera* L.), is caused by a soil fungus *Fusarium oxysporum* f. sp. *albedinis* (Killian and Mayor) Gordon (Foa). At the site of infection, an accumulation of hydroxycinnamic acid ware observed. The purpose of this study is to evaluate the effect of artificially applied cinnamic acid at different concentration (100, 200, 400, 600 mg/L) on three strains of Foa. Results demonstrated that hyphal growth of Foa arestrongly inhibited by cinnamic acid. However, mycotoxin production were greatly stimulated. The accumulation of AF in the Liquid Dextrose Potato culture medium is very important from 200 mg / L of cinnamic acid added. These data suggest that high concentrations of cinnamic acid is a risk factor and damage to the date palm.

## ACKNOWLEDGEMENT

This work was supported by PRFU (Research Projects University-Training) in Algeria.

# Physicochemical and Rheological Properties of Bunium Bulbocastanum Earth-Nut Flour

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## ABSTRACT

The species of the Bunium kinds are aromatic plants with medicinal properties, especially the earth nut. Which the fruits are used as aromatic and the leaves and roots consumed as vegetables. The present study was conducted in order to valorize its roots by studying the physicochemical and rheological properties of its flour. Samples were harvested at two different time periods from 4 regions in Algeria.

The results made it possible to conclude that this gluten-free flour contains about 7% of proteins; lipid and ash contents are 3.38 and 3.96% respectively. On the sensory level, the color parameters (L \*, a \*, b \*) conclude that there is a significant region effect and the harvest period on the color of the flour.

Our results reveal that the physicochemical characteristics of composite flours (FBT & FNT), when going from 5 to 10% substitution, improves MG rates, ash tenacity rate and the gluten index. However, it has a negative influence on the W force, the swelling and the P / L ratio of the dough.

This study would suggest that earth nut tubers could be properly incorporated in wheat flour up to 5-10%, but at higher doses with gluten-free flours.

Keywords: Earth nut, gluten-freeflour, roots, Bunium Bulbocastanum

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# Maturation, Fecundity and Spawning Season of the Cuttlefish Sepia officinalis in Oran Bay (Western Algeria coasts)

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#### ABSTRACT

In this study, from January to December 2013, 581 cuttlefish (*Sepia officinalis*), of which 328 females and 253 males were collected monthly from the Western coast of Algeria. Dorsal mantle lengths (ML) of all sampled specimens ranged between 6.5 and 23.8 cm with an average of  $12.57 \pm 2.59$  cm. The female-male ratio was 1.3:1. The size at first maturity was estimated as 9.2 cm ML for females and 8.9 cm ML for males. Monthly values of the gonadosomatic index (GSI) suggest that the spawning period of this species occurs between February and April, with a maximal GSI of 6.22% in March for females and between March and May, with a maximal GSI of 3.39% in April for males. The absolute fecundity of the cuttlefish, calculated from a sample of 46 female gonads, was between 122 and 885 oocytes, with an average value of  $424 \pm 161$  oocytes per specimen. The mean diameter of the oocytes was  $6,45 \pm 2,03$  mm and ranged from 1,28 mm to 9,89 mm.

#### ACKNOWLEDGEMENT

This work was supported by MESRS (the Algerian Ministry of Higher Education and Scientific Research.

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# Study of Genital Pathologies in Dairy Cows Encountered at the Ferdjioua Slaughterhouse

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#### ABSTRACT

It is obvious that causes of infertility are numerous, including the genital anomalies that play an important role in the failure of cattle breeding. In order to minimize economic losses, it is important to first define the genital pathologies encountered in Algéria in postpartum dairy cows. The aim of this study is to count the various pathologies of génital apparatus of the cow at the slaughter-house of Ferdjioua. The results obtained following the macroscopic study of 75 apparatus of the cow show a high frequency of the pregnant females reformed (10.66%), the physiological stage during whith our cows are most frequently cut down is the diestrus and incidence of the uterine infections proves to be the most significant followed by the cystic ovary. The frequencies of the ovarian affections obtained following are by descending order : Cystic ovary =8.95%, Smoth ovaries =4.48%, ovarian adherences =2.98%. The frequencies of the oviduct affections are : Salpingitis=4.47%, Pyosalpinx =1.49%, hydrosalpinx=1.49%. The frequencies of the uterus affections are ; Infectious diseases : 10.44%, 5.97% : congestive states, Double cervix: 1.49%. Our results generally consistent with the literature data.

Keywords: ovary, pathology, cow, cystic ovary, slaughter-house, Ferdjioua

# Characterization of Some Local Algerian Varieties of Durum Wheat (*Triticum durum*) Under Semi-Arid Environment

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#### ABSTRACT

Algeria is a country with significant biological wealth in all existing ecosystems. Nevertheless this wealth undergoes constantly and for several reasons, risks of loss or what is denominated commonly the erosion. The agricultural ecosystem is one of the ecosystems threatened by its biological diversity, particularly the biological diversity of local varieties of certain crops such as durum wheat. Local varieties of durum wheat are known as varieties resistant to the constraints of their environment but unproductive, like all local varieties of different cultivated species. In the context of protecting the genetic heritage of Algerian durum wheat from the risk of loss, studies are being undertaken to help its conservation. Among these studies, the characterization of local varieties in order to valorize them within the framework of the varietal selection, which constitutes a non negligible means to keep them on the circuit, thus avoiding the loss of a local genetic resource.

To this end and in the wake of these studies, six local Algerian varieties of durum wheat are being tested in a semi-arid environment in Sétif, Algeria, during the 2017/2018 crop year. And while following a conventional technical route and conducted under rainfed, the six local varieties studied have undergone the measurements of the yield and its components to have an idea of what a local variety can offer compared to another and consequently to characterize each variety for these criteria, and this after having in a subsequent study compared the behavior of local varieties with the varieties introduced. The results indicate that the local variety Oued Zinati is the most productive at 244.44 g/ m2 while Hedba 3 is the least productive of the local varieties studied with 201.16 g/m2. For yield components varieties; Oued Zinati, Glory of Montgolfier and Guemgoum R'khem are respectively distinguished by the number of spikes per square meter, the number of grains per spike and the weight of a thousand grains with respective values 208.00 spikes /  $m^2$ , 32, 01 grains / spike and 61.96 g. In this same order the local varieties that record the lowest values are Mohamed Ben Bachir with 142.67 spikes /  $m^2$ , Oued Zinati with 21.62 grains / spike and Mohamed Ben Bachir with 42.12 g. It was then that the variety Oued Zinati would be a potentially interesting local variety for grain yield and number of spikes, the variety Gloire de Montgolfier for the number of grains per spike and the variety Guemgoum R'Khem for the weight of a thousand grains. The characterization of each local

variety for the yield criteria and its components gives it an identity that will indicate and facilitate the correct choice in crosses for varietal improvement. Although local varieties are varieties known to be stable for these traits, it would be interesting to multiply the tests in time and space to confirm the results presented in this study.

**Keywords:** Durum wheat, local varieties, grain yield, components of yield, characters, variety selection.

# Fertilization of Cultivated Agricultural Soil of *Solanum tuberosum L*.by Waste Sludge from the Wastewater Treatment Plant of Tiaret (Algeria)

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#### ABSTARCT

Sewage treatment always produces a significant amount of waste sludge. The different uses of sludge disposal have negative consequences for the environment. Agricultural use may appear in some situations as an alternative to current solutions, both to optimize the degradation and recycling of organic and mineral elements. The objective of this study is to valorize waste sludge from the Tiaret wastewater treatment plant in agriculture. *Solanum tuberosum L.* has been selected because it is a demanding plant in mineral elements. The species fertilized with different sludge doses: 20%, 40% and 60%, mixed with soil and 100% sludge. Biometric measurements were carried out during three months of planting, showing the positive effect of the mud on the growth of the plant. The biometric values recorded, for all the doses, far exceed those of the control plants (100% soil), with high values recorded in the mixture of 60% sludge.

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This work was supported by the Tiaret wastewater treatment plant

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# Sodium Benzoate Affects Male Reproductive System in Swiss albino Mice

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#### ABSTRACT

Sodium benzoate (SB) is a commonly used as a chemical preservative in a variety of foods, beverages and condiments to prevent alteration or degradation caused by microorganisms during storage. The present study investigates the effect of sub-chronic consumption of sodium benzoate on male reproductive function in Swiss mice. Eighteen Swiss male mice were randomly divided into three groups of 6 mice each. Control group was given drinking water, and treated groups were given sodium benzoate at doses of 0.1 and 1%. After 13 weeks, we observed a significant decrease in sperm count, sperm motility, and serum testosterone levels as well as an impaired testes histology in 1% sodium benzoate induces impair spermatogenesis and sperm quality which affects the reproductive performance of male Swiss mice.

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# Wheat Seedlings Growth Response to drought Under PEG6000-induced Osmotic Stress Conditions

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#### ABSTRACT

Adaptation to climate change is legitimately the primary concern of Algeria's agricultural sector. An effective way to increase or at least stabilize soft wheat production in the face of new climate change challenges is to develop improved varieties through plant breeding. The latter is based, on the one hand, on the exploration of a wide genetic diversity within cultivated types and wild relatives and, on the other hand, on the implementation of screening tests on adaptive traits. The present study was conducted at the Algerian National Institute of Agricultural Research (INRAA), research unit of Sétif whose purpose is to evaluate the responses of growth and germination to water stress induced by PEG 6000 at different doses (0, 150 and 250 g/l) of a collection of common wheat varieties with different breeding origin. from a variety of common wheat varieties. The results indicate significant genotypic differences within the plant material evaluated. The minimum and maximum extreme values are variable according to the trait measured and the genotype evaluated, no variety has the desired values for all traits at once. Means of the measured traits are reduced as the degree of water stress increases in intensity. These reductions can go, under 250 g/l PEG6000, up to 80.36%. In this study, Anapo, Nesser, Ziad, Mahon-Démias, Tidis, Guadalupe, Ain Abid, Tamezghida, Zidane, Hodna, Mezghana and Buffalo are the most tolerant varieties to water stress. The outperforming genotypes under drought stress conditions can be useful in future wheat breeding programs, and early selection for the traits recommended in this study will be effective for developing high-yielding and droughttolerant wheat varieties.

Keywords: Triticum aestivum, diversity, water stress, climate change, selection, tolerance.

#### ACKNOWLEDGEMENT

The authors would like to express special thanks to the personnel of INRAA-Sétif for facilities and help in conducting the experiment.

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# The First Association of Hb Knossos: (*HBB*: c.82G>T) WITH (*HBB*: c.118C>T) Mutation Causs Thalassemia Homozygous in Algerian Children

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#### ABSTRACT

Beta-thalassemia is the most common disease among hemoglobinopathies in Algeria. Mutations found in Algerian beta-thalassemia patients constitute a heterogeneous group, consisting mostly of point mutations. Only in very rare cases did deletions or insertions cause affected or carrier phenotypes. Hb Knossos (*HBB*: c.82G> T) is a rare variant. In this study, we aimed to investigate the effect of compound heterozygosis for Hb Knossos (HBB: c.82G > T) and (*HBB*: c.118C > T). To our knowledge, this is the first report of such a combination related with beta-thalassemia major phenotype in a Algerian family, we used the minisequencing assay as a rapid screening procedure to identify most common HBB genetic variants and direct DNA sequencing to detect the rare mutations of HBB gene. Heterozygous inheritance of the mutation results in severe beta-thalassemia phenotype. The proband was a 13-year-old boy when first studied. He was referred because of severe anemia. Hematological analysis of the reveals Hb 7.2 g / dl; with microcytosis of 71.1 fl, hypochromia 25 pg and the number of red blood cells is  $2.9 \ 106 \ / \ mm^3$ . In addition, a significantly secondary thrombocytosis and leukocytosis were reported in patient. Electrophoresis of hemoglobin in an alkaline medium shows Hb A2 = 4% HbF = 65% and blood smear confirms microcytosis hypochromia, and showing the presence of many dacryocyte with hyper eosinophilia.

The combination of these mutations Hb Knossos (*HBB*: c.82G>T) and (*HBB*: c.118C>T) causes the beta-thalassemia major phenotype, and this is important for genetic counseling.

Keywords: Beta-thalassemia, Hb Knossos, HBB gene, genetic counseling, Algeria

# Investigation of Genotoxic and Antigenotoxic Effect of Amygdalin Using Comet Assay on Isolated Lymphocytes

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#### ABSTRACT

Amygdalin is a cyanogenic glycoside, mainly present in the seeds of Rosacea family such as apricots, peaches, bitter almond, plum, and apple. Several studies have reported on the prevention or treatment of various diseases including cancers, migraine, chronic inflammation, hypertonia, asthma, bronchhitis, diabetes and vitiligo (1, 2). In this study, the genotoxic and antigenotoxic effect of Amygdalin against H<sub>2</sub>O<sub>2</sub> induced DNA damage using comet assay in human lymphocyte were investigated. For this purpose, the lymphocytes isolated from 3 healthy volunteer (2 females and 1 male) were incubated with different Amygdalin concentrations (0.86, 1.72, 3.43, 6.86 and 13.75 µg/mL) alone and simultaneously with H<sub>2</sub>O<sub>2</sub> (100 mM) at 37°C for 1 hour. A negative (distilled water) and a positive control (H<sub>2</sub>O<sub>2</sub>) were also run. 300 cells for each concentration were analysed with regards to comet parameters that are the tail length  $(\mu m)$ , tail intensity (%) and tail moment using Comet Assay IV (Perceptive Instruments Ltd., UK). Amygdalin alone did not significantly increase DNA damage at all concentrations compared to negative control. Amygdalin+H<sub>2</sub>O<sub>2</sub> treatments were statistically significant decreased DNA damage occurring by H<sub>2</sub>O<sub>2</sub> compared to positive control for all comet parameters. According to our results, Amygdalin exhibited chemopreventive activity H<sub>2</sub>O<sub>2</sub>-promoted DNA damage in human lymphocytes.

## ACKNOWLEDGEMENT

This study was supported by TUBITAK (Project Code:118Z574).

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# The Examination of the Genotoxic Effects of Vermicompost (Obtained from *Eisenia foetida*) Using to Allium Test in Tissue Culture

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#### ABSTRACT

Vermicompost is obtained of digesting various plant and animal organic wastes by worms into humus-like substances. It contains nitrate, phosphate, exchangeable calcium and soluble potassium which are necessary for plant growth (Joshi and Pal Vig, 2010). This study investigated possible genotoxic effects of vermicompost obtained from Red California Worm (*Eisenia foetida*) at plant tissue culture with *Allium* test. The *Allium cepa* bulbs were rooted on agar solidified Murashige and Skoog (MS) medium for 3 days. Thereafter, the bulbs were transferred to MS medium (control) or MS medium containing 5000, 10000, 20000, 40000 and 80000 µg/mL vermicompost for 24 hours. The result indicates that all concentrations of vermicompost did not significantly increased the mitotic abnormalities compared to the control group. The Mitotic Index (MI) was significantly decreased at 10000 and 40000 µg/mL and significantly increased at 80000 µg/mL concentrations of vermicompost. This study is a preliminary study and all evaluations were made on five roots. These preliminary results demonstrated that vermicompost may not genotoxic potential in *Allium cepa* at tissue culture conditions.

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# In Vitro Genotoxic and Antigenotoxic Effects of an Exopolysaccharide Isolated from Lactobacillus salivarius KC27L Strain Using Mitotic Index and Sister Chromatid Exchanges (SCEs) Test

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#### ABSTRACT

The aim of this study was to investigate the *in vitro* genotoxic and antigenotoxic effects of an exopolysaccharide (EPS) isolated from Lactobacillus salivarius KC27L strain using mitotic index (MI) and sister chromatid exchanges (SCEs) test in cultured human lymphocytes. Peripheral blood samples collected from two healthy, non-smoker volunteers (a male and a female) were treated with four different concentrations (12.5, 25, 50 and 100 µg/mL) of EPS alone and simultaneously with 0.2 µg/mL mitomycin-C (MMC) for 24 and 48 hours. A negative and a positive controls (MMC) were maintained for each treatment. EPS did not significantly increase the SCE frequency at all concentrations compared to control groups at both 24 h and 48 h. Simultaneous treatment of EPS and MMC significantly reduced the frequency of SCE in all concentrations compared to positive control in both treatment periods. In addition, EPS did not decrease the MI in all concentrations compared with the negative control at both 24 and 48 h periods (except 100 µg/mL). Simultaneous treatment of EPS and MMC increase the MI in all concentrations compared to positive control in 24 and 48 h treatments. However, the EPS or simultaneous treatment of MMC did not affect replication index. This study demonstrated that the exopolysaccharide (EPS) isolated from Lactobacillus salivarius KC27L strain has no genotoxic risk at these concentrations and may have antigenotoxic potential in human peripheral lymphocytes.

# Variations of Some Primary Metabolites under the Effect of Dust Emitted by the Cement Plant of Bouira (Algeria)

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## ABSTRACT

The objective of our work is a toxicological study of the dust emitted by Bouira cement plant (Algeria) on a cryptogamic species: a lichen (*Xanthoria parietina*). Our samples collected at the "Errich" forest, an area considered to be unpolluted, did undergo a series of laboratory contamination tests during one month of treatment.

The toxicity of this dust is investigated through morphological observation and the determination of biochemical and physiological parameters. These tests showed that lichens show biochemical (proline) and physiological (Chlorophylls a, b and a+b and the ratio Fresh Matter/Dry Matter) changes.

The results obtained highlighted the impact of the dust emitted by the Bouira cement plant on our bio indicator used.

Keywords: lichens, dust, air pollution, toxicity, cement plant.

# Determination of Genotoxicity of Xylitol, A Sweetener, by Comet Assay in Isolated Human Lymphocytes

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#### ABSTRACT

Sweeteners are food additives that provide a sweet taste like that of sugar while containing significantly less food energy. There are positive and negative opinions about their effects on human health. Xylitol is a naturally occurring sugar alcohol, which has a wide variety of applications in food industry. It is present naturally in small amounts in fruits and vegetables, algae, and mushrooms (1). In this study, comet assay was conducted to evaluate the potential genotoxic effects of xylitol in isolated human lymphocytes. For this purpose, peripheral venous blood was collected from healthy nonsmoking adults; one male and two females, aged 27 years. Various concentrations of xylitol (125, 250, 500, and 1000  $\mu$ g/mL) were applied to isolated lymphocytes at 37°C for one hour. A negative and a positive control (100  $\mu$ M H<sub>2</sub>O<sub>2</sub>) were also applied for each treatment. According to test results, xylitol increased the comet tail intensity, tail length, and tail moment at two highest concentrations in a dose-dependent manner. In our previous studies, xylitol induced chromosomal aberrations, sister chromatid exchanges and micronucleus formation especially at high concentrations.

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# Effects of Water Stress on Gonads in Desert Rodent Gerbillus Tarabuli

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## ABSTRACT

The aim of this study is to investigate the possible effects of hydration by a water-rich diet on body weight, on male and female reproductive activity in desert rodent Gerbillus tarabuli. This is achieved by a prolonged hydration of three weeks to study the effect of hydration on body weight, while a hydration of five weeks to know the effect of hydration on ovarian and testicular histological level, the food of the non-hydrated groups is close to their biotope and differs from the hydrated groups by the addition of "ad libitum". The results show that there is a less significant increase in body weight, possibly due to growth, the amount of stored fat and low muscle activity. The histological study reveals structural changes observed in interstitial level, in testicular capsule, and also in the cortex and ovarian medulla. In conclusion, the data obtained suggest that the hydrated diet has a state of alert on reproduction in gerbillus tarabili.

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# Achievement of the Grain Yield of Some Local and Improved Durum Wheat Varieties in Relation to Its Components and the Duration of the Vegetative Phase under Semi-Arid Conditions of Algeria

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#### ABSTRACT

In Algeria, durum wheat is sown on an average extent of 1.2 million hectares, and its production represent for 45% of total cereal production (Fellahi *et al.*, 2013), which puts it at the forefront of cultivated cereals. Most of the durum wheat area is concentrated on the semi-arid high plains, region characterized by an altitude of more than 700 meters, cold winters, low and erratic rainfall, associated with warm and drying winds at the end of the cycle of culture (Chennafi *et al.*, 2006). These characteristics constitute the main constraints of the weakness of the production in these regions. The characterization of varieties of hard wheat grown there is one of the ways to benefit from it in varietal selection. This characterization makes it possible to determine the criteria associated with the grain yield in the constrained regions and the varieties concerned.

Sixteen durum wheat varieties, local and improved, were tested under a semi-arid environment in Algeria during the 2017/2018 crop year. The results obtained indicate that the number of spikes per unit area is positively correlated with grain yield (r = 0.825) and the duration of the vegetative phase is negatively correlated (r = -0.584). In this same order, the coefficient of determination ( $R^2$ ) of the linear regression takes the values of 0.656 and -0.410, respectively. It was then that under the semi-arid conditions of this study, grain yield is positively related to the number of spikes per unit area, in this case per square meter, and to short vegetative phase duration (at the earliest heading). The Vitron variety aligns these two traits, with respective values of 270.67 spikes /  $m^2$  and 117 days from January 1st, for the second largest grain yield at 347.04 g /  $m^2$ . Possibly showing itself as a breeding variety to favor in varietal selection to improve yields in semi-arid regions.

**Keywords:** Durum wheat, local varieties, improved varieties, grain yield, components of yield, date of heading.

# Assessment of the Apoptotic Effect of Bioinsecticide Pyrethrum Extract in Human Hepatocellular Carcinoma HepG2 Cells

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#### ABSTRACT

Nowadays, due to the many adverse side effects of synthetic pesticides on human health and the environment, the demand for natural pesticides has increased [1]. Pyrethrum extract (PE), a natural bioinsecticide derived from the flowers of Chrysanthemum cinerariaefolium, is extensively used across the world [2]. This natural pyrethrin has excellent insecticidal properties to control pest insects in household and farms [3, 4]. Although this bioinsecticide is of natural origin, the possible toxic and apoptotic effects should be investigated. In literatüre, there are no data on apoptotic effects of PE in human hepatocellular carcinoma (HepG2) cells. The aim of this study was to evaluate the apoptotic effect of PE on human hepatocellular carcinoma HepG2 cells using Annexin V-FITC/ Propidium iodide (PI) assay. Briefly,  $2x10^{5}$ /mL (per well) HepG2 cells were grown in 6 well culture plate and treated with 1000, 2000 and 4000 µg/mL concentrations of PE for 24 and 48 hours. At the end of the treatment, cells were stained with Annexin V-FITC and PI and analyzed with flow cytometry (Beckman Coulter, Navios). Cells were classified in stages of live cells (FITC- PI-), early apoptosis (FITC+ PI-), necrotic cell (FITC- PI+) and late apoptosis/necrosis (FITC+ PI+) with the Kaluza software program. At early stages of apoptosis, the cells bind Annexin V but exclude PI. At later stages the cells show both, Annexin V binding and PI fluorescence [5]. As a result, there was a significant reduction in the percentage of live cells at all treatments. Additionally, the percentage of necrotic (FITC-PI+) cells were significanly increased in all concentrations and treatments compare to control. However the percentage of late apoptotic/necrotic (FITC+ PI+) [cell population for 24 and 48 hour, 14.47%, 48.60%, respectively] cells were significantly higher increase only at 4000 µg/mL PE in both treatment times.

## ACKNOWLEDGEMENT

This work was supported by Gazi University Research Fund (The Scientific Research Projects Coordination Unit) under Project number: 64/2018-06, Turkey.

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# Anti-inflammatory and Antioxidant Activities of the Aqueous Extract of Capparis spinosa Buds

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#### ABSTRACT

Flower buds of *Capparis spinosa* are consumed as food and used in traditional medicine for treatment of various diseases including inflammatory and oxidative stress disorders [1]. Thus, this study aimed to explore the ant-inflammatory potential effect and to assess the antioxidant efficacy of the aqueous extract of this part of plant. Firstly, the content of the extract in total phenolic and flavonoid was assessed by colorimetric methods. The antiinflammatory activity was determined in vivo by evaluating the anti-edematous and the antinociceptive effects of the extract by using croton oil-induced ear edema and acetic acidinduced writhing assay, respectively. In vitro, the antioxidant properties were investigated by using 1,1-diphenyl -2-picrylhydrazyl (DPPH) and reducing power tests. Results showed that the aqueous extract of Capparis spinosa buds contents an important amount of polyphenols and flavonoids (90.04  $\pm$  6.26 µg/mg gallic acid equivalent and 11.81  $\pm$  1.18 µg/mg quercetin equivalent, respectively). Furthermore, the topical application of the extract (1 mg/ear) was able to reduce ear edema with an inhibitory effect of 66%. Similarly, the oral treatment of mice with 200 and 400 mg/kg of the extract inhibited the writhing response by 67.69% and 70.90%, respectively. On another hand, the extract exhibits a very important antioxidant activity, since it scavenge DPPH radicals and reduce ferrous ions with IC<sub>50</sub> of 226.97  $\pm$  7.11 µg/ml and 199.28  $\pm$  11.84 µg/ml, respectively. Taken together, these results revealed that *Capparis spinosa* buds extract possess a good anti-inflammatory and antioxidant activities which may be related to its content of phenolic compounds and confirm the nutritional and medicinal value of this part of plant.

#### ACKNOWLEDGEMENT

This work was supported by the Ministry of Higher Education and Scientific Research.

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# Control of *Trialeurodes vaporariorum* Westw. in Tomatoes in the Greenhouse

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#### ABSTRACT

Trialeurodes vaporariorum Westw. (Homoptera: Aleyrodidae) is a cosmopolitan and polyphagous species that is one of the most important pests of vegetable crops and ornamental plants in greenhouses and greenhouses the direct damage, it caused by sucking on vegetable juices and creating the conditions for the development of saprophytic fungi by secreting honeydew. Greenhouse whitefly is also significant as a vector of tomato virus [1]. Due to the presence of this pest, the yield of tomatoes can be reduced by 40%. These trials were conducted in 2019 by standard EPPO methods [2, 3, 4] at locality Bačko Gradište (Vojvodina) in tomatoes (cultivar Summer sun) in the greenhouse in accordance with Commission Regulation (EU) 2018/783, which prohibits the use of imidacloprid in the field due to the risk to pollinators, including only authorized use in greenhouse. The pesticide preparations based on imidacloprid (200 g a.i./l, SL formulation) were applied at a amount of 0.5 and 0.6 l/ha and acetamipride (200 g a.i./l, SP formulation) at a amonut 0.25 kg/ha. Pesticides were foliar applied, using a backpack sprayer with water consumption of 400 l/ha when tomatoes were in the flowering stage BBCH62. The efficiency of the insecticide was calculated by Henderson & Tilton and the significance of differences was determined by ANOVA for a confidence interval of 95%. Before the experiment, the average number of adult amounted from the 7.75-11.2, while the number of larvae was 19.8-22.8. Three days after the application of the insecticide, the size of T. vaporariorum was substantially lower in comparison to the control variant, and the efficacy ranged from 82.9 to 94.1%. After seven days from application, the number of aphids, adults and larvae, was also at a significantly lower level compared to the control with an efficacy of 95.8-98.2. The population of T. vaporariorum in the tomato crop in the greenhouses at Backo Gradište showed high susceptibility to insecticides from the neonicotinoid group, but there is also a need to include them in the monitoring of insecticide susceptibility since these are pests that are capable of quickly develop resistance.

## ACKNOWLEDEMENT

This study was part of the project III46008, funded by the Ministry of Education, Science and Technological Development of the Republic of Serbia.

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# Effects of Whole Oat, Oat Bran and Refined Oat Incorporation in a High-Fat Diet on the Cardio-Metabolic Risk Biomarkers in Rats with Type 2 Diabetes

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## ABSTRACT

The purpose of this study was to compare the effects of whole oat, oat bran and refined oat incorporation in a high-fat diet (HFD) on the cardio-metabolic risk biomarkers in rats with type 2 diabetes (T2D). T2D was induced by feeding young male rats (n = 24 and weighing  $160 \pm 10$  g) with an HFD for 10 weeks, followed by a low dose (35 mg/kg of body weight (BW)) of streptozotocin (STZ). T2D rats were then divided into four homogeneous groups. Three groups consumed an HFD containing 45% of whole oat (T2D-WO), oat bran (T2D-OB) or refined oat (T2D-RO). The fourth untreated group (T2D) continues to receive the HFD and serves as a control. After 28 days of nutritional experiment, the BW was similar in all four groups of rats. In contrast, a significant reduction in food and calories intake, leptinemia, arterial blood pressure (diastolic and systolic), glycemia, insulinemia, glycosylated haemoglobin, and insulin resistance (HOMA-IR) was noted in groups consuming whole oat or oat bran compared with the group fed refined oat and T2D untreated group. In addition, some parameters were even higher in T2D-RO vs T2D group (calories intake (+12%), insulinemia (+43%) and HOMA-IR (+42%)). Incorporation of whole oat and oat bran into HFD, improved serum and hepatic lipid levels as well as the reverse cholesterol transport by reducing, the values of total cholesterol (TC), triacylglycerols (TG), very-low (VLDL) and low-density lipoproteins cholesterol (p < p0.05), and by increasing lipids and cholesterol faecal excretion and high-density lipoprotein cholesteryl ester concentrations (p < 0.05) and stimulating lecithin: cholesterol acyltransferase activity. Also, T2D-WO and T2D-OB groups reduced the atherogenic index (TC/ HDL<sub>2+3</sub>-C, VLDL-LDL-HDL<sub>1</sub>/ HDL<sub>2+3</sub>-C)compared to those of T2D and T2D-RO groups (p < 0.05) and are even lower in the T2D-OB group vs T2D-WO (1.4- and 1.8-fold). In conclusion, in T2D rats, the incorporation of whole oat and its bran into a fat-enriched diet improves arterial blood pressure, provides better glycemic control and more efficient cholesterol and triacylglycerols transport. In contrast, refined oat accentuates the risk factors associated with diabetes.

**ACKNOWLEDGEMENT:** This work was supported by the Algerian Ministry of Higher Education and Scientific Research.

# Hypotensive and Antioxidant Effects of Algerian *Opuntia ficusindica* (*OFI*) Nopalitos in Rats with Type 2 Diabetes

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#### ABSTRACT

The effect of *Opuntia ficusindica* (*OFI*) nopalitos supplementation in a high-fat diet (HFD) was studied on blood pressure and pro/antioxidant balance, in Wistar rats with type 2 diabetes (T2D).Diabetes was induced by HFD ingestion containing 30% sheep fat for 10 weeks, followed by injection of a low dose of streptozotocin (35 mg/kg body weight).The T2D rats were then divided into 2 homogeneous groups and consumed each for 30 days, the HFD supplemented with freeze-dried *OFI* nopalitos at a rate of 50 g.kg<sup>-1</sup> of diet (HFD-*OFI* group) or not supplemented (HFD group).

After 1 month of nutritional experimentation, *OFI* nopalitos supplementation decreased arterial diastolic (-20%) and systolic (-16%) pressures. Evaluation of lipid peroxidation showed a reduction of thiobarbituric acid reactive substances (TBARS) in VLDL (-20%), LDL-HDL<sub>1</sub> (-23%), HDL<sub>3</sub> (-35%) and erythrocytes (-37%) in the HFD-*OFI* group compared with HFD. Similarly, in tissue, hydroperoxides and TBARS levels were decreased respectively in the liver (-20%, -26%), heart (39%, -48%), muscle (-25%, -29%) and adipose tissue (-25%, -31%). Protein oxidation evaluation revealed that *OFI* nopalitos decreased carbonyl levels in serum (-32%), erythrocytes (-38%) and some tissues (muscle -44% and kidney -27%) levels.In treated T2D group, the reduction of prooxidant markers was concomitant with increased activities of serum antioxidant enzymes (glutathione peroxidase (GSH-Px) and catalase (CAT)) (p<0.05); of erythrocytes (superoxide dismutase (SOD) and CAT) and tissues (SOD in aorta, muscle and adipose tissue); GSH-Px (liver, heart, muscle, kidney and adipose tissue) (p < 0.05); and CAT only in the liver and heart (p < 0.05).

In conclusion, in rats with T2D and exposed to a diet enriched in lipids, OFI nopalitos, seems to have interesting nutritional, functional and therapeutic properties (hypotensive and antioxidant), on account to their richness in soluble fiber and natural antioxidants (flavonoids and polyphenols). The nopalitos of Opuntiaficusindica cactus offers a better alternative to vegetable products a therapeutic, preventive, economic perspective and sustainable development.

**ACKNOWLEDGEMENT:** This work was supported by the Algerian Ministry of Higher Education and Scientific Research

# Effect of Repeated Administration of Hcg on Ovarian Response in PMSG-Superovulated Ouled Djellal Ewes (Algeria)

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## ABSTRACT

The objective of this study was to evaluate the effect of repeated administration of hCG on ovarian response in PMSG-superovulated ewes. Intravaginal pessaries containing 40 mg fluorogestone acetate (FGA) were inserted in all ewes (n=9) and remained in situ for 14 days. Two days prior to pessary removal, all ewes were treated with 1000 IU of PMSG. On the day of sponge removal (day 0), the females were randomly assigned to 2 treatments. The first group (n=3) did not receive any hCG, while the second group (n=6) treated intermuscular with hCG (500 IU) during days 0-2. On day 8, laparotomy was performed to assess numbers of corpora lutea (CL) and anovulatory follicles (AF). Blood samples were collected for analysis of serum progesterone (P4) using radioimmunoassay (RIA) method. The results obtained for first and second group was in number of CL ( $6.33\pm1.15$  and  $10.50\pm5.54$ ), number of AF ( $2\pm3.46$  and  $4.16\pm5.70$ ), then the levels of P4 ( $5.75\pm4.45$  and  $13.22\pm6.80$  ng/ml), respectively. These results indicate that the repeated administration of hCG post-sponge removal increases number of CL and improves luteal function in ewes after PMSGsuperovulatory treatment.

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# First Screening of Streptococcus Agalactiae in Pregnant Women in Specialized Mother and Child Hospital - Meriem Bouatoura - Batna, Algeria

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# ABSTRACT

Streptococcus Agalactiae can cause severe pneumonia, sepsis and meningitis in neonates and causes one of the most prevalent causes of invasive neonatal infections. Prenatal screening and perinatal antibiotic prophylaxis can prevent maternal transmission of S. Agalactiae during delivery. The objective of this study is to determine the maternal risk of maternal carriage of group B streptococcus over time, to offer reliable epidemiological data to the health staff working at maternity Meriem Bouatoura Batna or even all maternity hospitals in Algeria. than to inform them of the real risk of carriage of group B streptococcus correlating with the literature. In this prospective study, a vaginal swab (lower third) followed by a rectal swab that substantially improves the sensitivity of screening. Two methods of confirming the identification of SGB:

- Agglutination using the PASTOREX® Strep Kit.

- VITEK automated methods identification 2.

A total of 50 patients received a sample in 34 weeks of amenorrhea. A total of 5 patients were GBS positive, a percentage rate of 10%. Regular vaginal culture in pregnant third trimester screening be done, with evaluation of risk factors and treatment will reduce risk of infection in infants is regarded.

Keywords: Streptococcus Agalactiae, Pregnant Women, Batna, Algeria

# Accumulation of Nitrite and Nitrate and pb in the Spinacia oleracea in Iran, Markazi Province

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#### ABSTRACT

As the population grows, the demand for food and so on the use of chemical and organic fertilizers to achieve more production has increased. Therefore it is necessary to ensure the health of this kind of valuable food. Excess consumption of nitrogen fertilizers has caused a high nitrate uptake by the plant. On the other hand, heavy metals due to their nondegradability and physiological effects on humans are essential. These elements, due to low mobility, accumulates in the soil and eventually enters the food cycle and effects on human health. Therefore continuous measurement and monitoring of Heavy metals concentration are essential. Leafy vegetables are rich in vitamins, minerals, and antioxidant compounds that have been proven to have anti-cancer properties as well as reduce cardiovascular disease. Vegetables are the most important source of exposure and contribute to more than 80% of the nitrate intake. Since vegetables are capable of absorbing and retaining large amounts of Pb, nitrite, and nitrate, the consumption of such products may endanger human health. Spinach is a herb that belongs to the family Chenopodiaceae. Spinach is widely distributed and cultivated in Southwest Asia, including Iran. Spinach is a good source of minerals like iron, copper, phosphorus, zinc, selenium, niacin, folic acid, ascorbic acid, carotenoids, phenols, apocynin, and Omega-3-fatty acids. It is a relatively fast-growing vegetable. Nitrates accumulated in vegetables during a series of chemical reactions in the human gastrointestinal tract convert to nitrite and nitrose acid, and in combination with the first and second amino acids, cause the formation of nitrosamine, which causes a different kind of cancers like stomach, intestine, mouth, and hemoglobinemia in children. Heavy metals concentration is one of the most critical factors in determining the health of vegetables. Vegetable contamination with heavy metals can be due to different factors like fertilizers, pesticides, and wastewaters. Because of the importance of Pb, nitrite, and nitrate accumulation in leafy vegetables, this study investigated this issue. Twenty-five samples from different areas of Markazi province were collected. The amount of Nitrite concentration was determined in samples by spectrophotometric methods at a wavelength of 538 nm, and nitrate concentration was determined after reducing nitrate to nitrite by using cadmium column. ICP-OES determined concentrations of Pb and it was estimated three times from each sample of Spinach. The changes were calculated by one way Anova, and for analysis of the role of multiple factors, univariate analysis was used by SPSS 21. Probability values of <0.05 were considered significant. The results show a high amount of nitrite and nitrate in some samples; nevertheless, in most samples, the amounts of it were

lower than the maximum standard limite of nitrite and nitrate in leafy vegetables that set by WHO. The amount of Pb in the three samples was higher than the standards of leafy vegetables that set by WHO. For the rest of the samples, the amount of Pb was lower than 0.05 mg/kg. It should be noted that the process of spinach preparation for consumption affects the amount of Pb, nitrite, and nitrate, and this issue needs further studies.

Keywords: Pb, Nitrite, Nitrate, Spinacia oleracea, spectrophotometer, ICP-OES.

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# Epidemiological Study of non-Hodgkin's Large Cell B Malignant Lymphoma

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## ABSTRACT

Large B-cell non-Hodgkin's lymphoma results from the uncontrolled multiplication of abnormal B-cells, the very large cells that compose it will be dispersed in the ganglion or the lymphatic tissue. It accounts for 30 to 40% of all cases of LMNH. To date, the etiology of the disease is unknown but many viral or bacterial infections are criminalized, as well as other environmental factors.

Our epidemiological study was conducted at the University Hospital Center (CHU) of Batna (Algeria) on 33 patients with LDGCB (11 women and 22 men.) This descriptive analysis of the epidemiological factors associated with LDGCB allowed us to obtain interesting results;

There are several epidemiological factors associated with this type of lymphoma:

The age group [49-59] years is the most affected by LDGCB (24.24%) with an average age of 54 years and with a male predominance 67%; the sex ratio H / F 2. Our results also show that the high incidence of LDGCB cases was recorded in stage IV patients (33.33%). Biologically (60.60%) patients had normal HB levels of 6 to 12 g / 1 and the majority of patients had a normal blood platelet count (45.45%). The markers (CD20, CD45, CD3) are expressed in the great majority of cases (90.90%, 39.39%, 21.21%, respectively).

In the light of this research, we suggest applying molecular biology methods to the diagnosis of NHNH by studying the clonality of a lymphoid population using Southern-Blot and PCR techniques.

**Keywords:** Large B-cell non-Hodgkin's lymphoma (LDGCB), epidemiological study, clinical parameters, biological analysis, University Hospital Center of Batna.

# Antioxidant and DNA Damage Protective Activities of Methanol and Aqueous Extracts of Algerian *Cistus salvifolius*

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#### ABSTRACT

Cistus species are wide spread in north western Africa, used as general remedies in folk medicine for treatment of various skin diseases and inflammation disorders related to oxidative stress. This study was aimed to evaluate antioxidant activity of methanolic and aqueous leef extracts of *Cistus salvifolius*, sampled from Setif, Algeria. Total phenolic content was determined in both extracts. The antioxidant capacity of the two extracts was pointed out by ABTS, DPPH, FRAP and TBARS methods. Free radical (DPPH• andABTS•) scavenging potential revealed that both extracts to be active radical scavengers. Reducing (Fe+3-Fe+2) power and lipid peroxidation inhibition efficiency (TBARS assay) of the extracts was also evaluated. Results showed that aqueous and methanolic extracts inhibited linoleic acid oxidation by 76 and 79% respectively and exhibited a high reducing power, suggesting that extracts had strong electron-donating capacity. Furthermore, the ability of the extract to protect plasmid DNA (pBluescript M13+) against UV-induced photolysed H2O2 - oxidative damage was investigated. Methanolic and aqueous extracts was able to protect DNA from oxidative damage by 78-98%. The results obtained suggest that methanolic and aqueous extracts of Cistus salvifolius has promising therapeutic potential and may be useful in the food industry. They could be considered as potential source of effective synthetic antioxidants or for drug development by pharmaceutical industries.

Keywords: Cistus salvifolius, DNA damage, free radical, lipid peroxidation, Phenolics.

## ACKNOWLEDGEMENTS

The authors are thankful to the Algerian Ministry of High Education for providing facilities to carry out the research work.

# Insecticidal Effects of Asphodel (Asphodelus microcarpus) and Calycotome (*Calycotme spinosa*)

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# ABSTRACT

This study aims to propose alternative solutions to plant protection products based on biopesticides derived from vegetable extracts. The choice is based on two plants, roots and flowers of the asphodel (*Asphodelus microcarpus*) and roots and flowers of calycotome (*Calycotome spinosa*). The plants are harvested from different regions in Bouira. After drying, the plants are ground and then subjected to aqueous extraction. The insecticidal effect is evaluated against 3 species of insects (*aphis fabae*, *Ephestia kuihinella* and *Tribolium castaneum*) at concentrations of 100% 50% 25% 12.50%. Treatment by contact with adults of these insects at different doses reveals the efficacy of these extracts. Indeed, after 72 hours of the treatment on insects, one obtains 100% mortality in *A. fabae* for the 4 extracts and with the 4 doses. In the case of *E. kuihinella* treatments, we have obtained mortalities that vary from one extract to another, between 30% and 100% for high doses, and 13% to 92% for low doses. Adults of *T. castaneum*, treatments show mortalities ranging from 34% to 100% depending on dosages and extracts. The results show that mortality reported using the extract of *A. microcarpus*.

# ACKNOWLEDGEMENT

This work was done with the help of the plant protection laboratory of the SNVST faculty of the University of Bouira.

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# Effect of Phytosanitary Treatments on the Content and Biological Activity of 'Golden delicious' Apple Secondary Metabolites.

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# ABSTRACT

Polyphenols are major compounds of plants, possessing various modes of action and inhibitory and lethal activities with respect to a large number of microorganisms. In addition, secondary metabolites, including phenolic compounds, contain substances that are highly sought after by industries.

Apples are known for their richness in phenolic compounds, the composition of which depends on various factors such as cultivar, stage of maturation, agricultural practices, environmental factors and apple crop region.

The aim of this work is to study the effect of phytosanitary treatments on the content and biological activity of total polyphenols and flavonoids extracted from *Golden delicious* fruit peels harvested from two orchards that have undergone phytosanitary treatments at different levels intensities, situated in northern Algeria. The extraction of the polyphenols is carried out from an apple powder obtained by drying the peels in a ventilated oven at a temperature of 50°c, followed by grinding and sieving. The extraction of total polyphenols (PPT) was done by the maceration method in aqueous methanol, while flavonoids (Flv) are extracted by clashing with petroleum ether. After the detection of PPT and flavonoids in the methanolic solutions, their determination is carried out by the colorimetric method Folin –Ciocalteu and Aluminum trichloride, respectively.

The antimicrobial activity of the phenolic extracts is evaluated according to the solid medium diffusion method. The microbial range used for this test is composed of 3 bacteria, 2 filamentous fungi and a yeast namely: *Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, Aspergillus niger, Penicillium digitatum* and *Candida albicans*. The results show that PPT and flavonoid contents are higher in apple peels obtained from the least treated orchard with PPT levels = 0.183 mgEAG / mg MS and Flv = 1.531 mgEQ / mg MS versus of PPT = 0.075 mgEAG / mg MS and Flv = 0.045 mgEQ / mg MS of the orchard intensively treated. *E. coli* is highly resistant to all the extracts, whereas *C. albicans and P. digitatum* are highly sensitive to the PPTs from peel apple of the two orchards. the statistical analysis of the orchard effect showed a very highly significant difference.

# ACKNOWLEDGEMENT

The authors would like to thank laboratory for research on organic chemistry of Blida 1 University for critical analysis.

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# Validation of the Method for the Analysis of Fungicide Dodine Residues in Sour Cherry Fruits

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# ABSTRACT

Blumeriella jaapii (Rehm) Arx., causing agent of sour cherry leaf spot, represents widely spread and a significant problem in sour cherry production. It is present in all regions where the sour cherry is grown. In the control of B. jaapii (Rhem) Arx. in this fruit species, the most effective and indispensable is chemical control. Dodine is one of the fungicides that is highly effective in protecting cherries from this pathogen. The aim of this study was validation of the method for the determination of fungicide dodine in sour cherry fruits. The analyses was carried out by gas chromatography (Shimadzu GC-2010, Japan), equipped with a <sup>63</sup>Ni electron-capture detector and an InterCap capillary column (0.25 µm film thickness, 0.32 mm i.d.  $\times$  30 m). Injector was operated in splitless mode at 250 °C. Oven temperature was programmed as follows: the initial temperature of 150 °C was increased at 20 °C/min to 250 °C and was held for 6.0 min. The ECD temperature was set at 300 °C. A volume of 1 µl of was injected into the GC system. The extraction was conducted through QuEChERS procedure. The proposed method was validated in terms of accuracy, linearity, precision, limits of detection and quantification. The obtained limit of detection (LOD) for dodine fungicide was 0.016 mg/kg, while limit of quantification (LOQ) was 0.05 mg/kg, lower than prescribed MRL of 3 mg/kg. The obtained data demonstrated the good reproducibility in concentration of 0.5 µg/ml, with relative standard deviation (RSD) of 0.55%. Recoveries for spiked sour cherry samples at three concentraction levels (0.05 mg/kg, 0.5 mg/kg and 3 mg/kg) were from 93.6% to 101.2%. Matrix effect was 138,5%, thus for the analysis of dodine fungicide residues in sour cherry samples matrix-match calibration has to be applied. The results obtained for all parameters are completely in accordance with SANTE/11813/2017 criteria [1], and the proposed methodology is applicable for the detection and monitoring of fungicide dodine in sour cherry fruits.

# ACKNOWLEDGEMENT

This work was supported by Ministry of Education, Science and Technological development of the Republic of Serbia, No. TR31038.

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# Etude Comparative de Quelque Composes Entre Quelque Variétés de Dattes Algérienne et Saoudienne

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#### ABSTRACT

Dates are the date palm fruit phoenix dactylifera commonly called "nakhla". It has many therapeutic virtues that require further study. Indeed, phenolic compounds are secondary metabolites of great pharmacological importance, given their various biological activities confirmed in a large number of studies. It is in this context that we conducted a study that had as objective, on the one hand, the determination of certain phenolic compounds, flavonoids and condensed tannins in the methanolic extracts of six Algerian date varieties and five Saudi varieties using the methods of Folin-Ciocalteu, aluminum trichloride and vanillin respectively and secondly to compare the Algerian and Saudi varieties at the level of sugars and some micronutrients. Quantitative determination of total phenolics yielded levels ranging from 51.66 to 528.33 EAG / 100 g extract for Algerian varieties and from 51.66 to 205 EAG / 100 g extract. That of flavonoids varies from 12.5 to 37 EQ / 100 g of extract for Algerian varieties and from 3.95 to 23.95 EC / 100 g of extract for the Algerian varieties and from 8.56 to 16.59 EC / 100 g of extract for the saudi varieties. Sugars are the most important constituents of the date, their content is determined by Brix degree.

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# Evaluation of Salt Tolerance of a Transgenic *Nicotinia tabacum* with the UGT74E2 Gene

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# ABSTRACT

In Arabidopsis thaliana, overexpression of the UDP-glucosyltransferase UGT74E2 resulted in an altered homeostasis which was linked with architectural changes and resulted in drought and salt stress tolerance. Tobacco was transformed with Agrobacterium tumefaciens with p35S::UGT74E2 + p35S::NPTII. Kanamycin-resistant plants were selected for rooting on hormone free MS medium with 50 mg/l kanamycin and subsequently micropropagated. Small shoots were transferred to test tubes with 0, 6, 9, 12 and 15 g/l NaCl in order to assess the root formation. Only at 15 g/l it was possible to differentiate between control and transgenic plants, almost half of seawater.

Keywords: Tobacco, Genetic Transformation, Salt Stress.

#### ACKNOWLEDGEMENT

This work was supported by the Laboratory for Applied *In Vitro* Plant Biotechnology, Faculty of Bioscience Engineering, Ghent University.

# QQPFP; The Main Allergic Linear Peptidic Epitope Derived from Wheat Gliadin: A Review

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#### ABSTRACT

QQPFP (glutamine-glutamin-proline-phenylalanine-proline) is an amino acid sequence or potential celiac-toxic repetitive penta peptide epitope. There is no direct relationship between the estimated gluten content and the presence of this epitope. This allergic linear peptidic epitope degraded by crude proteases from germinated wheat seeds (PGW). Other idea is the proteolysis by microbial proteases from Lactic acid bacteria (LAB). The other bacteria is *Rothia mucilaginosa* that cleaves gliadin at regions that are resistant to degradation by mammalian enzymes. A monoclonal antibody that recognizes a potential coeliac-toxic repetitive pentapeptide epitope in gliadins is "R5". The R5 antibody recognizes mainly the epitope QQPFP and homologous epitopes LQPFP, QLPYP, QQSFP, QQTFP, PQPFPF, QQPYP, and PQPFP to a weaker degree and one of the modified method detection quantification of celiac disease related gluten proteins from wheat is RIDASCREEN®Gliadin competitive ELISA as proposed by the recent Codex Alimentarius Draft Revised Standard.

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# Study of the Morphological Characters of Larvae Oestrus ovis LINNAEUS, 1761 (Insecta, Oestridae) and Adult Breeding (fly) in the Region of Constantine-Algeria

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# ABSTRACT

Sheep estrosis is a disease that causes serious economic losses in small ruminant livestock especially in sheep, stunting, and nervous symptoms called "false tournaments", it is also a disease that we can find in humans, it is wide spread in the world, particularly in pastoralist areas. Our study is divided into two parts, the first one consists in rearing larvae III and we obtained three Oestrus ovis flies, we then studied in the second part, the morphological characteristics of larvae L1, L2 and L3 using a binocular loupe. The L1 measures 2.5mm in length and 0.5mm wide weighing 0.0120 g, its cephalic segment has 2 bull-shaped hooks. The L2 weighed (0.2568  $\pm$  0.27) g and measured (12  $\pm$  7) mm long and (2.8  $\pm$  1.4) mm wide, the cephalic segment had less robust and more curved horns, and finally the L3 had a weight of (0.3813  $\pm$  1.5) g, a length of (20  $\pm$  6) mm and a width of (4  $\pm$  2) mm the pair of hooks will reappear with a larger size, dorsally there are large plates in the form of "parallelograms" to rounded edges except the cephalic, at the last segment we find a pair of stigmatic plates in the form of D. Our results are consistent with those of Giannetto et al 1999, as well as the results of Guitton et al, 1997,

KEY WORDS: Oestrus ovis, larvae, flies, breeding, morphological characters

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# Optimization of Mature Embryo Based regeneration of Mirzabey 2000 Cultivar (*Triticum turgidum durum* L.)

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#### Abstract

The objective of this study was to optimize tissue culture and regeneration parameters of mature embryo based tissue culture of Triticum turgidum durum cv. Mirzabey 2000. The effects of auxin type of hormone at different concentrations and dark incubation periods on capacity regeneration were evaluated. Two different hormone types 2.4dichlorophenoxyacetic acid (2,4-D) and picloram were used at three different concentrations 2, 4 and 8 mg/L. Mature embryo derived calli were incubated in 6 different induction media at dark for 4 and 6 weeks for initiation of primary callus induction. After dark incubation periods, average callus fresh weight and primary callus induction rate were determined. The primary callus induction rates for 4 weeks and 6 weeks old dark incubated Mirzabey calli were found to be 91 % and 93.25 % respectively. The primary calli were transferred to embryogenic callus induction medium. The embryogenic callus formation was 94.88 % in 6 weeks dark incubated and including 2 mg/L 2,4-D medium for Mirzabey cultivar. The necrosis was observed at high concentration of 2,4-D. After embryogenic callus induction, embryogenic calli were transferred into hormone free regeneration medium. The maximum regeneration rate (62.31 %) and culture efficiency (44.13 %) were observed in 4 weeks dark incubated and including 2 mg/L 2,4-D medium for Mirzabey.

Keywords: Regeneration, mature embryo, wheat, 2,4- dichlorophenoxyacetic acid.

# Introduction

Wheat which is an annual plant is one of the most important crop plant produced, traded and consumed in the world. It is thought that the cultivation of wheat reaches far back into history. Wheat has been a major food source in the human diet for 8000 years in Europe, West Asia and North Africa. According to Food and Agriculture Organization of United Nations, wheat was grown over 200 million hectares (nearly 17 % of the world's cultivable land) and produced nearly 690 million tons in 2008. From 8000 years ago to today, wheat continues its important role as a food source.

Wheat evolved from wild grasses found growing in the Eastern Mediterranean and the Near East and Middle East areas and in places where other similar cereal crops such as barley and rye possibly developed [1]. Wheat is a member of Angiosperm class, the monocot subclass and grass family. Within grass family, wheat is a member of the tribe Triticeae and genus *Triticum* [2]

The basic chromosome number of wheat species is seven. Thus, diploid wheat species have 14 chromosomes (2N=2X=14), the tetraploid emmer and modern durum wheat species have 28 chromosomes (2N=4X=28) and the common hexaploid wheat species have 42 chromosomes (2N=6X=42) [2]. *Triticum durum*, durum wheat genome size is nearly 10 billion base pairs. This genetic code is located into 28 chromosomes being diploid AABB. The structure of A- and B- genome chromosomes of durum wheat is essentially identical to the corresponding homologues of bread wheat [3]. *Triticum aestivum*, bread wheat has a genome size of 16 billion base pairs of DNA organized into 21 pairs of chromosomes, seven pairs belonging to each of the genomes A, B, and D [3-5]

The wheat kernel composition varies widely. For example, usual range of protein is from 8% to 15%, however 7% or as high as 24% can occur. While 26% protein can be detected in embryo, 24% protein can be detected in aleurone. Gluten, a water insoluble protein fraction, can be isolated from endosperm. Gluten is especially important for the leavening in bread making [6].

Biotechnological approaches are considered the latest tools for agricultural researches. Besides plant breeding applications, biotechnology focuses on to development of novel methods for genetically alteration and control of plant development, performance and products. The delivery, integration and expression of defined genes into plant cells are parts of plant biotechnology. While conventional breeders use domestic crop cultivars and its relative genus as a gene source to improve new cultivars, biotechnologists can use defined genes from any organism. After exogenous genes are introduced to as a heritable character to wheat by using biotechnological techniques, the availability of desired genes is important for development of wheat [7].

Transformation, cell and tissue culture, genome mapping's and molecular markers, double haploids, gene isolation, sequencing and bioinformatics are wheat biotechnology research approaches. Tissue culture and transformation studies are two important points in wheat biotechnology. There are three parameters about these studies. The suitable and highly efficient regeneration system improvement is first step of wheat biotechnology research. Secondly, gene delivery technique, reliable and highly efficient, should be developed to introduce the desirable traits to the wheat plants. Finally, a good working screening and selection methods should be used to achieve healthy and usable transformants. Scientists continue to study in order to improve new wheat cultivars having desired new traits by using both regeneration and transformation studies.

Production of a large number of regenerable cells is the main aim of the plant tissue culture studies. After development of regeneration system, transformation studies should be done to deliver of desirable genes into plants. It is considered that regeneration studies is the most difficult part of tissue culture [8].

The growth medium and the external environment are very important components of *in* vitro cell culture studies. The growth medium includes all the essential mineral ions required for growth and development. Macronutrients (or macroelements) such as nitrogen, phosphorus, potassium, magnesium, calcium and sulphur, micronutrients (or microelements) such as manganese, iodine, copper, cobalt, boron, molybdenum, iron and zinc, and iron source as iron sulphate can be categorized essential elements in vitro cell culture. Some amino acids (glycine, arginine, asparagine, aspartic acid, alanine, glutamic acid, glutamine and proline and vitamins (thiamine and myoinositol) can be used as additive organic molecules in many cases. Sucrose is the most commonly added to medium as a carbon source. Glucose, maltose, galactose and sorbitol can also be used as carbon source. One other vital component that must also be supplied is water, the principal biological solvent. Liquid or solidified using agar, plant agar and phytagel media can be used in cell culture. Physical factors, such as temperature, pH, the gaseous environment, light (intensity and duration) and osmotic pressure, also have to be maintained within acceptable limits [8].

Plant growth regulators, plant hormones or their synthetic analogues, are used to directly manipulate the development of the plant cells in the cell culture. Auxins, cytokinins, gibberellins, absisic acid and ethylene are plant growth regulators used in plant cell culture. Cell division and cell growth are promoted by using auxins. The most commonly used auxin is 2,4-Dichlorophenoxyacetic acid (2,4-D) [8].

Cultures are most commonly started from an explant a sterile piece of a whole plant. Some pieces of organs, such as leaves, roots, pollens, endosperms and embryos can be used as explants. Culture initiation is affected form many characteristics of explants. Generally, younger, more rapidly growing tissue is reliable to regeneration [8].

Somatic embryogenesis is formed from somatic tissues. Directly or indirectly somatic embryogenesis can occur. A cell or small group of cells form directly an embryo structure without callus formation in direct somatic embryogenesis. In indirect somatic embryogenesis, firstly, callus is produced, after that embryo is formed from produced callus tissue [8].

Several factors affect *in vitro* cell culture. It is thought that the most important factor is genetic structure of explants. The culture medium needs differ from species to species and cultivars to cultivars. Media components (such as alternative carbon sources, macro- and microelement concentrations and composition), media preparation method and donor plant condition and growth conditions are other factors affecting plant cell culture. One of the main aims of in vitro cell culture is therefore to optimize the components of medium and improve highly efficient regeneration system [8].

Proceedings of the 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences (ICOFAAS 2019) Antalya, Turkey, 8-11 November 2019 The type of explants [9, 10], the genotype of cultivar used [11, 12] and tissue culture media composition [13-15] are important factors affecting in vitro wheat tissue culture.

A highly efficient and reproducible in vitro regeneration system is an absolute prerequisite to produce transgenic plants [16]. Immature embryos, immature inflorescences and mature embryos are the most widely used explants source in wheat regeneration system. Immature embryos [17,18], immature leaves [19], immature inflorescences [20,21], endosperm supported embryos [11], thin mature embryo fragments [22], and mature embryos [13, 23-26] have been used as various explants source in wheat tissue culture.

The aim of this study was to optimize of mature embryo based regeneration of Turkish durum wheat cultivar Mirzabey 2000 for hormone types, concentrations and dark incubation period.

#### **Materials and Methods**

#### **Plant material**

In this study, mature embryos of winter durum wheat *Triticum durum* cultivar Mirzabey 2000 obtained from Agricultural Research Institute, Ankara.

#### Seed surface sterilization

Mirzabey 2000 seeds were surface sterilized with 70 % of ethanol for five minutes, firstly. After that, they were rinsed with sterile distilled water several times. 30 % of sodium hypochloride was used as a surfactant agent for further step for twenty minutes. Surface sterilized seeds were rinsed with sterile distilled water again. Mirzabey seeds were imbibed in 8 mg/mL 2,4-D solution for 16-20 hours [27]. Imbibed seeds were surface sterilized with 70 % of ethanol for one minute and washed with sterile distilled water.

# **Isolation of mature embryos**

Mature embryos were aseptically removed from imbibed seeds using blade and forceps under stereomicroscope. The instruments were sterilized at 250°C in sterilizer. Radical portion of mature embryo was slightly damaged and cultured with scutellum in contact with the medium to start initiation of callus formation.

#### Tissue culture media and regeneration

Two different hormone (2,4-D and Picloram) with three different concentrations (2, 4 and 8 mg/L) and dark incubation period (4 and 6 weeks) were tested for tissue culture optimization of Mirzabey 2000 wheat cultivar. Abbreviations of tissue culture media were used further part of the text. 4W2D medium means 4 weeks dark incubated mature embryos treated with 2 mg/L 2,4-D auxin hormone (Table 1).

Medium	Dark Incubation (Week)	Auxin Type	Concentration (mg/L)
4W2D	4	2,4-D	2
4W4D	4	2,4-D	4
4W8D	4	2,4-D	8
4W2P	4	Picloram	2
4W4P	4	Picloram	4
4W8P	4	Picloram	8
6W2D	6	2,4-D	2
6W4D	6	2,4-D	4
6W8D	6	2,4-D	8
6W2P	6	Picloram	2
6W4P	6	Picloram	4
6W8P	6	Picloram	8

**Table 1.** Tissue culture media tested in this study and their abbreviations.

A modified culture conditions [25] were tested. Isolated mature embryos were firstly incubated in callus induction medium (4.4 g/L MS, 135 mg/L L-aspartic acid, 150 mg/L L-glutamine, 115 mg/L L-proline, 100 mg/L casein hydrolysate, 40 mg/L L-tryptophane, 30 g/L sucrose, 2.6 g/L phytagel and different concentrations of auxin hormones) for 4 and 6 weeks in dark. Calli were weighted end of the 4 weeks and 6 weeks dark incubation and primary callus induction rate was calculated.

After that, calli were transferred into embryogenic callus induction medium (the same with callus induction medium and including additionally 0.1 mg/L naphtalaacetic acid and 0.5 mg/L benzyl adenine) for 4 weeks under low light conditions. After 4 weeks embryogenic callus induction medium incubation, calli were weighted and embryogenic callus induction rate was determined.

Embryogenic calli were carried out regeneration medium (the same with callus induction medium except auxin hormones and additionally including 10 mg/L silver nitrate and 2 mg/L copper sulphate) to regenerate plantlets under 16/8 photoperiod. Additionally, regeneration rate and culture efficiency were calculated for each medium. Moreover, shoot number per plantlets and root formation rate were calculated.

Regenerated plantlets were transferred into root strength medium (2.2 g/L MS, 20 g/L sucrose and 2.6 g/L phytagel) to induce root formation. Rooted plants were incubated at 4°C for 4 weeks for vernalization. After that, plantlets were planted into soil in greenhouse conditions.

# Statistical Analyses

All statistical analyses were performed by using Minitab Statistical Software 13.0. One-Way (Unstucked) (ANOVA) was used for correlation between variables.

#### **Results and Discussion**

According to Table 2, 4 weeks old dark incubated mature embryo derived Mirzabey callus gave the highest average fresh weight of callus from 4W2D medium. However, there was no significantly differences including other concentration of 2,4-D and picloram. It was observed including 4W4P medium had the highest callus induction frequency (91 %). 4W8P medium primary callus induction rate was the minimum (81.86 %). Table 3 shows average callus fresh weight and callus induction rate of 6 weeks dark incubated Mirzabey mature embryo derived callus. Average callus fresh weight (86.38 mg) was the highest in 6W2P medium. Also, 6W2D medium was significantly different than other used hormone concentration according to average fresh weight. The minimum average fresh weight was 44.00 mg in 6W8D medium. According to primary callus induction rate, the maximum rate (93.25 %) was in 6W2P medium. It was reported fresh weight of Bobwhite a model cultivar in tissue culture for including 2 mg/L 2,4-D between 133-281 mg, 4 mg/L 2,4-D 30-86 mg, 2 mg/L picloram 499-607 mg and 4 mg/L picloram 405-565 mg [28]. They also reported auxin type and concentration had significantly effect on callus fresh weight. This approach has similar results in this study. Hormone type and concentration significantly affected Mirzabey 6 weeks old average callus fresh weight. Callus induction was found between 68.75-96.20 % in 2 mg/L 2,4-D from four wheat mature embryo based calli [24]. The callus induction frequency was recorded between 11.6-89.6 % including 8 mg/L 2,4-D medium [29]. In this study, callus induction rate in 8 mg/L 2,4-D varied between 57.40-90.75 %. Chauhan and colleagues [30] used 2 mg /l 2,4-D and found primary callus formation rate between 82-85% for *T.aestivum* and 77-79 % for *T.durum* 2 mg/L 2,4-D in medium being critical point for callus induction was used by Yu et.al [25] and the callus induction rate was between 70.8-95 %.

Medium	Average callus fresh weight (mg) <sup>1</sup>	Primary callus induction rate $(\%)^2$
4W2D	43.38 ± 12.57	86.75 ± 12.27 ab
4W4D	34.88 ± 10.58	88.25 ± 11.54 ab
4W8D	35.00 ± 5.73	82.38 ± 6.07 b
4W2P	$33.83 \pm 8.28$	85.67 ± 7.82 ab
4W4P	$39.83 \pm 5.74$	91.00 ± 6.78 a

**Table 2.** Mirzabey callus fresh weight and primary callus induction rate after 4 weeks dark incubation.

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4W8P	$36.29 \pm 8.16$	$81.86 \pm 11.98b$

Means denoted by different letters in a column are significantly different at P < 0.05 according to One-way ANOVA test. <sup>1</sup>Weight of calli/No.cultured mature embryo. <sup>2</sup>No. explants forming callus/Cultured embryo X 100.

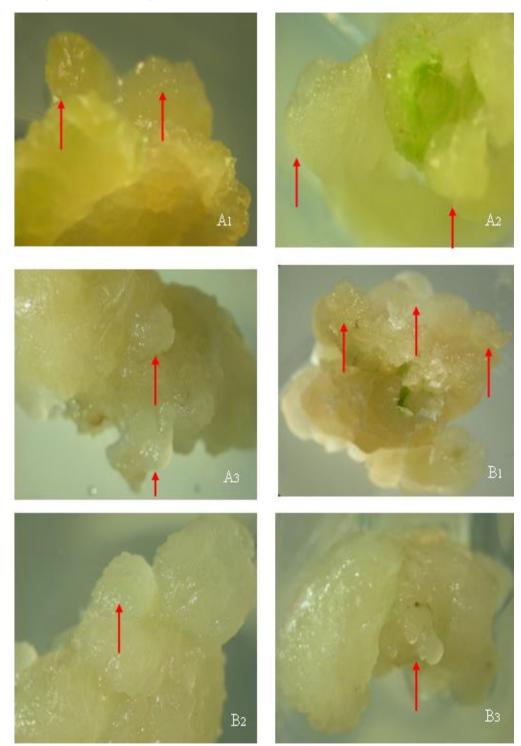
Table 3. 6 weeks old Mirzabey callus fresh weight and primary callus induction rate.

Medium	Average callus fresh weight <sup>1</sup> (mg)	Primary callus induction rate <sup>2</sup> (%)
6W2D	81.38 ± 19.78 a	88.375 ± 10.596 ab
6W4D	59.88 ± 9.60 b	$89.875 \pm 4.824$ ab
6W8D	$44.00 \pm 9.50$ c	90.750 ± 5.970 ab
6W2P	86.38 ± 16.27 a	93.250 ± 4.921 a
6W4P	58.50 ± 15.08 b	84.333 ± 10.053 b
6W8P	$72.50 \pm 16.04$ ab	84.125 ± 7.060 b

Means denoted by different letters in a column are significantly different at P < 0.05 according to One-way ANOVA test. <sup>1</sup>Weight of calli/No.cultured mature embryo. <sup>2</sup>No. explants forming callus/Cultured embryo X 100.

Weight of embryogenic callus and embryogenic callus induction rate was determined for each medium. Calli had globular or heart shape sites (Figure 1). According to Table 4, 4W2D and 4W4P media gave the significantly difference results in terms of average callus fresh weight. The weight was 177.25 mg for 4W2D and 147.00 mg for 4W2P. The minimum weight was 98.75 for 4W8D medium. 8 weeks old mirzabey calli 4 weeks dark incubated embryogenic callus induction rate was evaluated, the significance difference from other media was observed in 4W2D medium and rate was 86.38 %. The minimum embryogenic callus induction rate was 13.25 % in 4W8D medium. If Table 5 was evaluated, callus average fresh weight was the highest in 6W8P medium (256.75 mg). There was no significantly difference between 6W8P and 6W2D media according to fresh weight. These two media were significantly different other four media. When the embryogenic callus induction rate was evaluated, 6W2D medium had the maximum rate (94.88 %). The minimum rate of embryogenic callus induction was observed in 6W8D medium (5.13 %). The necrotic tissues were observed in high concentrations of 2,4-D. The embryogenic callus formation rate for 2 mg/L 2,4-D (63.34 %), 3 mg/L (28.34 %) and 4 mg/L (18.44) was reported [27]. Mendoza and Kaeppler [28] observed detrimental effect of high concentration of 2,4-D resulting brownish and necrotic appearance of 90 % calli. This structure was observed in this study for 4 mg/L and 8 mg/L 2,4-D concentration. Higher concentration of 2,4-D increases probably somatic mutation [31]. It was reported that the rate was between 30.9-48.5 % in 2mg/L 2,4-D [25]. In this study, the embryogenic

capacity was 94.88 % in 6W2D and 86.38 % in 4W2D for Mirzabey. The results recorded in this study show similarity with literature.



**Figure 1.** Embryogenic calli structures of Mirzabey. Red arrows show globular or heart shape embryogenic sites of callus.  $A_1$  from 7 weeks old,  $A_2$  from 8 weeks old and  $A_3$  from 9

Proceedings of the 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences (ICOFAAS 2019) Antalya, Turkey, 8-11 November 2019 weeks old in 2 mg/L 2 4 D. R. from 10 weeks old in 2 mg/L rielorem. R. fr

weeks old incubated in 2 mg/L 2,4-D. B<sub>1</sub> from 10 weeks old in 2 mg/L picloram, B<sub>2</sub> from 9 weeks old in 4 mg/L picloram and B<sub>3</sub> from 8 weeks old in 8 mg/L picloram.

Mediu m	Average callus fresh weight (mg) <sup>1</sup>	Embryogenic callus induction rate (%) <sup>2</sup>
4W2D	177.25 ± 34.67 a	86.38 ± 11.58 a
4W4D	$105.38 \pm 18.86$ c	$36.38 \pm 8.83$ c
4W8D	$98.75 \pm 26.80$ c	$13.25 \pm 14.12 \text{ d}$
4W2P	$116.67 \pm 20.23$ bc	$72.67 \pm 17.20$ ab
4W4P	$147.00 \pm 24.81$ a	$52.17 \pm 18.02$ bc
4W8P	$158.57 \pm 42.69$ ab	58.57 ± 38.78 abc

B<sub>2</sub> from 9 weeks old in 4 mg/L picloram and B<sub>3</sub> from 8 weeks old in 8 mg/L picloram.

Table 4. 8 weeks old Mirzabey callus fresh weight and embryogenic callus induction rate

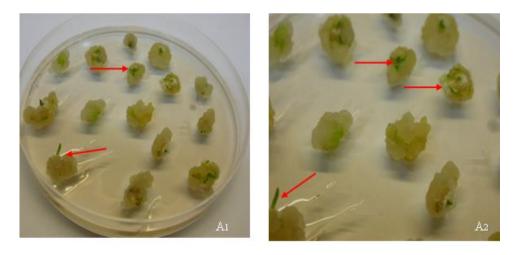
Means denoted by different letters in a column are significantly different at P < 0.05 according to One-Way ANOVA test. <sup>1</sup>Weight of calli/No.Primary calli transferred onto embryogenic callus induction medium. <sup>2</sup>No. explants forming callus/ No.Primary calli transferred onto embryogenic callus induction medium X 100.

Table 5. 10 weeks old Mirzabey callus fresh weight and embryogenic callus induction rate

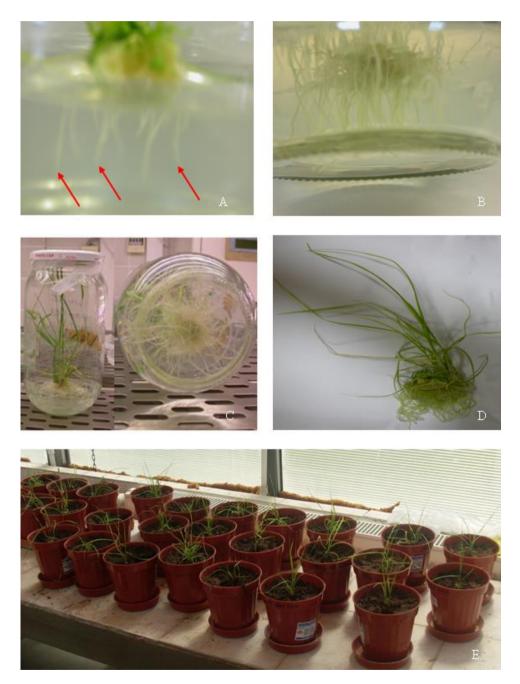
Mediu m	Average callus fresh weight (mg) <sup>1</sup>	Embryogenic callus induction rate (%) <sup>2</sup>
6W2D	251.38 ± 34.60 a	$94.88 \pm 6.24$ a
6W4D	$123.50 \pm 16.59$ c	$59.00 \pm 17.81$ c
6W8D	$72.75 \pm 21.03 \text{ d}$	5.13 ± 7.10 d
6W2P	194.38 ± 23.75 b	79.88 ± 11.23 b
6W4P	190.17 ± 21.93 b	88.17 ± 14.43 ab
6W8P	256.75 ± 58.99 a	83.00 ± 12.86 b

Means denoted by different letters in a column are significantly different at P < 0.05 according to One-Way ANOVA test. <sup>1</sup>Weight of calli/No.Primary calli transferred onto embryogenic callus induction medium. <sup>2</sup>No. explants forming callus/ No.Primary calli transferred onto embryogenic callus induction medium X 100.

Figure 2 shows shoot formation and plant regeneration of Mirzabey in regeneration medium. Figure 3 shows root formation and greenhouse transfer of Mirzabey. The mature embryo based regeneration rate and culture efficiency of Mirzabey was demonstrated in Table 6. The regeneration frequency was the highest in 4W2D medium, 62.31 %. The rate was 47.29 % for 6W2D, 45.02 % for 4W2P, 42.69 % for 6W2P. The rate was determined between 15.63-20.41 % in including 4 mg/L 2,4-D media, 4W4D and 6W4D. For 8 mg/L 2,4-D, regeneration data was not recorded. The embryogenic capacity was 33.42 % for 4W4P, 19.95 % for 6W4P, 16.34 % for 4W8P and 11.95 % for 6W8P. If culture efficiency was evaluated, the 4W2D medium having maximum regeneration capacity had significantly higher culture efficiency rate other media. For picloram, the maximum culture efficiency rate was in 6W2P (39.25 %) and the minimum in 6W8P medium (8.38 %). It was reported that regeneration rate between 2.17-32.40 % and culture efficiency was between 0.60- 27.70 % [24]. According to a wheat tissue culture study, the regeneration rate was maximum 50 % and the minimum 2.17 % from including 2 mg/l 2.4-D medium depending on different genotypes [27]. In 2008, the regeneration rate was reported between 49.1-67.0 using 2 mg/l 2,4-D among the different genotypes [25]. In the same study culture efficiency was changed from 17.8 % to 36.8 %. Including 2 mg/l 2,4-D media gave the maximum regeneration rate and culture efficiency for 4 and 6 weeks dark incubated Mirzabey mature embryo based culture. The significantly effects of cultivar genotype, hormone type and concentration on plant regeneration were demonstrated [24, 28]. The regeneration results obtained in this study showed similarity with literature.



**Figure 2.** Shoot formation and plant regeneration in regeneration medium. Red arrows show shoot initiation. A<sub>1</sub> and A<sub>2</sub> from 10 weeks old Mirzabey incubated in 4W2D medium.



**Figure 3.** Root strength medium and transfer to greenhouse. A shows 3 days after root strength medium incubation of Mirzabey. The red arrows shows root structures. B and C show 3 weeks incubated in root strength medium (nearly 15 weeks old plantlets of

# Proceedings of the 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences (ICOFAAS 2019) Antalya, Turkey, 8-11 November 2019 Mirzabey). D shows plantlet of Mirzabey ready to carry on greenhouse. E shows planted plantlets of Mirzabey 1 week after from soil transfer

Medium	Regeneration rate <sup>1</sup> (%)	Culture efficiency <sup>2</sup> (%)
4W2D	62.31 a	44.13 a
4W4D	15.63 b	5.13 c
4W8D	0.00	0.00
4W2P	45.02 a	27.83 ab
4W4P	33.42 ab	19.00 bc
4W8P	16.34 b	10.43 c
6W2D	47.29 a	39.25 ab
6W4D	20.41 b	9.13 c
6W8D	0.00	0.00
6W2P	42.69 a	32.63 ab
6W4P	19.95 bc	14.33 c
6W8P	11.95c	8.38 c

**Table 6.** Regeneration and culture efficiency rate for Mirzabey

Means denoted by different letters in a column are significantly different at p< 0.05 according to One-Way ANOVA test. <sup>1</sup>No.Regenerated plantlets/No.Cultured callus onto regeneration medium X 100. <sup>2</sup>No. Rgenerated plantlets/ No.Mature embryo cultured onto induction medium X 100.

According to Table 7, the maximum average shoot number per plantlets (8.31) produced Mirzabey mature embryo based callus was determined in 6W8P medium. The shoot formation rate was 6.04 in 4W2D medium having maximum regeneration rate. The minimum shoot formation rate was reported in 4W4D medium. There was no shoot and root formation for 8 mg/L 2,4-D medium because of its detrimental effect. The root formation rate for Mirzabey was the highest in 4W2D medium. Picloram including media gave the variable results between 45.67-78.38 % for root formation rate. It was reported that average shoot number changing between 3.2 and 8.8 in high concentration of 2,4-D [23]. In this study, 4 mg/l 2,4-D media average shoot number was varied between 3.75 and 5.34 and no shoot formation in 8 mg/l 2,4-D media for Mirzabey. They only incubated calli 3 weeks in high concentration of 2,4-D and transferred to regeneration medium. Average shoot formation was found for 2 mg/L 2,4-D between 0.1-0.5, no formation for 4

mg/L [28]. When Mirzabey average shoot number results given in Table 7 were evaluated, the average shoot number was higher than Mendoza and Kaeppler's study [28]. Because, the silver nitrate was used in this study to promote shoot induction as reported by Yu and colleagues [25].

Mirzabey plantlets vernalized 1 month were grown in 3 months. Mirzabey plantlets gave the healthy and normal spikes and seeds. However, the normal and abnormal spikes and seeds were also harvested.

Medium	Average shoot number per plantlets <sup>1</sup>	Root formation rate <sup>2</sup> (%)
4W2D	6.04	81.63
4W4D	3.75	23.38
4W8D	0.00	0.00
4W2P	6.85	56.50
4W4P	5.10	45.67
4W8P	4.90	47.43
6W2D	8.01	64.13
6W4D	5.65	29.88
6W8D	0.00	0.00
6W2P	5.34	63.25
6W4P	6.57	70.88
6W8P	8.31	78.38

Table 7. Average shoot number per plantlets and root formation rate for Mirza	abey
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<sup>1</sup>No.Shoot on regenerated plantlets/No.Rgenerated plantlets. <sup>2</sup>No.Callus with root/ No.Mature embryo cultured onto induction medium X 100.

# Conclusion

In this study, the regeneration paramaters of mature embryo based culture of pasta wheat cultivar Mirzabey 2000 (*Triticum durum*) was optimized. The maximum regeneration rate (62.31 %) was observed in 4W2D medium and also, culture efficiency (44.13 %) was the highest for this medium. The rate was 47.29 % for 6W2D, 45.02 % for 4W2P and 42.69 % for 6W2P media. There was no record of regeneration of Mirzabey calli in 6W8D and 4W8D media.

As a conclusion, the usage of including 2 mg/L 2,4-D medium was important for primary callus induction, embryogenic callus induction and regeneration in mature embryo based wheat tissue culture studies. The use of high concentration of 2,4-D caused browning of tissues and death of calli and decreased the regeneration potential. The picloram can be used as an alternative hormone type instead of 2,4-D. The further optimization studies must be performed to increase effect of picloram.

# Acknowledgements

This work was supported by the research fund: BAP-08-11-DPT-2002-K120510. This study was a part of Master of Science Thesis of Abdulhamit BATTAL. Van Yüzüncü Yıl University Scientific Research Project Department supported this study to present in this congress with project number: FAP-2019-8082.

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# Nicking on Seed Coat Increases Germination Rate of Medicinally Important Devegülü Marshmallow Plant

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#### Abstract

The medicinal plant deve gülü which belongs to the Malvaceae family is used in the treatment of tonsilitis, gastric ulcers, pneumonia, excretory system infections and pelade. The aim of this study was to develop an in vitro germination method for deve gülü plant as a source of sterile explants in tissue culture studies. Three different sodium hypochlorite concentrations (5%, 7.5% and 10%), incubation at 42 °C and nicking of the seed coat with scalpel were tested for germination. According to germination rate results, it was determined that incubation at 42 °C did not affect germination rate. On the contrary, seeds damaged by scalpel germinated rapidly. Cotyledon, cotyledonary node and hypocotyl became available as explant source for tissue culture studies on the seventh day. Leaf formation started on the eighth day. Leaf, petiole and shoot tip explants were available to use in tissue culture on the thirtieth day. As a conclusion, an efficient germination method for deve gülü marshmallow having medicinal importance was developed with this study.

Keywords: Alcea, Deve gülü, Hollyhock, Germination rate.

# Introduction

*Alcea* genus which has a medical importance is a member of Malvaceae family. The most well-known species is *Alcea rosea* L., which extends from the southwestern parts of the People's Republic of China to many parts of the world [1]. Its leaves, flowers, stem and roots are consumed by humans [2-4]. While petals and buds are consumed by joining salads, fresh leaves are eaten with or without cooking [5]. In addition, the colors in the petals are used as food additives in apparel products, jams and jellies and soft drinks [2].

Alcea's aerial parts contain bioactive substances such as mucilage, glucuronic acid, galacturonic acid, rhamnose, galactose, pectin, ferulic acid, valinic acid, comaric acid, p-hydroxybenzoic acid and quercetin, while its seeds contain malvalic, stretchic, myristic, palmitic, stearic, oleic and linoleic fatty acids [6-9]. The antiproliferative activity, antiinfluenza effect, antioxidant properties and anti-ulcer activity of *A.rosea* have been demonstrated by studies [10-13].

Medicinal plants can be propagated quickly and in desired amounts through plant tissue culture. The source, type and freshness of the explants to be used in plant tissue culture studies are important for the micro-propagation studies. In the studies with *Althae officinalis* L. (Marshmallow), which is the same family and having the medicinal importance, they were generally used either from sources grown in botanical gardens or from plants germinated from seeds under sterile conditions [14-16]. However, plants from botanical gardens are highly susceptible to contamination since they are not grown under sterile conditions. For the sterilization of these plants, environmentally harmful chemicals such as mercury chloride are often used [14, 15]. At the same time, physiological conditions may vary from plant to plant as well as environmental stress factors and vegetation periods. These factors negatively affect the regeneration system for medicinal plants.

Deve gülü marshmallow is a medicinal plant belonging to the genus *Alcea* used against tonsillitis, gastric ulcer, pneumonia, excretory system infections and pelade in western Iran and north of Iraq [17, 18]. Dried aqueous extract of deve gülü plant has been shown to have antimicrobial and antioxidant activity [19]. While there is a proposed germination method for *Althae officinallis* L., there is no germination procedure for tissue culture studies for deve gülü marshmallow. The aim of this study is to create an effective germination method for deve gülü plant used in folk medicine and to produce explants that can be used in plant tissue culture studies under sterile conditions.

# **Material and Method**

# **Plant Material**

In this study, seeds of deve gülü marshmallow species collected from Van region were used. Seeds were collected in August and September 2017. The seeds that were brought to the laboratory were removed from the impurities and then laid on drying papers and dried. Dried seeds, in which dust and pests were removed, were packaged and stored at  $+4^{\circ}$ C.

# **Surface Sterilization of Seeds**

Seed wings were manually removed prior to seed sterilization. Different concentrations of sodium hypochlorite (NaOCl) (5%, 7.5% and 10%) were used for sterilizing the seeds. The seeds were incubated for 20 minutes at 300 rpm in a shaking incubator at these concentrations, then washed with sterile distilled water 4-5 times in a biosafety cabinet and treated with 70% ethyl alcohol for 30 seconds. Seeds were washed again with sterile distilled water 4-5 times. Seeds were divided into 3 groups for each sodium hypochlorite concentration (5%, 7.5% and 10%) in order to investigate the effect of incubation at 42°C and nicking of seed coat on germination of deve gülü seeds in plant tissue culture (Table 1). Bacterial and fungal growth was observed during germination of the seeds.

# Preparation of MS media for incubation of Deve gülü Seeds

The seeds that were sterilized were incubated in 1/2 (half strength) MS medium. To prepare the medium, 2.2 g / L MS medium and 15 g / L sucrose were weighed and dissolved in distilled water and adjusted to pH 5.8. Then 8 g / L plant agar was weighed and added to the solution. The prepared medium was autoclaved and poured into sterile 90 mm petri

Proceedings of the 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences (ICOFAAS 2019) Antalya, Turkey, 8-11 November 2019 dishes in a biosafety cabinet. Ten seeds of each group were sown on 1/2 MS medium and germinated in photoperiod for 16/8 hours in plant tissue culture chamber.

# Germination of Deve gülü Seeds

Germination conditions were followed at the 1st, 2nd, 3rd, 5th, 10th and 30th periods and germination rates (%) on the 5th and 30th days were determined according to the formula below;

Germination Rate (GR) % = (Number of germinated seeds / number of sown seeds) X 100

Group Name	Application
5-SH	Surface sterilized seeds with 5% sodium hypochlorite.
5-SH+42°C	Seeds were surface sterilized with 5% sodium hypochlorite and incubated for 24 hours at 42 ° C.
5-SH+42°C+N	Surface sterilized with 5% sodium hypochlorite, incubated at 42 ° C for 24 hours and nicked on seed coat with scalpel. After incubation, the seeds were subjected to surface sterilization with the same sodium hypochlorite for 10 minutes before nicking.
7.5-SH	Surface sterilized seeds with 7.5% sodium hypochlorite.
7.5-SH+42°C	Seeds were surface sterilized with 7.5% sodium hypochlorite and incubated for 24 hours at 42 $^{\circ}$ C.
7.5-SH +42°C+N	Surface sterilized with 7.5% sodium hypochlorite, incubated for 24 hours at 42 $^{\circ}$ C and nicked on seed coat with scalpel. After incubation, the seeds were subjected to surface sterilization with the same sodium hypochlorite for 10 minutes before nicking.
10-SH	Surface sterilized seeds with 10% sodium hypochlorite.
10-SH +42°C	Seeds were surface sterilized with 10% sodium hypochlorite and incubated at 42 ° C for 24 hours.
10-SH +42°C+N	Surface sterilized with 10% Sodium hypochlorite, incubated at 42 ° C for 24 hours and nicked on seed coat with scalpel. After incubation, the seeds were subjected to surface sterilization with the same sodium hypochlorite for 10 minutes before nicking.

# **Statistical Analysis**

Experiments were performed with at least three replicates and statistical analyses were performed using Minitab12 One-Way ANNOVA. In the statistical comparisons between the groups, p <0.05 values were evaluated as statistically significant changes.

# **Results and Discussion**

In this study, the effect of three different concentrations of sodium hypochlorite (5%, 7.5% and 10%) on germination of seeds of deve gülü plant by using plant tissue culture media was evaluated. The abundance of explant sources to be used in plant tissue culture is important. Different parts of plants such as root, stem, cotyledons, leaves, embryo, cotyledon stem and leaf stem can be used as explant source in plant tissue culture studies [14,15]. Therefore, sterile germination of plants using artificial plant tissue culture media is very important to provide sufficient explant source. Bacterial and fungal growth was determined in the 5-SH groups at the end of day 2, while 5-SH + 42°C group was found to have bacteria and fungus at the end of day 3. At the end of day 1, fungal growth was observed in the medium containing 7.5-SH+42°C+N group, while bacterial and fungal growth was determined in the medium containing 7.5-SH, 7.5-SH+42°C and 7.5-SH+42°C+N groups (Figure 1). The media contaminated with bacteria and fungi were kept up to the 5th day and after germination rates were determined, they were destroyed by autoclave. In groups treated with 10% sodium hypochlorite, no contamination was observed until day 30. It was determined that the application of 10 % sodium hypochlorite to deve gülü seeds for 20 minutes was sufficient for sterilization. (Table 2)

Sterilization agents such as ethanol, sodium hypochlorite (NaOCl), Tween-20 and mercury chloride (HgCl<sub>2</sub>) are generally recommended for surface sterilization of seeds [20, 21]. The seeds of *Althaea officinalis* L. were treated with fungicide and multi-purpose detergent followed by sterilization with 0.1% mercury chloride (HgCl<sub>2</sub>) and their nodes were used for tissue culture studies [16]. In another study using the same plant, different concentrations of HgCl<sub>2</sub>, NaOCl, ethanol and Tween-20 sterilization agents were applied at different times. Contamination rates of 14.66% (0.3% HgCl<sub>2</sub>, 7 minutes) and 85.33% (sterile distilled water, 20 minutes) were observed, and optimum sterilization was achieved by using 4% NaOCl for 5 minutes [22]. In the same study, it was reported that 4% NaOCl administration for 10 minutes caused contamination. In this study, it was found that 10 minutes treatment with 10% sodium hypochlorite and 30 seconds treatment with 70% ethanol were sufficient for sterilization.

Application	Bacterial Contamination	Fungal Contamination
5-SH	+	+
5-SH+42°C	+	+
5-SH+42°C+N	+	+
7.5-SH	+	+
7.5-SH+42°C	+	+
7.5-SH +42°C+N	+	+
10-SH	-	-
10-SH +42°C	-	-
10-SH +42°C+N	-	-

**Table 2.** Bacterial and fungal contamination.

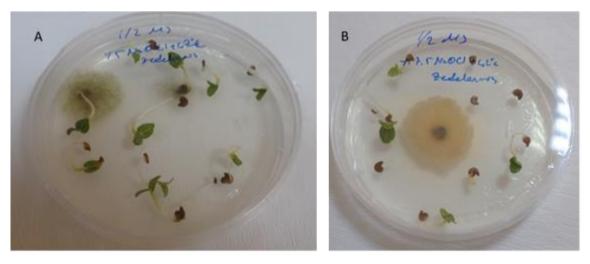


Figure 1. Contamination on 5% and 7.5% SH treatments. A: 5-SH+42°C+N; B: 7.5-SH+42°C+N treatments.

# Germination Rate of Deve gülü Plant Seeds

In the preliminary studies, different concentrations of sterilizing agents such as HgCl<sub>2</sub>, ethanol and sodium hypochlorite were tested on seeds that were not scratched with scalpel for different periods of time and germination was not observed. Then, germination of  $\frac{1}{2}$ 

MS medium of seeds of deve gülü plant, which was scored by scalpel, was observed on days 1, 2, 5, 10 and 30. On the 5<sup>th</sup> day, germination rates were calculated for all applications, while on the 30<sup>th</sup> day germination rate was calculated only for groups sterilized with 10% sodium hypochlorite without contamination. At the end of day 1 it was observed that germination was started only in seed groups damaged by scalpel after the sowing of the seeds. Seeds did not germinate in groups that were incubated at 42°C and the seed coat was not damaged and treated only with sodium hypochlorite. 5<sup>th</sup> day of germination rate with a maximum rate of 90.00  $\pm$  10.00 5-SH+42°C+N application was determined (Table 3). However, both bacteria and fungi were observed in this application.

Germination was observed at 7.5-SH+42°C+N ( $66.67 \pm 25.166\%$ ) and 10-SH+42°C+N ( $66.67 \pm 20.817$ ) treatments at the end of day 5, while 7.5-SH+42°C+N application bacterial and fungal contamination occurred (Table 2 and Figure 1). Contamination was not observed in 10-SH+42°C+N application.

At the end of 5 days, 5% and 7.5% SH applications were discontinued and the experiment was continued with 10% SH groups without contamination. At the end of the 30th day, germination rate was only  $3.33 \pm 5.774$  % in 10-SH application and germination rate was calculated as  $86.67 \pm 5.774\%$  in 10-SH+42°C+N application (Table 3 and Figure 2). When the results were evaluated, it was determined that incubation of seeds at 42 °C for 24 hours did not encourage germination of deve gülü plant, while seeds nicked on their coat had high germination rates. Damage to the seed coat accelerated the water uptake of the seeds and the enzyme activities required for germination were initiated.

Germinated seeds sprouted rapidly and cotyledons formed. One week after germination, the cotyledon, cotyledon stems, hypocotyl and epicotyl of the shoots were ready to be used as explants in tissue culture. The leaves started to form on the 8th day and at the end of the 30th day the leaf, leaf stem and shoot tip explants became available for tissue culture (Figure 3).

In a study with *Althea officinalis* L. germination rate was reported to be quite low [23]. In another study with *Alcea aucheri* (Boiss.) Alef., the effect of different sulfuric acid concentrations and application times, different concentrations of the gibberellic acid and the different application times of the sandpaper on the germination of the plant seeds were examined. The highest germination rate (42%) was obtained with the application of 5 minutes of sanding has been reported [24]. In the study, where an effective germination procedure was reported for the hollyhock plant, the highest germination rate was obtained by using sterile distilled water with 76% and the lowest germination rate was found to be 28% on solidified MS medium with 3% sucrose added agar [22]. In a study conducted with *Rheum ribes* L., a medicinal plant, different concentrations of sulfuric acid, gibberellic acid, calcium chloride and potassium nitrate were tested at different stratification times, and the highest germination rate (57.3%) was achieved with giberellic acid at 25 days at 4 °C. incubated samples have been reported [25]. When the results were evaluated, germination of deve gülü plant, which is one of the marshmallow species, was realized at a rate close to 90%.

	Germination Rate (%)	
	Mean ± Standard Deviation	
Applications	5 <sup>th</sup> Day	30 <sup>th</sup> Day
5-SH	3.33±5,774 <sup>a</sup>	N.D
5-SH+42°C	0.00±0.000 <sup>a</sup>	N.D
5-SH+42°C+N	90.00±10.00 <sup>ab</sup>	N.D
7.5-SH	3.33±5.774 <sup>a</sup>	N.D
7.5-SH+42°C	$0.00 \pm 0.000$	N.D
7.5-SH +42°C+N	$66.67 \pm 25.166^{ab}$	N.D
10-SH	0.00±0.000 <sup>a</sup>	3.33±5.774 <sup>a</sup>
10-SH +42°C	0.00±0.000 <sup>a</sup>	0.00±0.000 <sup>a</sup>
10-SH +42°C+N	$66.67 \pm 20.817^{ab}$	$86.67 \pm 5.774^{ab}$

**Table 3.** Deve gülü day 5 and day 30 germination rates

Different letters in the same column indicate statistically significant p < 0.05 difference between the means. N.D: No data.

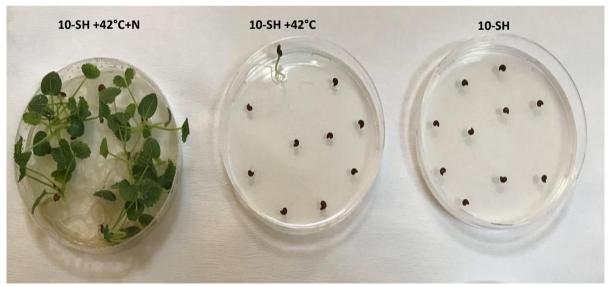


Figure 2. Germination conditions of 10% SH applications

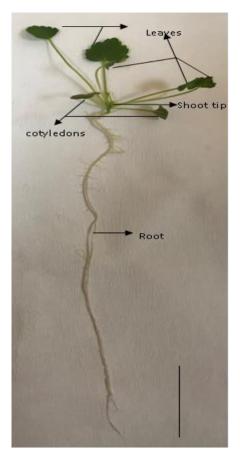


Figure 3. 30 day old deve gülü plant.

# Conclusion

As a result, an effective germination procedure was achieved by nicking on seed coat for deve gülü plant and it was provided to obtain explant sources which can be used without problem for plant tissue culture studies. The applicability of the developed germination method can be tested for other medicinal plants with low germination rate as well as other marshmallow species.

# Acknowledgements

We would like to thank to Assoc. Prof. Dr. Abdullah Dalar and Dr. Muzaffer Mükemre for supplying of plant seeds and Prof. Dr. Murat Ünal for its identification. This study was supported by Van Yüzüncü Yıl University Scientific Research Projects Department (FDK-2017-6160). This study is a part of Doctorate Thesis of Gül Görmez. This study was published in Bitlis Eren Üniversitesi Journal of Science.

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# Physiological Approaches to Improve Terminal Drought Tolerance in Corn (Zea mays L.)

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# Abstract

Corn is an essential dietary component in human food and also in animal feed across the globe. With the rising trend of global climate change, growth, productivity and quality losses of corn are expected to increase, because of various biotic and abiotic stress in all over the world. Among the abiotic stress, drought stress is considered one of the greatest limiting factors in agriculture. Therefore, the ability of corn, adapt to drought conditions is crucial for sustaining crop production worldwide. The critical growth stages of corn to water deficit stress are seedling establishment, tillering and reproductive stages. There are different ways to alleviate drought stress in corn production, either by developing and practicing improved drought management practices or by developing drought-tolerant cultivars. The present review explores the recent advances of physiological approaches to improve drought tolerance in corn and also highlights the possible ways to identify or develop the promising drought-tolerant genotypes as well as mitigation strategies to meet the food security of increasing population in the era of climate change.

Keywords: corn, physiology, drought stress, climate change, economic yield

# Introduction

Corn (*Zea mays* L.) is an important cereal that is used as human food (20-25%), livestock and poultry feed (60-75%) and also a raw material in industry (5-15%) (Emam, 2004). Due to its high content of sugar and starch, corn is also considered one of the best economic crops for producing quality green and silage forages yield (more than 80 tons forage per hectare) and high-quality grain (Tajbakhsh and PourMirza, 2003).

While as a result of the changing climate, growth, productivity and quality losses of crops including corn are expected to increase, due to the various abiotic stresses. Among the abiotic stress, drought stress is a major restrictive factor that limits crop productivity worldwide by altering the physio-biochemical activity of plant process. This is especially true for the cereal crops all of which demonstrate a range of detrimental effects from drought stress (Muthomi and Musyimi, 2009). Although the morphological, physiological and biochemical responses of plants to drought stress is varied with the intensity of drought stress and genotype-environment interaction. Saini and Westgate (Saini and Westgate, 2000) have been reported that the early growth and reproduction stages of corn are the most sensitivite to water deficit stress, which leads to reducing the final yield. Similarly, other researchers found that two weeks after pollination to physiological maturity of corn is the most sensitive stage to drought stress, and significantly decreases the grain weight of affected plants (McPherson and Boyer, 1977). The critical growth stages of cereals to drought stress are the seedling establishment, tillering and reproductive stages which may result in significant yield losses of affected crops and even lethal to the crops (Ludlow and Muchow, 1990). Grain yield in corn is strongly correlated with yield components and any reduction in the yield components leads to a reduction in the final yield (El Sabagh et. al., 2018). The seed setting stage is highly sensitive to adverse environmental conditions, particularly the tasselling and silking stages are highly sensitive to drought stress (Tollenaar, 1977).

The responses of drought stress on plants depend on its intensity and duration and also genotype-environment interactions due to the different genetic makeup of particular cultivars as well as harshness of the environment. Plants have a short term or long term physiological response to the drought stress, depend on survival ability. During drought stress, tolerant plants can change their leaf relative water and leaf chlorophyll content to survive for a short term response against drought (Ahmadi and Ceiocemardeh, 2004). The present review explores the recent advances of physiological approaches to improve drought tolerance in corn and also highlights the possible ways to identify or develop the promising drought-tolerant genotypes as well as mitigation strategies to meet the food security of the increasing population in the era of climate change.

# **Physiological responses**

During drought stress, a remarkable genotypic variation in the stomatal conductance of corn genotypes was observed by Bahar et al. (Bahar et. al., 2009). Since the stomatal conductance which is a key trait of the photosynthetic leaf was significantly influenced by abiotic stresses (Jiang et. al., 2006). Delay canopy senescence due to various light interceptions by green leaf area has been reported to be necessary for the high productivity of hybrid corn under normal and drought stress conditions (Cairns et. al., 2012).

The performance of crops under drought conditions is a complex phenomenon when drought occurs during the reproductive growth stage, plant reduces the demand for carbon by decreasing the size of sink which eventually diminished the grain yield of crops (Reynolds et. al., 2006). The Identification of phenotypic, ideotype traits and donors are important in breeding programs for development of drought-tolerant genotypes (Cairns et. al., 2012). The changes in leaf morphology induced by drought caused higher reflectance

*Proceedings of the* 2<sup>nd</sup> **International Conference on Food, Agriculture and Animal Sciences (ICOFAAS 2019)** *Antalya, Turkey, 8-11 November 2019* in the visible spectra between stressed and unstressed corn leaves also observed by (Genc et. al., 2013).

Physiological trait such as stomatal conductance is an important trait which influences the yield significantly under drought stress and it is an important indirect crop selection criterion for drought-tolerant cultivars (Koc et. al., 2008).

# **Grain-filling stage**

Various metabolic processes occurring in the leaves, especially production and translocation of photo-assimilates, importing precursors for biosynthesis of seed reserves, minerals, and other functional constituents influenced the grain-filling. These processes are highly sensitive to drought and heat stresses due to their involvement with diverse enzymes and transporters, located in the leaves and seeds (El Sabagh et. al., 2018; Barutçular et al., 2017; Barutçular et al., 2016). Abdelaal et al. (Abdelaal et. al., 2017) reported that the grain weight and other yield traits were increased significantly under well-irrigated conditions (without moisture stress) as compared to drought conditions. Yield attributes such as stem length, ear height, number kernels row<sup>-1</sup>, grain weight, grain yield, biomass yield and harvest index of corn were highly affected by drought stress (El Sabagh et. al., 2018).

During the critical period of corn such as silking to early grain-filling drought inhibits photosynthesis rate, and consequently lowers the carbohydrate reserves that are insufficient to support optimum reproductive development, causes reduction in the photosynthates mobilization to seeds and thereby reduction of grain weight (Eck, 1986). Kamara et al. (Kamara et. al., 2003) found that water deficit at the grain-filling stage decreases grain weight due to the reduction of the remobilization of photosynthates into the grains. Similarly, Farooq et al. (Farooq et. al., 2009) found that water shortage from vegetative to reproductive stage of corn leads to reduce the grain yield of the crop. Earlier findings also reported that anthesis period is the most sensitive stage to drought in corn growth and development that ultimately reduced that grain yield (El Sabagh et. al., 2018; Zharfa et. al., 2011).

# Genotypic and phenotypic variation of corn genotypes under drought stress

The significant variations among different corn genotypes with respect to grain yield and yield traits indicate the existence of genetic variation and possibility of selection for drought tolerance genotypes (El Sabagh et. al., 2018). The reality of high diversity among corn hybrids studied for drought tolerance had been reported by several investigators (Mostafavi et. al., 2011). The adverse effect of drought stress on the physiological traits of corn genotypes reduces the production of dry matter, disturbs the partitioning of carbohydrates to grains and finally reductions of the harvest index (Mostafavi et. al., 2011).

# Correlation analysis between growth traits and yield attributes

A significant correlation coefficient between grain yield and grain weight was noted, while kernels row<sup>-1</sup> was negatively correlated with grain yield (El Sabagh et. al., 2018). A positive correlation between grain yield and drought resistance index (DRI<sub>SC</sub>) (r=0.784, P<0.037) (in the young leaves) under drought environment was observed by EL Sabagh et al. (El Sabagh et. al., 2018). The correlation between stomatal conductance and grain yield were positive (but non-significant) at early milky stage (r = 0.165) but non-significant at late milky stage (r = -0.234) under normal watering, while no positive correlation between

stomatal conductance and grain yield was observed at early maturity stage under drought condition (Bahar et. al., 2009). Corn grain yield was negatively correlated with root and leaf growth rates under the drought.

# Selection of drought-tolerant genotypes based on tolerance indices

Several drought tolerance indices could be used for screening drought-tolerant genotypes, based on yield loss as compared with normal conditions (Naghavi et. al., 2011). Drought sensitivity and tolerance indices of the corn genotypes may be determined based on the grain yield obtained under drought stress reported by (Naghavi et. al., 2011; Hossain et. al., 2018). Stress tolerance index (STI), geometric mean productivity (GMP) and mean productivity (MP) can be used to select tolerant genotypes to drought (Hossain et. al., 2018). Furthermore, EL Sabagh et al. (El Sabagh et. al., 2018) noticed that tolerance index (TOL) and stress susceptibility index (SSI) appeared to be the most suitable indices for selection of high yielding genotypes under drought stress. The genotypes with high values of yield stability index (YSI), drought resistance index (DI) and harmonic mean (HM) might be selected as tolerant genotypes to drought (Hossain et. al., 2018).

# Quality traits of corn

Grain quality is governed by a number of factors particularly the duration and rate of grainfilling and the availability of assimilates that are negatively influenced under the drought stress (Brdar et. al., 2008). Usually, quality properties of corn are usually affected by genotypes, environmental factors, and their interactions. Grain-filling stage is the most environmentally sensitive phase in corn which strongly influenced the development of grain quantitatively and qualitatively (Yıldırım et. al., 2018). Oury and Godin (Oury and Godin, 2007), reported a negative correlation between protein contents and grain weight in corn under drought stress. Association analysis revealed that cob height, thousand-grain weight, and protein contents had significant relationship with grain yield of corn (Yousaf et. al., 2018). Generally, it is predicted that gene controlling cell division gets downregulated due to drought which could be responsible for decreasing cells number in cotyledons along with endosperm (Sehgal et. al., 2018).

# Adaptation strategies of corn to drought stress

Under stress conditions, plants exhibit various surviving mechanisms including long-term evolutionary phenological and morphological adaptations, and short-term avoidance or acclimation mechanisms of changing leaf orientation, transpirational cooling, or alteration of membrane lipid compositions (Hasanuzzaman et. al., 2011). The incorporating drought and heat tolerance into corn germplasm has the potential to offset predicted yield losses and sustain corn productivity under climate change including drought also (Tesfaye et. al., 2009).

Managing water consumption of the crops is one of the strategies which should be adopted locally and worldwide in current trends for sustaining agricultural productivity. Plants can survive under different stress conditions by mobilizing various defense mechanisms as well as altering their physiological metabolism, and growth pattern (Mittler, 2002). Therefore, the accumulation of osmolytes is a prerequisite for osmotic adjustment of all organisms under drought stress (Zhang et. al., 2009). Several studies reported that exogenous application of proline (Pro) help to survive plant under different abiotic stresses like

Proceedings of the 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences (ICOFAAS 2019) Antalya, Turkey, 8-11 November 2019 drought (Ali et. al., 2008). Although little attention has been given to the role of Pro in affecting the uptake and accumulation of inorganic nutrients in plants, however, many

studies have given a much attention to stress tolerance in plants as a compatible osmolyte for osmotic adjustment during drought stress (Ali et. al., 2008).

In recent decades, exogenous applications of protectants such as osmoprotectants, phytohormones, signaling molecules, trace elements, etc. have shown beneficial effects on plant growth under stress environment, due to the growth-promoting and antioxidant activities of these compounds (Hasanuzzaman et. al., 2011). While it is well established that glycine betaine (GB) a quaternary ammonium compound and a very effective compatible solute that accumulates in plants' leaves during various types of environmental stresses including drought (Ashraf and Foolad, 2007). Salicylic acid (SA) is a naturally occurring plant hormone that influences various physiological and biochemical functions in plants. It has been also proposed that SA acts as an endogenous signal molecule responsible for inducing environmental stress tolerance in plants (Muthomi and Musvimi, 2009). SA enhanced photosynthetic rates, leaf area and plant dry matter production (Elwana and El-Hamahmy, 2009). SA also acts as a potential non-enzymatic antioxidant in regulating a number of plant physiological processes including photosynthesis (Arfan et. al., 2007). Exogenously applied ascorbic acid (AA) ameliorates the adverse effects of drought. It has been reported that maintenance of water status in plants is regulated by stomatal conductance and rate of transpiration (Ashraf, 2009). Other researchers, Yazdanpanah et al. (Yazdanpanah et. al., 2009) also reported that AA in plants plays a significant role in regulation of mechanisms of photosynthesis and defense against oxidative stress under stress conditions. Application of abscisic acid (ABA) stimulated the morphological and anatomical modification in plants which could provide the roots to break through compacted soil (Hartung et. al., 1994). Abdelaal et al., (Abdelaal et. al., 2017), found that the yield traits (grains ear<sup>-1</sup>, 100-grain weight) and grain yield of corn were significantly influenced by (ABA) and yeast under droughtconditions. The application of ABA significantly improved the antioxidant enzymatic activity under drought stress in corn crops (Lee and Luan, 2012).

# Strategies to avoid and tolerate of drought stress at the grain filling stage of corn

In order to enhance crop productivity in the semi-arid region, nano-fertilizer can be introduced as a promising tool in agroecosystems which is economic and environmentally friendly. It is very necessary to know about the nano-technological information to enhance drought stress tolerance of various crops occurring at reproduction and grain formation phase. In a new type of nano-fertilizers, the nutrients can be released in response to environmental factors. Nano-technological discoveries could open up novel applications in the field of biotechnology and agriculture. Nano-fertilizers are innovative agricultural inputs that are aimed to release nutrients into the soil gradually and in a controlled way, thus avoiding environmental damages and improving crop growth and productivity (Sekhon, 2014). According the findings of Janmohammadi et al. (Janmohammadi et. al., 2016), indicated that mild deficit irrigation during the reproductive growth stage in corn with application of nano-chelated micronutrients would be efficient management tools for maximizing water use efficiency and yield in semi-arid regions.

Elucidating corn hybrid for drought tolerance could be an indispensable step towards a balanced yield under the changing climate. The base of further research should be focused on spring corn crop. Application of osmoprotectants, nano-technology, and use of

sustainable agriculture agents have become necessary for further aspect of research. From the present review, it can be concluded that, with the rising trend of global climate change, especially due to drought stress, growth and yield of corn reduce significantly all over the world especially in the arid and semi-arid regions across the globe. Hence, the development and use of drought-tolerant corn genotypes have a great potential to stabilize the grain yield in the era of climate change for meeting the food demand of the increasing population.

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# The Present Status of World Vegetable Seed Trade

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### Abstract

This study was conducted to assess the present status of world vegetable seed trade. According to 2016 data of International Seed Federation (ISF), vegetable seed export was 114 661 tons with a monetary value of 397 million dollars. The vegetable species with the greatest seed exports were identified as squash (46%), potato (23.3%), broad bean (12.7%), pea (5.3%), bean (3%) and onion (2.4%). The greatest revenue in export generating seed species were identified as tomato (25.5%), cucumber (15.5%), squash (14.8%), melonwatermelon (11.3%), onion (9.3%) and pepper (7%). The leading countries in world vegetable seed export were identified as the USA (15.6%), Netherlands (12.8%), Italy (12%), China (10.9%), France (9.5%), Denmark (9.2%) and New Zealand (9%). With the regard to monetary value of the exports, again the Netherlands (43.1%), USA (19.5%) and France (12.9%) were leading countries. World vegetable seed import was 124 664 tons with a monetary value of 3 828 million dollars. The seed vegetable species with the greatest imports were identified as potato (88.2%), pea (6.3%), spinach (1.8%) and bean (1.3%). The seed species with the greatest import monetary values were identified as tomato (33.2%), potato (14.2%), pepper (8.4%), cucumber (7.6%), melon-watermelon (7.6%) and squash (4.2%). With regard to vegetable seed imports, China (13.4%), the USA (12.8%), United Kingdom (12.6%), Netherlands (10.1%) and Italy (7.6%) were the leading countries. With regard to monetary value of the imports, The Netherlands (15.4%), USA (12.7%), Mexico (11.4%) and Spain (9.9%) were the leading countries.

Keywords: Vegetable seed, poduction, export, import.

# Introduction

Good and quality seeds constitute the bases of a successful agricultural production. World seed trade mostly includes cereals, oil seeds, vegetables, flowers and grass seeds. High quality seed is especially important in vegetable production activities. Vegetable culture is either practiced in open fields or undercover in greenhouses and it is a costly labor-intense production branch of agriculture. The sector has been more integrated into seeds than several other agricultural products. This paper was prepared to assess the present status of world vegetable seed trade and to put forth the place of Turkey in world vegetable seed trade.

# World Vegetable Seed Trade

# 2.1. Export Quantity and Monetary Value

The data for world vegetable seed trade between the years 2011 - 2016 and distribution of world vegetable seed export quantity (kg) based on species are provided in Table 1 and Table 2.

Year	Quantity	Annual change	Monetary Value	Annual change
	(tons)	(%)	(Million Dollars)	(%)
2011	117 144	-	3 309	-
2012	116 400	-1	3 447	4
2013	114 065	-2	3 564	3
2014	117 854	3	3 791	6
2015	115 747	-2	3 630	-4
2016	114 661	-1	3 976	9

**Table 1.** Quantity and monetary value of world vegetable seed export

Source: International Seed Federation (ISF)

Species					Change	Part
	2012	2013	2014	2016	(%)	(%)
Potato	149 250	2 200 175	3 134 640	833 020	458	23.3
Sweet corn	36 247	22 722	3 139	82 996	128	2.3
Squash	1 280 673	364 545	356 885	1 642 370	28	46.0
Cucumber	4 051	4 314	3 083	9 299	129	0.2
Melon-Watermelon	9 496	8 231	9 206	13 981	47	0.4
Tomato	2 994	581	466	1 903	-37	0.05
Pepper	1 209	1 593	688	2 261	87	0.06
Eggplant	287	186	52	531	85	0.01
Okra	93	151	1 038	3 932	4 128	0.11
Onion	40 940	47 070	10 754	84 893	127	2.4

**Table 2.** Distribution of world vegetable seed export quantity (kg) based on species

Total	2 461 946	3 092 240	3 770 208	3 573 781	45	100
Other vegetable seeds	24 991	46 627	30 516	77 506	210	2.1
Kidney bean	200	-	- 20 516	21 788	1089	0.6
Broad Bean	808 205	334 459	162 900	455 740	-44	12.7
Beans	10 627	20 941	22 508	106 433	900	3.0
Pea	83 800	35 149	18 000	189 048	125	5.3
Radish	96	1 173	1 856	6 112	6 266	0.2
Red beet	40	0	0	60	50	0.001
Cauliflower	94	100	302	65	-31	0.001
Spinach	3 300	3 721	3 753	3 686	11	0.1
Parsley	4 635	216	8 689	34 803	650	1.0
Lettuce	229	77	248	1 090	375	0.03
Carrot	489	209	1 485	2 264	362	0.06

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### Source: TUİK Foreign Trade Statistics

Despite the slight fluctuations in vegetable seed exports of the years, there were not distinctive differences. In 2016, there was 1% decrease in export quantity, but 9% increase in monetary value as compared to previous year. According to 2016 data, world vegetable seed export was 114 661 tons with a monetary value of 3 976 million dollars (Table 1). When the world vegetable seed exports were assessed based on plant species, it was observed that although there were distinctive decreases in squash seed exports in the years 2013 and 2014, squash had the first place with 46% (1 642 370 kg) share in seed exports of the year 2016. Squash seed exports of the years varied between 356 885–1 642 370 kg. Squash seeds were respectively followed by potato (23.3%, 833 020 kg), broad bean (12.7%, 455 740 kg), pea (5.3%, 189 048 kg), bean (3.0%, 106 433 kg) and onion (2.4%, 84 893 kg) seeds. While there was 37% decrease in tomato seed export, 31% in cauliflower and 44% in bean seed export, there were increases in export of the other vegetable seeds as compared to the year 2012.

Except for 2014, there were increases in monetary value of vegetable seed exports. Based on exports of the years, tomato seeds had the first place with a share of 25.5%. With regard to monetary value of the export, tomato seed was followed respectively by cucumber (15.5%), squash (14.8%), melon-watermelon (11.3%), onion (9.3%) and pepper seed (7%) (Table 3). Vegetable seed exports of the countries between the years 2011-2016 are provided in Table 4. With regard to quantity of vegetable seed export of the countries, the USA had the first place with a share of 15.6%. It was respectively followed by the

*Proceedings of the* 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences (ICOFAAS 2019) *Antalya, Turkey, 8-11 November 2019* Netherlands (12.8%), Italy (12%), China (10.9%), France (9.5%), Denmark (9.2%) and New Zealand (9%) (Table 4).

					Change	Part
Species	2012	2013	2014	2016	(%)	(%)
Potato	66 761	1 523 008	2 109 033	392 949	488	1.4
Sweet corn	214 955	376 414	77 651	981 553	356	3.4
Squash	3 009 043	2 212 792	1 401 274	4 253 623	41	14.8
Cucumber	2 450 018	2 661 168	2 007 301	4 469 932	82	15.5
Melon-Watermelon	2 361 988	2 038 768	1 564 330	3 253 792	38	11.3
Tomato	2 411 971	2 729 334	2 123 181	7 328 659	204	25.5
Pepper	418 910	1 100 631	627 211	2 018 306	382	7.0
Eggplant	1 962 590	1 542 899	124 782	373 708	-81	1.3
Okra	2 011	4 700	7 760	51 706	2 471	0.18
Onion	1 440 158	1 709 545	441 234	2 663 246	84	9.3
Carrot	25 923	20 527	40 573	112 877	335	0.4
Lettuce	13 778	12 370	5 480	73 606	434	0.3
Parsley	122 004	4 424	39 646	125 474	3	0.4
Spinach	22 519	21 711	13 551	54 284	141	0.2
Cauliflower	3 868	3 500	11 508	5 759	49	0.02
Red beet	1 549	0	0	1 345	-13	0.004
Radish	36 679	29 781	19 900	104 246	184	0.4
Pea	209 201	84 197	36 968	432 554	107	1.5
Beans	40 921	91 207	90 698	346 833	748	1.2
Broad Bean	1 642 518	756 096	307 500	1 150 957	-29	4.0
Kidney bean	1 071	-	-	83 283	7 676	0.3
Other vege. seeds	584 428	355 414	235 368	462 985	-21	1.6
Total	17 042 864	17 278 486	11 284 949	28 741 677	68.6	100

Table 3. Distribution of monetary value (dollars) of vegetable seed exports based on species

Source: TUİK Foreign Trade Statistics

Countries	2011	2012	2013	2014	2015	2016	Chang	Part
							(%)	(%)
France	8 700	8 084	8 254	8 696	9 454	9 674	11	9.5
Netherlands	10 426	11 596	12 340	13 178	12 364	13 088	25	12.8
USA	17 853	17 626	14 678	14 526	15 007	15 966	-11	15.6
Germany	1 691	1 271	1 429	1 789	1 715	1 914	13	1.8
Hungary	2 200	1 600	1 025	2 045	2 280	2 654	20	2.6
Ukraine	12 900	12 916	7 117	7 226	10 142	-	-22	-
Italy	10 827	10 153	9 548	11 000	12 390	12 212	12	12.0
Denmark	6 985	7 855	10 396	10 465	10 700	9 414	34	9.2
U.Kingdom	1 191	922	1 233	1 148	887	915	-23	0.9
Indonesia	1 453	2 944	3 992	4 788	-	1 767	21	1.7
China	6 280	7 776	7 619	6 041	7 202	11 101	76.8	10.9
Spain	3 108	2 011	887	2 379	2 445	2 248	-28	2.2
New Zeland	7 400	8 4 2 7	8 787	8 683	8 885	9 200	24	9.0
Thailand	3 000	2 037	1 809	2 198	2 500	1 700	-44	1.6
S. Africa	1 392	1 436	1 553	1 502	1 463	1 653	19	1.6
India	6 200	6 589	7 307	5 709	3 130	3 544	-43	3.4
Bulgaria	927	1 804	2 242	2 019	1 890	898	-3	0.8
Sweden	132	345	1 692	2 088	1 475	2 180	1 551	2.1
Turkey	2 544	1 458	1 470	1 174	1 335	1 870	-26	1.8
Total	105 209	106 850	103 378	106 654	105 264	101 998	-3.1	100

Table 4. Distribution of vegetable seed exports (tons) based on countries

Source: International Seed Federation (ISF)

Countries	2011	2012	2013	2014	2015	2016	Change	Part
							(%)	(%)
France	366	349	404	436	409	444	21	12.9
Netherlands	1 146	1 255	1 273	1 361	1 221	1 486	29	43.1
USA	507	529	550	581	624	671	32	19.5
Germany	73	58	66	79	70	84	15	2.4
Hungary	18	11	10	18	16	16	-12	0.5
Ukraine	32	26	16	18	15	-	-53	-
Italy	118	116	121	128	111	115	-3	3.3
Denmark	46	42	62	63	56	54	17	1.6
U.Kingdom	23	21	28	28	23	25	8	0.7
China	105	158	146	176	161	113	7	3.3
Spain	64	51	61	59	56	79	23	2.3
New Zeland	43	57	67	51	49	47	9	1.4
Thailand	80	73	74	71	94	72	-10	2.1
S. Africa	21	26	21	22	25	33	57	0.9
India	29	36	10	44	41	48	65	1.4
Israel	117	105	105	112	131	132	12	3.8
Turkey	18	12	12	18	16	22	22	0.6
Total	2 806	2 925	3 026	3 265	3 118	3 441	22.6	100

**Table 5.** Distribution of monetary value of vegetable seed exports based on countries (million dollars)

Source: International Seed Federation (ISF)

While the was 11% increase in vegetable seed export of the leading exporter country USA between 2011 - 2016, there were also increases in vegetable seed export of the other leading countries the Netherlands (25%), Italy (12%), France (11%), China (76.8%), Denmark (34%) and New Zealand (24%) (Table 4). With regard to monetary value of vegetable seed exports, the Netherlands (43.1%), USA (19.5) and France (12.9%) were again the leading countries (Table 5).

# **Import Quantity and Monetary Value**

World vegetable seed imports varied in years (Table 6 and Table 7).

e Annual change	Monetary Value	Annual change	Quantity	Year
) (%)	(Million Dollars)	(%)	(tons)	
- 52	3 152	-	132 752	2011
47 3	3 247	-4	128 039	2012
36 5	3 436	3	132 529	2013
50 6	3 650	2	135 869	2014
-4	3 521	-2	133 395	2015
28 8	3 828	-6	124 664	2016
55 52	3 6 3 5	2 -2	135 869 133 395	2014 2015

**Table 6.** Quantity and monetary value of world vegetable seed import

Source: International Seed Federation (ISF)

Table 7. Distribution of world vegetable seed import quantity (kg) based on species

	2012	2012	2014	2017	Chan (%)	Part
Species	2012	2013	2014	2016		(%)
Potato	18 966 835	8 040 979	18 823 420	26 386 320	39	88.2
Sweet corn	18 660	55 564	42 555	93 447	402	0.3
Squash	856 815	2 394 383	1 068 720	195 362	-77	0.7
Cucumber	25 007	16 842	12 193	30 383	21	0.1
Melon-Watermelon	75 623	22 425	243 459	37 227	-51	0.1
Tomato	9 582	9 635	5 172	8 439	-12	0.02
Pepper	2 125	1 029	130 778	1 557	-27	0.005
Eggplant	605	452	318	765	26	0.002
Onion	43 099	3 108	11 104	48 970	13	0.2
Carrot	29 273	28 374	13 182	26 964	-8	0.09
Lettuce	11 888	14 934	16 770	44 550	275	0.1
Parsley	63 232	35 685	34 175	114 111	80	0.4
Spinach	373 080	311 384	150 549	527 325	41	1.8
Cauliflower	1 498	997	855	1 543	3	0.005

Total	22 472 805	12 620 999	20 984 143	29 884 230	33	100
Other vegetab. seeds	29 683	12 479	8 068	43 359	46	0.1
Kidney bean	1 000	-	9 300	42 924	4 192	0.1
Broad Bean	89 055	23 514	11	3 000	-96	0.01
Beans	296 268	321 015	272 152	383 716	29	1.3
Pea	1 557 288	1 318 212	126 522	1 870 596	20	6.3
Radish	15 560	7 390	11 270	13 824	-11	0.04
Red beet	6 629	2 598	3 570	9 848	48	0.03

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Source: TUİK Foreign Trade Statistics

According to 2016 data, total vegetable seed import was 124 664 tons with a monetary value of 3 828 million dollars (Table 6). With regard to imported vegetable seeds, potato (26 386 320 kg) had the first place with a share of 88.2%. Potato was respectively followed by pea (6.3%, 1 870 596 kg), spinach (1.8%, 527 325 kg) and bean (1.3%, 383 716 kg) (Table 7).

While there were decreases in squash, melon-watermelon, broad bean, pepper, tomato, carrot and turnip seed imports in 2016, there were distinctive increases especially in sweet corn, lettuce, parsley and kidney bean seed imports (Table 7). With regard to monetary value of vegetable seed imports based on species, it was observed that tomato (33.2%) had the first place and it was respectively followed by potato (14.2%), pepper (8.4%), cucumber (7.6%) and melon-watermelon seeds (7.6%) (Table 8).

					Chan	Part
Species	2012	2013	2014	2016	(%)	(%)
Potato	14 821 698	7 123 648	16 505 141	19 285 929	30	14.2
Sweet corn	632 532	1 733 723	927 866	2 423 636	283	1.8
Squash	10 470 231	13 908 250	5 679 505	5 649 372	-46	4.2
Cucumber	15 990 694	11 705 508	3 235 308	10 375 848	-35	7.6
Melon-W.melon	9 392 236	6 581 640	2 766 265	10 351 508	10	7.6
Tomato	59 656 688	59 076 820	59 076 820	45 095 054	-24	33.2
Pepper	10 022 297	8 923 951	2 219 291	11 378 100	13	8.4
Eggplant	5 116 166	5 495 789	3 569 587	4 851 816	-5	3.6
Onion	2 706 491	376 126	633 442	2 065 514	-24	1.5
Carrot	4 479 523	5 111 516	1 613 058	3 182 167	-29	2.3

Table 8. Distribution of monetary value (dollars) of vegetable seed import based on species

Total	151 811 188	138 841 569	108 003 994	135 843 524	-11	100
Other veg. seeds	4 536 809	3 972 628	2 064 229	3 617 791	-20	2.6
Kidney bean	5 574	-	36 947	142 049	2 548	0.1
<b>Broad Bean</b>	75 337	27 002	217	118 409	57	0.08
Beans	1 712 343	2 084 611	1 803 876	1 964 673	15	1.4
Pea	2 640 979	2 242 899	193 625	2 623 028	-1	1.9
Radish	349 666	272 321	323 412	359 604	3	0.3
Red beet	222 196	118 245	92 824	332 204	49	0.2
Cauliflower	2 775 879	1 944 113	2 005 375	1 901 133	-32	1.4
Spinach	3 421 191	4 357 131	2 140 736	4 573 676	34	3.4
Parsley	308 771	172 339	158 062	434 523	41	0.3
Lettuce	2 473 887	3 613 309	2 958 408	5 117 490	107	3.8

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Source: TUİK Foreign Trade Statistics

According to ISF 2016 data on vegetable seed import quantities, the first 5 countries were ordered as China (13.4%), the USA (12.8%), United Kingdom (12.6%), Netherlands (10.1%) and Italy (7.6%) (Table 9). These leading countries in world vegetable seed export were also taking the leading positions in import (Table 4). For monetary value of the imports (in dollars), the Netherlands (15.4%), USA (12.7%), Mexico (11.4%) and Spain (9.9%) had the leading positions (Table 10). Turkey has a quite small share in world vegetable seed trade. According to ISF data, Turkey had 1.8% share (1 870 ton) in world vegetable seed export in 2016 and shared the 12<sup>th</sup> place with Germany among 19 countries (Table 4). For monetary value of the export, Turkey had 0.6% share (22 milyon dolar) and 16<sup>th</sup> place among 17 countries (Table 5). For world vegetable seed imports, Turkey had 1.1% share (1 067 ton) and 18<sup>th</sup> place among 20 countries (Table 9). For monetary value of the imports, Turkey with 98 million dollars had the 11<sup>th</sup> place among 18 countries (Table 10).

Countries	2011	2012	2013	2014	2015	2016	Cha(%)	Part
USA	15 562	14 616	13 540	15 854	14 190	12 350	-21	12.8
Netherlands	12 253	15 398	17 960	14 796	13 590	9 778	-21	10.1
China	12 291	11 394	11 300	14 088	13 055	13 365	8.7	13.4
Italy	5 856	5 539	5 725	14 292	6 866	7 298	25	7.6
U.Kingdom	5 627	4 162	3 700	6 813	13 688	12 163	116	12.6

Table 9. Distribution of vegetable seed imports (tons) based on countries

Total	102 370	99 827	95 038	105 565	100 192	96 345	-6.1	100
Turkey	1 844	1 468	2 830	1 992	771	1 067	-43	1.1
Australia	2 005	1 599	1 888	2 448	2 170	2 675	33	2.7
Korea	2 253	2 605	2 970	2 547	2 508	2 182	-3	2.3
Greece	2 195	1 945	2 100	2 133	2 055	2 031	-8	2.1
Belgium	2 872	2 726	3 300	3 508	3 000	3 452	20	3.6
India	2 023	1 862	2 555	2 340	3 385	4 040	99	4.2
Hungary	2 309	1 347	900	1 193	1 070	1 084	-54	1.1
Mexico	1 728	2 098	1 580	1 601	1 353	2 005	16	2.1
Russian Fed.	3 691	2 861	2 0 2 0 2 0	1 182	981	1 240	-67	1.3
Pakistan	4 254	4 604	2 540	3 118	3 035	3 110	-27	3.2
Japan	4 793	5 413	5 380	4 703	4 775	4 998	4	5.2
Canada	4050	2 933	3 460	3 491	3 751	4 250	5	4.4
Spain	6 462	7 201	5 300	2 578	2 965	2 217	-66	2.3
Germany	5 913	4 148	2 390	2 913	2 934	2 825	-52	2.9
France	4 389	5 908	3 600	3 975	4 050	4 215	-4	4.4

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Source: International Seed Federation (ISF)

Table 10. Monetar	y value of vegetable seed i	mports based on countries	(million dollars)
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Countries	2011	2012	2013	2014	2015	2016	Cha.(%)	Part(%)
France	150	137	151	154	143	168	12	5.7
Netherlands	330	373	421	436	420	450	36	15.4
USA	318	369	368	383	381	371	16	12.7
Germany	97	90	84	88	74	130	34	4.5
Russian Fed.	70	58	62	75	57	66	-6	2.3
Italy	177	170	188	193	180	198	11	6.8
U.Kingdom	83	70	72	76	73	74	-11	2.5
Mexico	215	221	230	265	295	331	54	11.4
China	114	111	125	152	172	177	55	6.1
Spain	195	197	211	238	214	289	48	9.9
Canada	78	75	81	85	97	113	44	3.9

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Total	2 238	2 401	2 551	2 707	2 648	2 908	29.9	100
Poland	45	50	54	56	46	47	4	1.6
Brazil	64	67	70	77	72	72	12	2.5
Turkey	104	122	119	109	105	98	-6	3.4
Australia	57	53	52	53	45	56	-2	1.9
Japan	94	113	126	121	125	135	43	4.6
Korea	-	73	81	79	76	69	-6	2.4
India	47	52	56	67	73	64	36	2.2

Source: International Seed Federation (ISF)

# CONCLUSION

Seeds, especially the vegetable seeds are significant inputs of agriculture. They are produced with the aid of high technology and they have quite high economic value. There are quite distinctive relationships between seed production levels and agricultural success of the countries. The leading countries of world vegetable seed trade generally do not have export/import deficits or a balanced export/import. Turkey had the last places in world vegetable seed trade. According to 2016 ISF data, monetary value of seed import of Turkey was 98 million dollars and monetary value of seed export was 22 million dollars. So, the export deficit of Turkey in vegetable seed trade is around 76 million dollars. Despite decreasing export deficit in years, international payments for vegetable seeds are still at high levels and still negatively influence export/import balance of the country.

# ACKNOWLEDGEMENT

This work was supported by Van Yuzuncu Yil University Scientific Research Projects Coordination Unit (Project Number: FAP-2019-8529).

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# **Emission of Methane from Enteric Fermentation of Ruminants**

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# Abstract

The methane (CH<sub>4</sub>) gas from monogastric animals constitutes only 5% of total emission. Therefore, when the CH<sub>4</sub> emission is the matter of subject, ruminants should be understood. Enteric fermentation or enteric CH<sub>4</sub> emissions have the greatest share in agriculture-induced greenhouse gas emissions. Annual animal-induced emission is around 7.1 million tons  $CO_2$  (carbon dioxide) and such a value corresponds for 14.5% anthropogenic greenhouse gas emission. For enteric methane emissions from cattle, sheep and goat of the continents between the years 1990 – 2016, the greatest increases were respectively observed as 73.4%, 69.49% and 119.34% in Africa. On the other hand, the greatest rational decreases were respectively observed as 49.7% in Europe, 58.31% and 45.16% in Australia. Considering the global greenhouse gas emissions, increases were observed in enteric emissions of sattle and goats and decreases were observed in enteric emissions of sheep. In this study, greenhouse effect-generating gas emissions of ruminants in years were investigated, significance of enteric methane emissions were put forth and measures to be taken were recommended.

Keywords: World, ruminant, enteric fermentation, emission.

# Introduction

Solar flares and periodic changes in earth's motion and resultant natural factors contributes to global climate change. Besides these natural factors, anthropogenic factors through depletion of ozone layer and greenhouse effect also contribute to global climate change. Especially after Industrial Revolution, direct (through intensive use of fossil fuels) or indirect (through deforestation to open fields to agriculture, anthropogenic effects) changes were observed in natural structure of atmosphere [1]. While the temperature of the globe was largely stable before Industrial Revolution, temperature increased by 0.76°C since 1850 in which temperature measurements started to be taken reliably. In case relevant measures are not taken, temperatures are expected to increase by about 1.8 - 4.0°C and even up to 6.4°C within the present century. In other words, if the global greenhouse gas emissions were not reduced to half of 1990 levels by the year 2050, the dangerous threshold (2°C increase in temperature) will be reached and in turn climate change will generate irreversible impacts [2].

Nitrogen (78.08%), Oxygen (20.95%), Argon (0.93%) and trace quantities of Neon, Helium and Krypton are fixed atmospheric gases without any temporal and spatial changes in

quantity. On the other hand, greenhouse gasses like water vapor (0-4%), carbon dioxide  $(CO_2)$  (0.039%) and trace quantities of methane  $(CH_4)$  (0.00019%), ozone  $(O_3)$  (0.05%) and nitrous oxide  $(N_2O)$  (0.0000325%) may have changing ratio in atmosphere based on air circulations, changing temperatures and emissions of various sectors [3]. Thusly, since the Industrial revolution, with the impacts of anthropogenic activities, atmospheric CO<sub>2</sub>, CH<sub>4</sub>, and N<sub>2</sub>O contents increased respectively by 31\%, 200\% and 17\% [4].

Greenhouse gasses differentiate in their duration of stay in atmosphere and heat they absorb [3]. Global warming potential (GWP) was developed to determine differences in greenhouse gas emissions and heat absorption capability and in turn to make comparisons and to take relevant measures [5]. In this sense, GWP values were indicated as 1 for CO<sub>2</sub>, 25 for CH<sub>4</sub> and 310 for N<sub>2</sub>O. The duration of stay in atmosphere was calculated as 120 years for CO<sub>2</sub>, 12 years for CH<sub>4</sub> and 114 years for N<sub>2</sub>O [3].

Agriculture-induced global emissions including plant and livestock production activities doubled during the last 50 years. Agriculture-originated global gas emission of 2.7 billion tons equivalent CO<sub>2</sub> in 1961 increased to 5.3 billion tons in 2011. In a study covering 2001-2010 period, agriculture-originated gas emissions of the continents were measured as 44% in Asia, 25% in America, 15% in Africa, 12% in Europe and 4% in Australia. For the same period, agricultural gas emissions were composed of 40% from enteric fermentation, 16% from pasture manure, 13% from synthetic fertilizers, 10% from paddy production, 7% from manure management and 5% from agricultural waste and meadow wildfires [6]. Such cases indicate that livestock activities constituted a significant portion of greenhouse gas emissions worldwide. Thusly it was indicated in several resources (IPCC, FAO, EPA, etc.) that about 7-18% of anthropogenic global gas emissions came from livestock activities [7]. Annually 7.1 billion tons  $CO_2$  equivalent animal-originated emissions corresponding to 14.5% of anthropogenic greenhouse gas emissions is a quite significant ratio. When such a ratio was assessed based on animal species, it was observed that 41% came from beef cattle and 20% came from dairy cattle operations. In livestock industry, the most important emission sources include feed production (45%), enteric fermentation (30%) and livestock manure (9%) [8]. The primary animal-originated greenhouse gasses are CH<sub>4</sub>, CO<sub>2</sub> and NO<sub>2</sub> [9].

# **1.1. Methane Emission:**

Annual methane emission from natural resources is around 250 million tons. Anthropogenic methane emission is about 320 million tons and such a value constitute about 56% of total methane emission [3]. Methane with a relative ratio of 12-20% in anthropogenic greenhouse gasses constitute a significant factor in global warming [10]. With respect to sectoral contributions to global methane emissions, agriculture has a ratio 50.6%, energy has a ratio 28.7%, waste has a ratio 20.6% and industry has a ratio of 0.1% [11]. The CH<sub>4</sub> gas from monogastric animals constitutes only 5% of total emission [12]. Therefore, when the CH<sub>4</sub> emission is the matter of subject, ruminants should be understood. Mature cattle produce annually about 80-110 kg methane [9]. Such a value seems to be low

considering single cattle, considering 1.4 billion cattle worldwide, animal-originated emissions have significant contributions to world greenhouse gas emissions [11].

In livestock operations, except for  $CO_2$ , the main sources contributing to greenhouse gas emissions are enteric fermentation and manure wastes. These sources are the greatest  $CH_4$  and  $N_2O$  greenhouse gas production points [7].

# **1.2. Enteric Fermentation:**

Methane is produced by symbiotic microorganisms of ruminants, in other words, it is a natural part of the digestive process of ruminants and produced through anaerobic fermentation of feeds coming to rumen [11]. It is released through belching [10]. About 87% of enteric methane emission is originated from the rumen and it is 95% released through belching [12].

It was reported in a study conducted in in the USA in 2011, about 137 million tons methane were produced through enteric fermentation and such a value constituted about 70% of total agricultural methane emission of the country. Of this enteric methane emission, 100 million tons were constituted by beef cattle and this value was 3 times of dairy cattle and 50 times of hogs [3].

Agricultural greenhouse emissions can be classified under the following categories [13]:

Greenhouse gas emissions from agricultural production processes

Nitrogen (N) fertilizer production (CO<sub>2</sub>)

Direct or indirect emissions from animal or chemical fertilizer applications to agricultural lands

Emissions from liquid or solid manure over the pastures

Energy consumption in soil tillage, fertilization, product process, drying, threshing and similar agricultural processes

Product processing as either main or by product

Feed transport from the field

Changes in carbon stocks due to afforestation and

Nitrogen losses related to changes in carbon stocks.

Enteric fermentation of ruminants

Direct or indirect emissions from manure storages

In this study, enteric fermentation-originated greenhouse emissions constituting the greatest share in agricultural emissions will be investigated for Turkey and the world and possible reductions will be discussed.

# 2. Greenhouse Gas Emissions of Ruminants in Turkey

# 2.1. Gas Emission from Enteric Fermentation

In Turkey in 2010, agriculture-originated  $CO_2$  equivalent greenhouse emission (34 864 thousand tons) constituted about 8.5% of greenhouse gas emissions from all sectors (410 058 thousand tons) [14]. According to the statistics of the same year, enteric emissions ( $CO_2$  equivalent) (11 822 thousand tons) from ruminants constituted about 34% of agricultural greenhouse emissions. Such a case indicates the significance of enteric fermentation of ruminants in greenhouse gas emissions.

Table 1. Distribution of enteric fermentation-originated greenhouse gas emissions from the ruminants in Turkey (thousand tons) [14]

		1990		2000		2010		2016	Change
	CH <sub>4</sub>	CO <sub>2</sub> eq	CH <sub>4</sub>	CO <sub>2</sub> eq	CH4	CO <sub>2</sub> eq	CH <sub>4</sub>	CO <sub>2</sub> eq	(%)
Cattle	465.8	9780.8	421.9	8859.2	397.9	8355.3	515.3	10821.1	+10.6
Sheep	218.2	4582.9	151.3	3176.9	109.0	2288.4	157.5	3308.3	-27.8
Goat	59.7	1253.9	38.9	816.3	25.6	538.5	52.1	1093.7	-12.7
TOTAL	743.7	15617.6	612.1	12582.4	532.5	1182.2	724.9	15223.2	-2.5

CH<sub>4</sub>: Methane; CO<sub>2</sub> eq: Carbon dioxide equivalents

Enteric  $CH_4$  gas emissions from ruminants and  $CO_2$  equivalents in Turkey are provided in Table 1. Increase was observed in years only in enteric emissions of cattle and cattle constituted the greatest share. Besides, total metal emission of ruminants decreased by 2.5%.

# 2.2. Gas Emission from Manure Management

According to 2010 data, the emissions from manure management practices of ruminants in Turkey (465 thousand tons) constituted about 7.5% of total emissions from agricultural sector (34 864 thousand tons) [14].

**Table 2.** Distribution of manure management -originated greenhouse gas emissions from the ruminants in Turkey (thousand tons) [14]

	19	1990 2000		201	10	201	Change		
	CH <sub>4</sub>	CO <sub>2</sub> eq	CH <sub>4</sub>	CO <sub>2</sub> eq	$CH_4$	CO <sub>2</sub> eq	CH <sub>4</sub>	CO <sub>2</sub> eq	(%)
Cattle	18.1	483.0	16.3	437.1	15.1	407.2	19.5	525.7	+7.2
Sheep	4.3	91.7	3.0	63.5	2.2	45.8	3.2	66.2	-25.6
Goat	1.3	27.9	0.9	17.9	0.5	11.8	1.1	24.1	-15.4
TOTAL	23.7	602.6	20.2	518.5	17.8	464.8	23.8	616.0	-0.1

	199	0	200	0	201	0	2016	, ,	Change %
	CH4	CO <sub>2</sub> eq	CH <sub>4</sub>	CO <sub>2</sub> eq	CH <sub>4</sub>	CO <sub>2</sub> eq	CH4	CO <sub>2</sub> eq	_ 70
Africa	6.36	133.66	7.66	160.95	9.87	207.28	11.13	233.72	+73.4
Ethiopia	0.98	20.66	1.09	22.90	1.81	38.11	2.02	42.45	
Nigeria	0.45	9.54	0.49	10.34	0.54	11.37	0.67	14.08	
S. Africa	0.42	8.90	0.44	9.17	0.44	9.25	0.43	9.04	
Sudan	0.72	15.16	1.29	27.08	1.51	31.81	1.06	22.37	
America	25.32	531.66	26.84	563.54	29.67	623.06	30.33	637.00	+19.8
Argentina	3.00	62.95	2.76	58.06	2.77	58.27	2.98	62.65	
Brazil	8.54	179.40	9.80	205.51	12.10	254.12	12.54	263.24	
USA	5.83	98.20	5.89	123.78	5.67	119.08	5.57	117.00	
Asia	14.90	312.80	17.32	363.66	18.37	385.70	19.20	403.14	+28.9
China	3.72	78.12	5.02	105.34	4.20	88.18	4.24	89.03	
India	6.41	134.61	6.20	130.23	6.57	137.94	6.53	137.10	
Indonesia	0.49	10.40	0.52	11.02	0.65	13.62	0.77	16.12	
Pakistan	0.61	12.75	0.81	16.92	1.24	26.03	1.52	31.87	
Russia	-	-	2.15	45.17	1.54	32.37	1.40	29.33	
Turkey	0.45	9.78	0.42	8.86	0.40	8.36	0.51	10.82	
Europa	17.94	376.78	11.04	231.84	9.12	191.56	8.90	186.74	-49.7
France	1.55	32.58	1.47	30.89	1.34	28.09	1.32	27.71	
Germany	1.54	32.29	1.11	23.38	0.98	20.60	0.96	20.24	
United Kingdom	0.86	18.07	0.76	16.29	0.69	14.43	0.68	14.30	
Oceania	2.05	42.97	2.41	50.57	2.43	51.04	2.36	49.51	+20
Australia	1.44	30.23	1.72	36.13	1.65	34.66	1.55	32.45	
New Zealand	0.56	11.84	0.64	13.46	0.73	15.38	0.77	16.07	
WORLD	66.56	1397.86	65.26	1370.55	69.46	1458.61	71.91	1510.11	+7.9

**Table 3.** Greenhouse gas emission from enteric fermentation of cattle in the world (million tons) [14]

Methane emissions from enteric fermentation of cattle of the continents and  $CO_2$  equivalents are provided in Table 3. As can be inferred from the table, the greatest rational

increase in enteric methane emissions were observed in Africa (73.4%) and Ethiopia was most remarkable African country in this increase. The other continents with rational increase in enteric emissions are respectively ordered as Asia (28.9%), Oceania (20.0%), and America (% 19.8). Contrarily to other continents, there was significant decreases in methane emissions (-49.7%) in Europe. While Europe had the second place in cattle methane emissions in 1990 (with 17.9 million tons), it was identified as the country together with Oceania with the least methane emission (8.9 million tons) in 2016. In general, there was 7.9% increase in global gas emissions.

	1990		2000		2010		2016		Change %
	CH <sub>4</sub>	CO <sub>2</sub> eq	CH <sub>4</sub>	CO <sub>2</sub> eq	CH <sub>4</sub>	CO <sub>2</sub> eq	CH <sub>4</sub>	CO <sub>2</sub> eq	_
Africa	1.03	21.77	1.24	25.93	1.55	32.55	1.75	36.90	+ 69.49
Algeria	0.09	1.86	0.09	1.85	0.11	2.40	0.14	2.95	
Ethiopia	0.11	2.41	0.05	1.15	0.13	2.68	0.15	3.22	
Nigeria	0.06	1.31	0.13	2.73	0.18	3.73	0.21	3.42	
S. Africa	0.16	3.43	0.14	3.00	0.12	2.57	0.12	2.45	
Sudan	0.10	2.17	0.23	4.84	0.26	5.47	0.20	4.26	
America	0.66	13.85	0.47	9.95	0.47	9.77	0.44	9.19	- 33.65
Argentina	0.14	3.00	0.07	1.42	0.08	1.58	0.07	1.56	
Brazil	0.10	2.10	0.07	1.55	0.09	1.82	0.09	1.94	
Asia	1.75	36.75	2.07	43.42	2.22	46.70	2.56	53.75	+ 46.25
China	0.56	11.68	0.66	13.76	0.67	14.07	0.81	17.02	
India	0.24	5.11	0.30	6.24	0.34	7.11	0.32	6.62	
Iran	0.23	4.68	0.27	5.66	0.24	5.00	0.21	4.46	
Pakistan	0.13	2.70	0.12	2.53	0.14	2.91	0.15	3.13	
Russia	-	-	0.11	2.12	0.16	3.33	0.18	3.82	
Europa	2.36	49.60	1.19	25.02	1.04	21.86	1.05	22.02	- 55.61
Spain	0.18	3.82	0.19	4.03	0.15	3.12	0.13	2.68	
Turkey	0.22	4.58	0.15	3.18	0.11	2.29	0.16	3.31	
United Kingdom	0.35	7.36	0.34	7.10	0.25	5.22	0.27	5.70	
Oceania	1.82	38.33	1.29	27.02	0.80	16.91	0.76	15.98	- 58.31
Australia	1.36	28.61	0.95	19.92	0.54	11.44	0.54	11.35	

**Table 4.** Greenhouse gas emission from enteric fermentation of sheep in the world (million tons) [14]

*Proceedings of the* 2<sup>nd</sup> **International Conference on Food, Agriculture and Animal** Sciences (ICOFAAS 2019) *Antalva, Turkev, 8-11 November 2019* 

Sciences (10)		/=01///	incary a,	rankey, e	11110		15			_
New Zealand	0.46	9.72	0.34	7.10	0.26	5.47	0.22	4.63		
WORLD	7.63	160.31	6.26	131.34	6.08	127.89	6.56	137.84	- 14.04	

Methane emissions from enteric fermentation of sheep of the continents and  $CO_2$  equivalents are provided in Table 4. According to the table, the greatest gas emissions were observed in Africa (% 69.49) and Asia (% 46.25) contents and there were decreases in gas emissions of Europe (% 55.61), Australia (% 58.61) and worldwide (% 14.04). The Sudan, Nigeria and Ethiopia played a significant role in increasing gas emissions of Africa.

Table 5. Greenhouse gas emission from enteric fermentation of goat in the world (m	illion tons)
[14]	

	1990		2000		2010		2016		Change %
	CH <sub>4</sub>	CO <sub>2</sub> eq	CH <sub>4</sub>	CO <sub>2</sub> eq	CH <sub>4</sub>	CO <sub>2</sub> eq	CH <sub>4</sub>	CO <sub>2</sub> eq	_
Africa	0.88	18.56	1.18	24.85	1.66	34.97	1.94	40.71	+ 119.34
Kenya	0.05	1.07	0.05	1.05	0.14	2.96	0.13	2.81	
Nigeria	0.12	2.45	0.21	4.46	0.28	5.94	0.37	7.76	
Sudan	0.08	1.60	0.19	4.05	0.22	4.56	0.16	3.29	
America	0.18	3.88	0.17	3.66	0.19	3.89	0.19	3.98	+ 2.58
Asia	1.75	36.77	2.29	48.18	2.60	54.53	2.78	58.38	+ 58.77
China	0.48	10.12	0.74	15.60	0.75	15.82	0.74	15.65	
India	0.57	11.89	0.62	12.97	0.69	14.42	0.67	14.06	
Iran	0.12	2.60	0.13	2.70	0.11	2.25	0.10	2.01	
Pakistan	0.18	3.72	0.24	4.98	0.30	6.29	0.35	7.39	
Turkey	0.06	1.26	0.04	0.82	0.03	0.54	0.05	1.09	
Europa	0.11	2.33	0.09	1.99	0.09	1.84	0.08	1.78	- 23.61
Oceania	0.01	0.31	0.01	0.25	0.02	0.41	0.02	0.45	- 45.16
WORLD	2.95	61.8	3.76	78.9	4.56	95.64	5.01	105.30	+ 70.39

According to table again, there were remarkable increases in rational gas emissions of Africa (119.34%), Asia (58.77%) and America (2.58%), but decreases were remarkable in gas emissions of Australia (45.16%) and Europe (23.61%). In general, there was about 70.39% increase in enteric emissions from goats.

# **3. CONCLUSION**

Enteric CH<sub>4</sub> emissions of the ruminants can be reduced through the use of fertile animals, increasing concentrate feed ratios in ruminant rations, supplementing rations with vegetable oils, use of probiotics [9] and widespread of biogas production from livestock manure [15]. In this way, reductions can be achieved in anthropogenic global greenhouse gas emissions

and energy savings can be achieved through reducing  $CH_4$  release from the livestock manure and such a saving in turn can be used in activities of the livestock operations [12]. It should also be considered that 1 kg concentrate feed production generates greater  $CO_2$  emissions than the roughage production [9].

# ACKNOWLEDGEMENT

This work was supported by Van Yuzuncu Yil University Scientific Research Projects Coordination Unit (Project Number: FAP-2019-8532).

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# The Relationship Between Linear Body Measurements with Final Weight in Brahman Cross Cattle

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# Abstract

The research aims to study the relationship between various linear body measurements and Body Condition Score (BCS) with final weight in Brahman Cross cattle at PT. KASA Lampung, Indonesia. The relevance of the research is the importance of finding methods to estimate final weight based on body linear measurements and BCS in order to facilitate the records of final weight in the field. Data is collected at PT. KASA, Tegineneng District, Lampung. The research material was 145 heifer (80 heads) and steer (65 heads). The data obtained were analyzed using simple correlation analysis and multiple regression. As with other studies, all measurements in this study, the measurements in steer is higher than in heifer. Body Height (BH), Chest Circumference (CC), Body Length (BL) and Body Condition Score (BCS) correlate very closely with Final Weight/ FW (r = 0.95; 0.91; 0.86 and 0.88, respectively). Multiple regression for estimating final weight using all variables is FW = -914+1.03CC+1.76BL+4.42BH+40.3BCS (R<sup>2</sup> adj. = 95%). It was concluded that linear body measurements and BCS are correlated strongly with FW, which the strongest predictors in estimating final weight is BH, compared to CC, BL and BCS. Besides that, the use of multiple regression using all variables turned out to produce a very high accuracy with  $R^2$  adj. above 90%.

**Keywords:** Brahman Cross cattle, linear measurements, multiple regression, final weight estimation.

# Introduction

Indonesia is one of the countries in the equator that has air temperature and relative humidity that is enough to give effect to living things in it. With a population of over 200 million, the fulfillment of meat consumption is a major concern for the government. To meet the increasing demand for meat today, the government is importing both meat and cattle that will be fattened domestically. The quite popular cattle that are imported from Australia is Brahman Cross cattle, which are originated from crossing between Brahman cattle and with Shorthorn cattle, Hereford, Angus or with Beefmaster. Cattle from this breed contain Bos indicus and also Bos taurus blood, so they have good growth ability, and

have high adaptability to high temperature and humidity. Cattle imported are generally still young, steer or heifer. Brahman cross steer has red and white skin color. This is due to the cow's parents. Brahman Cross (BX) is a new breed of cattle from the crossing between Brahman cattle (Bos Indicus) with Shorthorn cattle and Hereford or British cow nation (Bos taurus). The proportion of blood from Brahman Cross is 25% Brahman cattle, 25% Hereford cattle (Bos taurus) and 25% Shorthorn cattle (Boss taurus).

PT KASA Lampung is a large beef cattle fattening company in Central Lampung, located in Rengas Village, Tegineneng District. Because the main result of this farm is beef which is fattened in a particular time, then the carcass with high weight is important to note. The aim of the research is to to study the relationship between various linear body measurements and BCS with final weight in Brahman Cross cattle at PT. KASA Lampung, Indonesia. The relevance of the research is the importance of finding methods to estimate final weight based on body linear measurements in order to facilitate the records of body weight in the field.

# **Material and Methods**

# **Data collection**

Data were collected at the research location, namely at PT KASA by doing a total sampling. Data was collected by measuring in the field, namely for final weight and cattle BCS, as well as examining production data recording for CC, BL, BH data. The amount of research material is 145 animals which are divided into two groups, 80 heifers and 65 steers. Weather data that is daily temperature and humidity are recorded based on the recording by BPS weather recording.

# Data analysis

The obtained data was analyzed using Linear and Multiple Regression and Anova with One Way Classification Unbalanced design models, and using Minitab software version 13.1. In data analysis, CC, BH, BL and BCS function as independent variables and FW as dependent variables. To calculate the percentages of accuracy of final weight estimation is done by calculate the difference between the actual final weight and the estimated final weight divided by the actual final weight and then multiplied by 100% [1].

# **Result and Discussion**

# Average linear body size, BCS and final weight in Brahman Cross cattle

Seasonal data shows that the minimum daily temperature is 22°C and the maximum is 33°C with relative humidity at 85-86% during August to October 2019, with rainfall at 0 [2]. Such environmental conditions are similar to conditions in the Brahman Cross home country, Australia. It is likely that Brahman Cross cattle will grow better in cooler areas such as in Rockhampton, Queensland where temperatures are range between 10.7°C (minimum) to 32.2°C (maximum). This because there are more Brahman Cross in brown, which is the result of a cross with Beefmaster. Beefmaster cattle are the first American composite breed (combination of three or more breeds). They were developed by Tom Lasater in south Texas, beginning in 1931. The Beefmaster breed was recognized in by the

USDA in 1954, and today Beefmaster Breeders United is the fifth-largest breed registry in the United States. Average cattle performance in terms of BCS, CC, BH, BL, final weight can be seen in Table 1 below. BCS was measured based on Rasby, Stalker, Funston [3]; Farney, Blasi, Johson, Tarpoff, Wagoner and Weaber [4].

Variables	Sex	Mean ± Sd	Ν
BCS	Heifer	6.845 ± 0.338 **	65
	Steer	$7.09 \pm 0.317$	80
	Overall	$6.96\pm0.35$	145
FW (kg)	Heifer	417.8 ± 46.7 **	65
	Steer	$469.14 \pm 56.58$	80
	Overall	$440.81 \pm 57.23$	145
CC	Heifer	151.29 ± 5.44 **	65
	Steer	$154.91 \pm 9.07$	80
	Overall	$152,91 \pm 7.49$	145
BL	Heifer	132.63 ±5.59 **	65
	Steer	$137.12 \pm 6.60$	80
	Overall	$134.64 \pm 6.44$	145
ВН	Heifer	151.44 ± 5.12 **	65
	Steer	$156.85 \pm 5.77$	80
	Overall	$153.86 \pm 6.04$	145

Table 1. Average Final weight (FW), BCS, CC, BL and BH in the location of the research

\*\* means different significantly between heifer and steer (P<0.001)

For all measurements show that steer are higher than heifer. The results of this study are similar to study as reported by [5] that the growth and final weight of Limousine-Ongole Crossbred and Brahman Cross steer cattle are higher than heifers. Growth Hormone and Testosterone are hormones that work in the protein-anabolic mechanism, which they work in synergy [6].

# Relationship between linear measurements and BCS with FW

The results of statistical analysis for the relationship between all variables with the final weight show high accuracy (P<0.001) and high relationship (R2 adj.>70 %). Table 2 presents correlation coefficients and simple linear regression and multiple regression

functions to estimate the final weight. With these results, all variables can be used to estimate the final weight, especially body height (BH). Likewise, with all the variables used to estimate the final weights obtained high estimation accuracy by using muliple regression. This reslut is the same with the result that is reported by Maylinda, and Busono [7] in Fat Tailed Sheep where by using many variables such as Chest Circumference (CC), Body Height (BH), Body Length (BL) and Tail Circumference (TC) in multiple regression and obtain higher accuracy.

**Table 2.** Result of correlation analysis, linear regression and multiple regression analysis between CC, BH, BL, BCS and FW

Variables as predictors	r	Regression	R <sup>2</sup> (adj) (%)
$CC^1$	0.91 **	FW = - 621 + 6.95 CC **	82.5
BL <sup>2</sup>	0.86 **	FW = - 587 + 7.63 BL **	73.6
BH <sup>3</sup>	0.95 **	FW = -940 + 8.97 BH **	89.5
BCS <sup>3</sup>	0.88 **	FW = - 556 + 143 BCS **	76.8
All variables	I	W = - 914 + 1.03 CC + 1.76 BL + 4.42 BH + 40.3 BCS **	95

\*\* = Accuracy of analysis is highly significant (P<0.001), r = correlation coefficient, <sup>1</sup>Chest Circumference, <sup>2</sup>Body Length, <sup>3</sup>Body Height, <sup>4</sup>Body Condition Score.

Using the multiple regression formula by using all variables (CC, BH, BL, BCS) into the multiple regression formula FW = -914+1.03CC+1.76 BL+4.42BH+40.3BCS, then the difference between actual final weight and estimated final weight is very small, that is 4.4276 %. That value is obtained from by reducing the estimated FW with the actual FW then divided by the actual FW. This value indicates a very small value.

# Conclusion

All variables used in this research has a close and very high relationship with the final weight, especially body height/BH ( $R^2$  adj. = 89.5%). The use of all variables in the multiple regression function results in a very close relationship ang the highest relationship ( $R^2$  adj. = 95%).

Acknowledgement: This work was supported by (1) the Dean of Faculty of Animal Science, University of Brawijaya and (2) the Head of PT. KASA, where is the research conducted.

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# Effects of Humic Acid Applications on Mineral Content of Garden Cress Roots under Heavy Metal Stress

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### Abstract

This study was conducted for determination the effects of humic acid (HA) applications on mineral content of garden cress roots under cadmium stress. Three Cd doses (0, 100 and 200 mg/kg) and four HA levels (0, 10, 15 and 20 ml/l) were used in experiment. As a result of this study Cd stress caused decrease nutrient element content of garden cress roots. HA effects were significant. Also, Parallel to the increased Cd level decreased significantly N, P, K, Ca, Mg, Fe, Cu, Mn, Zn and B but not Cl. However, HA applications improved these elements uptake from the roots except Cl. In addition, with the increasing HA levels, significant increases occurred in these parameters. 20 ml/l was the most effective dose for improving uptake of nutrient elements. In conclusion, HA applications could improve nutrient element content in garden cress roots under cadmium stress.

Keywords: heavy metal stress, humic acid, macro and micro element content

### Introduction

Heavy metals belong to the most hazardous environmental pollutant group in terms of high toxicity and significant amounts released into the environment as a result of anthropogenic processes. Heavy metals can be toxic and lead to death of organisms, depending on the type of metal, oxidation state, pH of the environment, concentration and exposure time. Among environmental pollutants called heavy metals, cadmium (Cd) is a major problem due to the mobility in the plant-soil system. Although Cd is naturally present in small amounts in the environment (0.04 - 0.32mM), the fallout and agricultural applications of phosphate fertilizers and biosolites from some industrial activity have enriched the soil with this element [1]. Cd is particularly dangerous because plants growing in contaminated soils can absorb and accumulate large amounts of Cd in edible tissues without any visible signs, so that they can be found in edible parts of plants [2] and enter the human body by feeding [3]. In a field study conducted in Europe Peris et al. [4] reported that the edible parts of vegetables contain Cd above the specified thresholds. Therefore, low Cd uptake and plant

physiological responses are critical to the long-term safety and protection of agricultural resources and ecosystems.

0, 6, 9 mg L<sup>-1</sup> Cd applications on plant growth and leaf accumulation in a research on garden cress, it was determined that the concentration of Cd increased, the accumulation in the roots and leaves increased [5]. This study was conducted for determination the effects of humic acid (HA) applications on mineral content of garden cress roots under cadmium stress.

### **Materials and Methods**

### **Experiment set up**

This study was carried out in the Research Greenhouses and Laboratories of the Department of Horticulture, Faculty of Agriculture, Atatürk University In 2019. In the research, garden cress (*Lepidium sativum*) was used as plant material.

Seeds were sown into pots filled with 1 liter of garden soil: sand (1: 1 v: v) mixture to a depth of 1-1,5 cm and 10 seeds per pot. Five plants were left in each pot with the same appearance after seedling emergence. 15:10:15 kg of NPK per decare was mixed into the medium (Şalk et al. 2008). The experiment was carried out in 240 pots with 4 different levels of cadmium (0, 100, 200 and 250 mg / kg) × 4 HA levels (0, 10, 15 and 20 ml / L), 3 replications x 5 pots.

# **Heavy Metal Applications**

For heavy metal stress applications, cadmium (CdSO4.8H2O) was mixed with different concentrations (0, 100 and 200 mg / kg) and then incubated for 3 weeks. At the end of the incubation period, cress seeds were sown.

### **Humic Acid Applications**

HA applications were prepared with water in concentrations of 10, 15 and 20 ml L-1 and were applied to the soil three times with one week intervals starting one day before planting. 0 ml of L-1 will be used as control.

### **Root mineral content determination**

The experiment was terminated 50 days after sowing and root mineral contents were determined. The samples of roots have been taken in the harvest period and dried at oven at 68 °C for 48 h and passed 1mm sieve size. The Kjeldahl method and a Vapodest 10 Rapid Kjeldahl Distillation Unit (Gerhardt, Konigswinter, Germany) were used to determine total nitrogen (N). Macro- [phosphorus (P), potassium (K), calcium (Ca) and magnesium (Mg), and micro-elements [iron (Fe), manganese (Mn), zinc (Zn), cupper (Cu) and boron (B)] were determined after wet digestion (HNO3-H2O2 acid mixture (2:3 v/v)) of dried and ground sub-samples in microwave digestion (Bergof Speedwave Microwave Digestion Equipment MWS-2), by using an Inductively Couple Plasma spectrophotometer (Perkin-Elmer, Optima 2100 DV, ICP/OES, Shelton, CT 06484-4794, USA).

### **Results and Discussion**

Cadmium and HA treatments significantly affected the mineral content of garden cress roots. Mineral content of garden cress roots decreased with increasing Cd doses except for Cl. N, P, K, Ca, Mg, Fe, Cu, Mn, Zn and B concentrations at 200 mg/kg Cd reduced by 42, 39, 58, 48, 37, 46, 66, 20, 28 and 43%, respectively (Figure 1, Figure 2).. Similarly, several studies reported that the leaf and root content of mineral elements in lettuce and Indian mustard were negatively affected by the Cd exposure [1, 6, 7]. In one study,; It was found that 0.0; 1.3, 3.0 and 6.0 mg kg-1 Cd applications adversely affected plant growth and yield in lettuce and Cd concentration in edible parts exceeded the acceptable limit [8].

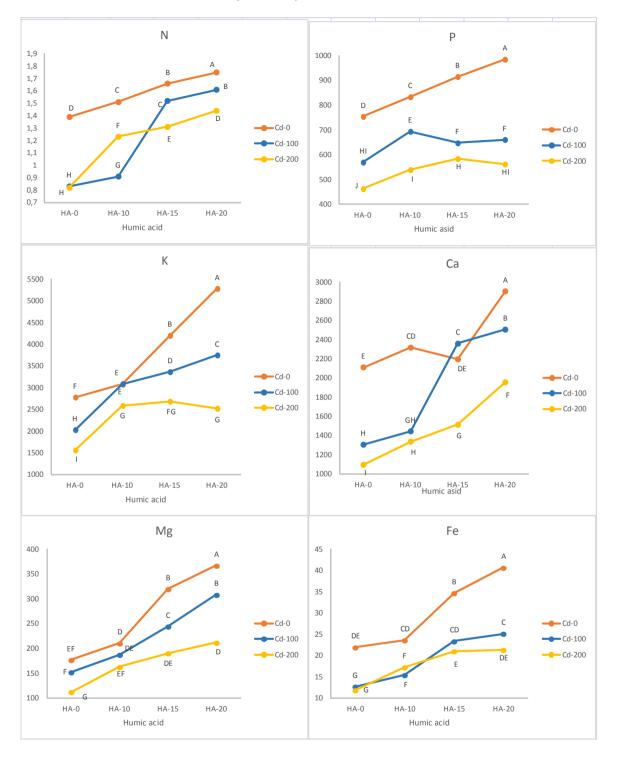
However, HA applications improved the concentrations of the minerals in roots compared to the control treatment. In all Cd treatments, the most effective HA dose for increasing concentration was 20 ml/L. 20 ml/L HA treatment at 200 mg/kg Cd increased N, P, Ca, Mg, Fe, Cu, Zn and B content by 44, 18, 44, 47, 56, 65, 38 and 51%, respectively compared to the control (Figure 1, Figure 2).

Humic substances affect physical, chemical and biological events in soil. In some studies, it has been reported that humic acid and similar organic materials increase the solubility of various nutrients with biochemical soil activities such as respiration, nitrification and nitrogen mineralization [9, 10, 11]. Humic acids help uptake of nutrients by increasing the permeability of cell membranes in plants [12]. Demirtaş et al. [13] reported that HA treatments enhanced macro and micro mineral element content of tomato. Cadmium at 100 and 150 mg kg-1 soil decreased significantly length, fresh and dry weights of shoot and root systems as well as leaf number per plant in both seasons. Similarly, Farouk et al. [14] showed that Cd stress decreased nitrogen, phosphorus and potassium content of radish. On the other hand, application of HA as soil addition increased all mentioned mineral content and decreased cadmium concentrations in plant tissues.

# Conclusions

In conclusion, HA applications could improve nutrient element content in garden cress roots under cadmium stress.

Acknowledgement: This work was supported by Scientific Research Projects Coordination Unit (7219).



Proceedings of the 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences (ICOFAAS 2019) Antalya, Turkey, 8-11 November 2019

Figure 1. Effects of HA applications on N, P, K, Ca, Mg and Fe content of garden cress roots under Cd stress

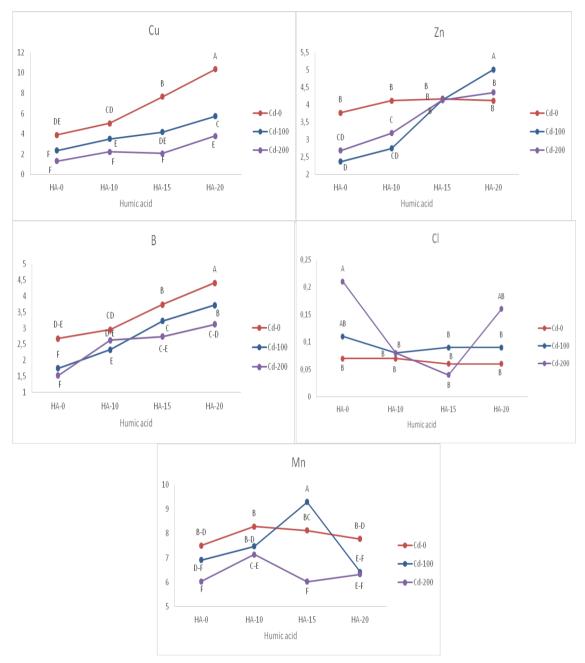


Figure 2. Figure 1. Effects of HA applications on Cu, Zn, B, Cl and Mn content of garden cress roots under Cd stress

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# Effects of Plant Growth Promoting Rhizobacteria (PGPR) and Different Fertilizer Combinations on Macro and Micro Element Contents of Cauliflower (*Brassica oleraceae* L. *botritys*)

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### Abstract

This study was conducted for determination the effects of plant growth promoting rhizobacteria (PGPR) and different (organic and mineral) fertilizer combinations on macro and micro nutrient contents of cauliflower (Brassica oleraceae L. botritys). 16 treatments consisting of PGPR (Paenibacillus polymyxa RC14, Bacillus subtilis RC63 and Pseudomonas fluorescens RC77), organic poultry manure (OPM) (4000 kg/ha), 3 levels of mineral fertilizer nitrogen (N), phosphorus (P) and potassium (K) [NPK1 (80 kg/ha N+50 kg/ ha P+80 kg/ ha K), NPK2 (160 kg/ ha N+75 kg/ ha P+160 kg/ ha K) and NPK3 (200 kg/ ha N+100 kg/ ha P+200 kg/ha K)] and combinations of these applications were used in the study. The experiment was established according to completely randomized block design as four replicates. In the study, the macro (N, P, K, Ca, S and Mg) and micro (Fe, Cu, Mn, Zn, Mo and B) elements of cauliflower leaves were determined. The results of two-year study showed that the highest N, P, K, Ca, S and Mg contents (2.743, 0.353, 0.205, 2.48, 1.525 and 0.210%) were obtained from the NPK2+OPM, PGPR+OPM, NPK3+PGPR, NPK2+PGPR, NPK2+PGPR and NPK1+OPM applications respectively. Also the highest Fe, Cu, Mn, Zn, Mo and B (121.94 ppm, 4.85 ppm, 51.43 ppm, 50,42 ppm, 1,46 ppm and 10,72 ppm) contents were obtained from the NPK2+PGPR, PGPR+OPM, PGPR+OPM, PGPR+OPM, NPK0 and NPK3+PGPR applications. According to the results of the study; the combined use of PGPR and organic poultry manure and their combination with mineral or chemical fertilizers significantly increased the nutrient intake of cauliflower and the effectiveness of mineral fertilizers.

**Keywords:** Cauliflower, PGPR, organic poultry manure, different combinations of fertilizers, macro and micro element content

### Introduction

Plant growth promoting rhizobacteria (PGPR) have been used for improving use efficiency of applied chemical fertilizers [1, 2, 3], biocontrol to pathogens [4, 5] and against abiotic stresses [6, 7]. Excessive use of chemical fertilizers causes environmental pollution [8]. So these fertilizers should be used adequately according to soil analyses. Another way of using less of these chemical fertilizers is to increase the uptake and use efficiency of the nutrients or converting them available forms for taking them by the plants easily.

Cauliflower is a commercially grown vegetable crop worldwide and a member of Brassicaceae family. Cauliflower and broccoli are produced in Turkey 265 815 tones in 11 692 ha area and produced in the world 25 984 758 tones in 1 395 152 ha area [9]. Therefore, Cauliflower is a vegetable crop with high production and consumption and has important ingredients such as fibers, vitamins and antioxidants for health. Cauliflower varieties with high yield have resulted in an increase in cauliflower production but require large amounts of chemical fertilizers which lead to health problems and environmental pollution [10]. In the current study, we investigated how the combined and single use of organic, biological and chemical fertilizers affects macro and micro element profile of cauliflower.

# **Material and Methods**

This study was carried out in the research and experiment area of Yozgat Bozok University Agricultural Faculty in 2014-2015. *Brassica oleracea* var. botritys cv. Tetris F1 was used as plant material. In this study, as PGPR a combination of three bacterial strains (*Pseudomonas putida* RC14, *Bacillus subtilis* RC63 and *Pseudomonas fluorescens* RC77) were used. The bacterial strains were isolated from the rhizospheres of different plants and had nitrogen fixation and phosphate solubilizing properties. As organic manure organic poultry manure was used in the study. Chemical fertilizers were used as three different doses (NPK1: 80-50-80 kg/ha N-P-K, NPK2: 160-75-160 kg/ha N-P-K, NPK3: 200-100-200 kg/ha N-P-K) of nitrogen (N), phosphorus (P) and potassium (K).

Seedlings were planted with 50x60 cm spacing in the middle of July and harvest was made in the early October. After harvesting, the leaf samples from plants were taken and cleaned. The samples were dried at 80° C until constant weight and passed through a 1 mm sieve and prepared for the determination of macro and micro elements (K, P, Ca, Fe, Mg, Cu, Mn, S, Zn, Mo, B) by dry burning method at 550° C in an ash oven. Subsequently, 4 ml of 3 N HCl was added to the samples removed from the ash oven and the samples were filtered with watman filter paper and readings were made in the samples diluted with ultra-pure water. Readings were performed on a Thermo Scientific ICAP-Qc brand ICP-MS. Total N content of plant samples was determined by micro Kjeldahl method after wet burning with salicylic-sulfuric acid.

The experiment was established according to completely randomized block design as four replicates. There were 16 applications in the study shown **Table 1**. Statistical analyses were made according to Duncan's multiple comparison method with using SPSS 16<sup>®</sup> package program.

Aplications (Combinations)							
NPK0 (Control, No fertilizer)							
NPK1							
NPK2							
NPK3							
NPK0+PGPR							
NPK1+PGPR							
NPK2+PGPR							

Table 1. Applications used in the study

NPK3+PGPR
NPK0+OPM
NPK1+OPM
NPK2+OPM
NPK3+OPM
NPK0+PGPR+OPM
NPK1+PGPR+OPM
NPK2+PGPR+OPM
NPK3+PGPR+OPM

#### **Results and Discussion**

The effects of treatments on macro and micro element content of cauliflower were found different. The highest nitrogen (N) contents were determined in NPK3+PGPR (2,647%), NPK2+OPM (2,743%), NPK1+PGPR+OPM (2,670%) and NPK3+PGPR+OPM (2,640%) applications (Table 2). As it can be understood from these findings, high nitrogen content could be obtained from high chemical fertilizer doses as well as organic and biological fertilizers in combination with lower chemical fertilizer doses. The highest phosphorus content was determined in NPK0+PGPR+OPM (0,353%) treatment. This shows that the combination of PGPR+OPM without the synthetic fertilizer could significantly increase the phosphorus content. In another study, the lowest phosphorus content was obtained from the control [10] as shown in our study. The results of the study demonstrated that the highest potassium (K) contents were found in NPK3+PGPR (0,205%) and NPK0+PGPR+OPM (0,201%) applications. It is reported that application of recommended or lower levels of N and P combined with bacteria may increase K content.

Calcium (Ca), sulfur (S) and magnesium (Mg) contents were determined the highest in NPK2+PGPR (2,48%), NPK2+PGPR (1,525%) and NPK1+OPM (0,210%) respectively. Here it is seen that Ca content increases with bacterial and the other applications compared to control, in other study, it has been reported that the Ca content of cauliflower increases with bacterial applications [11]. The vegetables belonging to Brassicaceae family receive their unique aroma and flavor from the sulfurous compounds they contain. Therefore, considering that cauliflowers are rich in sulfur, NPK2xPGPR can be considered as the most positive contribution in this regard. Obtaining the highest Mg content from the combination of organic manure and the lowest level of chemical fertilizers (NPK1+OPM) showed that this applications. In different studies conducted previously it is indicated that the organic manure and bacterial treatments could increase Mg content of cauliflower and broccoli [12, 13].

	Macro Elements							
	Ν	Р	K	Ca	S	Mg		
Applications	(%)	(%)	(%)	(%)	(%)	(%)		
1- NPK0 (Control, No								
Fertilizer)	2,073 c	0,243 k	0,153 h	1,53 k	0,620 i	0,160 g		
2- NPK1	2,547 ab	0,300 d	0,159 g	1,86 h	1,103 e	0,158 gł		
3- NPK2	2,067 c	0,253 ik	0,149 h	1,62 j	1,048 f	0,153 h		
4- NPK3	2,490 ac	0,288 e	0,173 f	2,30 b	1,230 d	0,180 de		
5- NPK0+PGPR	2,447 ac	0,263 gi	0,153 h	1,75 i	0,985 g	0,175 e		
6- NPK1+PGPR	2,127 bc	0,308 cd	0,190 bc	2,10 cd	1,300 c	0,153 h		
7- NPK2+PGPR	2,477 ac	0,258 hj	0,193 b	2,48 a	1,525 a	0,178 e		
8- NPK3+PGPR	2,647 a	0,313 c	0,205 a	2,07 de	1,393 b	0,188 bo		
9- NPK0+OPM	2,140 bc	0,253 ik	0,169 f	2,01 ef	1,010 g	0,185 cc		
10- NPK1+OPM	2,400 ac	0,330 b	0,182 de	2,16 c	1,228 d	0,210 a		
11- NPK2+OPM	2,743 a	0,270 fg	0,180 e	2,26 b	1,220 d	0,193 b		
12- NPK3+OPM	2,300 ac	0,250 jk	0,139 i	2,01 ef	1,020 fg	0,168 f		
13- NPK0+PGPR+OPM	2,110 bc	0,353 a	0,201 a	1,96 fg	1,133 e	0,190 bo		
14- NPK1+PGPR+OPM	2,670 a	0,258 hj	0,133 j	1,87 h	0,880 h	0,175 e		
15- NPK2+PGPR+OPM	2,310 ac	0,265 gh	0,173 f	2,32 b	1,140 e	0,180 de		
16- NPK3+PGPR+OPM	2,640 a	0,278 f	0,186 cd	1,92 gh	1,103 e	0,160 g		

### Table 2. N, P, K, Ca, S and Mg contents of cauliflower leaves\*

\* Differences between the means indicated by different letters in the same column are statistically significant ( $p \le 0.05$ ).

The effects of the applications on the micro element content of cauliflower were also different (**Table 3**). The iron (Fe) content was determined as the highest in the applications of NPK3 (119,16 ppm) and NPK2+PGPR (121,94 ppm). Rhizosphere bacteria that promote plant growth are known to convert iron into an available form [14]. In this study, the positive effect of PGPR's on Fe uptake was also observed. The highest copper (Cu, 4,85 ppm), manganese (Mn, 51,43 ppm) and zinc (Zn, 50,42 ppm) contents were found in the same treatment (NPK0+PGPR+OPM). Considering that no chemical fertilizer is used in NPK0+PGPR+OPM application, it could be said that the combination of organic fertilizers and bacteria increases Cu, Mn and zinc contents. The copper, manganese and zinc are important nutrients for the plant growth, chlorophyll formation, photosynthesis and etc. The positive effect of bacterial applications with organic fertilizers on plant growth and development is once again seen here.

Molybdenum was the highest in NPK0 or Control treatment. The reason of this is may be that the plants in this parcel due to the lack of the nutrients, may try to overcome this problem by taking molybdenum which is effective in nutrient uptake. In other words, the high amount of molybdenum in the plants in this parcel can be an indicator of the stress effect that occurs as a result of the lack of fertilizer application. The highest boron (B) was determined in NPK3+PGPR (10,72 ppm) treatment. Cauliflower is a species that is sensitive to boron deficiency. So it is observed that the PGPR applications had positive effect on uptake of boron in cauliflower.

2- NPK174,45 f3,34 g32,74 g34,01 d0,84 h9,49 eg3- NPK287,63 de3,69 cd39,31 c19,47 i1,06 d9,37 gh4- NPK3119,16 a3,79 bc42,29 b21,92 h1,04 de9,72 de5- NPK0+PGPR87,07 de3,31 g34,00 f40,77 b0,98 g9,66 df6- NPK1+PGPR104,15 c3,64 de38,05 d29,20 f0,84 h9,87 cd7- NPK2+PGPR121,94 a2,67 ij39,25 c26,52 g1,12 c10,37 b8- NPK3+PGPR101,81 c2,75 i39,72 c30,88 e1,01 eg10,72 a9- NPK0+OPM85,67 e3,51 f40,13 c39,92 b0,99 fg9,44 fg10- NPK1+OPM90,12 de3,81 b39,46 c36,31 c0,83 h9,12 h11- NPK2+OPM112,27 b2,92 h42,53 b26,13 g1,03 df10,05 c12- NPK3+OPM104,29 c4,85 a51,43 a50,42 a1,31 b10,59 ab14- NPK1+PGPR+OPM78,09 f2,57 j33,68 fg40,54 b0,64 k8,78 i		Fe	Cu	Mn	Zn	Мо	B
Fertilizer)69.48 g1,75 133.28 fg36,57 c1,46 a8,19 j2- NPK174,45 f3,34 g32,74 g34,01 d0,84 h9,49 eg3- NPK287,63 de3,69 cd39,31 c19,47 i1,06 d9,37 gh4- NPK3119,16 a3,79 bc42,29 b21,92 h1,04 de9,72 de5- NPK0+PGPR87,07 de3,31 g34,00 f40,77 b0,98 g9,66 df6- NPK1+PGPR104,15 c3,64 de38,05 d29,20 f0,84 h9,87 cd7- NPK2+PGPR121,94 a2,67 ij39,25 c26,52 g1,12 c10,37 b8- NPK3+PGPR101,81 c2,75 i39,72 c30,88 e1,01 eg10,72 a9- NPK0+OPM85,67 e3,51 f40,13 c39,92 b0,99 fg9,44 fg10- NPK1+OPM90,12 de3,81 b39,46 c36,31 c0,83 h9,12 h11- NPK2+OPM112,27 b2,92 h42,53 b26,13 g1,03 df10,05 c12- NPK3+OPM104,29 c4,85 a51,43 a50,42 a1,31 b10,59 ab14- NPK1+PGPR+OPM78,09 f2,57 j33,68 fg40,54 b0,64 k8,78 i	Applications	(ppm)	(ppm)	(ppm)	(ppm)	(ppm)	(ppm)
2- NPK174,45 f3,34 g32,74 g34,01 d0,84 h9,49 eg3- NPK287,63 de3,69 cd39,31 c19,47 i1,06 d9,37 gh4- NPK3119,16 a3,79 bc42,29 b21,92 h1,04 de9,72 de5- NPK0+PGPR87,07 de3,31 g34,00 f40,77 b0,98 g9,66 df6- NPK1+PGPR104,15 c3,64 de38,05 d29,20 f0,84 h9,87 cd7- NPK2+PGPR121,94 a2,67 ij39,25 c26,52 g1,12 c10,37 b8- NPK3+PGPR101,81 c2,75 i39,72 c30,88 e1,01 eg10,72 a9- NPK0+OPM85,67 e3,51 f40,13 c39,92 b0,99 fg9,44 fg10- NPK1+OPM90,12 de3,81 b39,46 c36,31 c0,83 h9,12 h11- NPK2+OPM112,27 b2,92 h42,53 b26,13 g1,03 df10,05 c12- NPK3+OPM104,29 c4,85 a51,43 a50,42 a1,31 b10,59 ab14- NPK1+PGPR+OPM78,09 f2,57 j33,68 fg40,54 b0,64 k8,78 i	1- NPK0 (Control, No						
<b>3. NPK2</b> 87,63 de3,69 cd39,31 c19,47 i1,06 d9,37 gh <b>4. NPK3</b> 119,16 a3,79 bc42,29 b21,92 h1,04 de9,72 de <b>5. NPK0+PGPR</b> 87,07 de3,31 g34,00 f40,77 b0,98 g9,66 df <b>6. NPK1+PGPR</b> 104,15 c3,64 de38,05 d29,20 f0,84 h9,87 cd <b>7. NPK2+PGPR</b> 121,94 a2,67 ij39,25 c26,52 g1,12 c10,37 b <b>8. NPK3+PGPR</b> 101,81 c2,75 i39,72 c30,88 e1,01 eg10,72 a <b>9. NPK0+OPM</b> 85,67 e3,51 f40,13 c39,92 b0,99 fg9,44 fg <b>10. NPK1+OPM</b> 90,12 de3,81 b39,46 c36,31 c0,83 h9,12 h <b>11. NPK2+OPM</b> 112,27 b2,92 h42,53 b26,13 g1,03 df10,05 c <b>12. NPK3+OPM</b> 89,87 de3,55 ef38,28 d29,03 f0,70 j9,18 h <b>13. NPK0+PGPR+OPM</b> 104,29 c4,85 a51,43 a50,42 a1,31 b10,59 ab <b>14. NPK1+PGPR+OPM</b> 78,09 f2,57 j33,68 fg40,54 b0,64 k8,78 i	Fertilizer)	69,48 g	1,751	33,28 fg	36,57 c	1,46 a	8,19 j
4- NPK3119,16 a3,79 bc42,29 b21,92 h1,04 de9,72 de5- NPK0+PGPR87,07 de3,31 g34,00 f40,77 b0,98 g9,66 df6- NPK1+PGPR104,15 c3,64 de38,05 d29,20 f0,84 h9,87 cd7- NPK2+PGPR121,94 a2,67 ij39,25 c26,52 g1,12 c10,37 b8- NPK3+PGPR101,81 c2,75 i39,72 c30,88 e1,01 eg10,72 a9- NPK0+OPM85,67 e3,51 f40,13 c39,92 b0,99 fg9,44 fg10- NPK1+OPM90,12 de3,81 b39,46 c36,31 c0,83 h9,12 h11- NPK2+OPM112,27 b2,92 h42,53 b26,13 g1,03 df10,05 c12- NPK3+OPM104,29 c4,85 a51,43 a50,42 a1,31 b10,59 ab14- NPK1+PGPR+OPM78,09 f2,57 j33,68 fg40,54 b0,64 k8,78 i	2- NPK1	74,45 f	3,34 g	32,74 g	34,01 d	0,84 h	9,49 eg
5- NPK0+PGPR87,07 de3,31 g34,00 f40,77 b0,98 g9,66 df6- NPK1+PGPR104,15 c3,64 de38,05 d29,20 f0,84 h9,87 cd7- NPK2+PGPR121,94 a2,67 ij39,25 c26,52 g1,12 c10,37 b8- NPK3+PGPR101,81 c2,75 i39,72 c30,88 e1,01 eg10,72 a9- NPK0+OPM85,67 e3,51 f40,13 c39,92 b0,99 fg9,44 fg10- NPK1+OPM90,12 de3,81 b39,46 c36,31 c0,83 h9,12 h11- NPK2+OPM112,27 b2,92 h42,53 b26,13 g1,03 df10,05 c12- NPK3+OPM104,29 c4,85 a51,43 a50,42 a1,31 b10,59 ab14- NPK1+PGPR+OPM78,09 f2,57 j33,68 fg40,54 b0,64 k8,78 i	3- NPK2	87,63 de	3,69 cd	39,31 c	19,47 i	1,06 d	9,37 gh
6- NPK1+PGPR104,15 c3,64 de38,05 d29,20 f0,84 h9,87 cd7- NPK2+PGPR121,94 a2,67 ij39,25 c26,52 g1,12 c10,37 b8- NPK3+PGPR101,81 c2,75 i39,72 c30,88 e1,01 eg10,72 a9- NPK0+OPM85,67 e3,51 f40,13 c39,92 b0,99 fg9,44 fg10- NPK1+OPM90,12 de3,81 b39,46 c36,31 c0,83 h9,12 h11- NPK2+OPM112,27 b2,92 h42,53 b26,13 g1,03 df10,05 c12- NPK3+OPM89,87 de3,55 ef38,28 d29,03 f0,70 j9,18 h13- NPK0+PGPR+OPM104,29 c4,85 a51,43 a50,42 a1,31 b10,59 ab14- NPK1+PGPR+OPM78,09 f2,57 j33,68 fg40,54 b0,64 k8,78 i	4- NPK3	119,16 a	3,79 bc	42,29 b	21,92 h	1,04 de	9,72 de
7- NPK2+PGPR121,94 a2,67 ij39,25 c26,52 g1,12 c10,37 b8- NPK3+PGPR101,81 c2,75 i39,72 c30,88 e1,01 eg10,72 a9- NPK0+OPM85,67 e3,51 f40,13 c39,92 b0,99 fg9,44 fg10- NPK1+OPM90,12 de3,81 b39,46 c36,31 c0,83 h9,12 h11- NPK2+OPM112,27 b2,92 h42,53 b26,13 g1,03 df10,05 c12- NPK3+OPM89,87 de3,55 ef38,28 d29,03 f0,70 j9,18 h13- NPK0+PGPR+OPM104,29 c4,85 a51,43 a50,42 a1,31 b10,59 ab14- NPK1+PGPR+OPM78,09 f2,57 j33,68 fg40,54 b0,64 k8,78 i	5- NPK0+PGPR	87,07 de	3,31 g	34,00 f	40,77 b	0,98 g	9,66 df
8- NPK3+PGPR101,81 c2,75 i39,72 c30,88 e1,01 eg10,72 a9- NPK0+OPM85,67 e3,51 f40,13 c39,92 b0,99 fg9,44 fg10- NPK1+OPM90,12 de3,81 b39,46 c36,31 c0,83 h9,12 h11- NPK2+OPM112,27 b2,92 h42,53 b26,13 g1,03 df10,05 c12- NPK3+OPM89,87 de3,55 ef38,28 d29,03 f0,70 j9,18 h13- NPK0+PGPR+OPM104,29 c4,85 a51,43 a50,42 a1,31 b10,59 ab14- NPK1+PGPR+OPM78,09 f2,57 j33,68 fg40,54 b0,64 k8,78 i	6- NPK1+PGPR	104,15 c	3,64 de	38,05 d	29,20 f	0,84 h	9,87 cd
9- NPK0+OPM85,67 e3,51 f40,13 c39,92 b0,99 fg9,44 fg10- NPK1+OPM90,12 de3,81 b39,46 c36,31 c0,83 h9,12 h11- NPK2+OPM112,27 b2,92 h42,53 b26,13 g1,03 df10,05 c12- NPK3+OPM89,87 de3,55 ef38,28 d29,03 f0,70 j9,18 h13- NPK0+PGPR+OPM104,29 c4,85 a51,43 a50,42 a1,31 b10,59 ab14- NPK1+PGPR+OPM78,09 f2,57 j33,68 fg40,54 b0,64 k8,78 i	7- NPK2+PGPR	121,94 a	2,67 ij	39,25 c	26,52 g	1,12 c	10,37 b
10- NPK1+OPM       90,12 de       3,81 b       39,46 c       36,31 c       0,83 h       9,12 h         11- NPK2+OPM       112,27 b       2,92 h       42,53 b       26,13 g       1,03 df       10,05 c         12- NPK3+OPM       89,87 de       3,55 ef       38,28 d       29,03 f       0,70 j       9,18 h         13- NPK0+PGPR+OPM       104,29 c       4,85 a       51,43 a       50,42 a       1,31 b       10,59 ab         14- NPK1+PGPR+OPM       78,09 f       2,57 j       33,68 fg       40,54 b       0,64 k       8,78 i	8- NPK3+PGPR	101,81 c	2,75 i	39,72 c	30,88 e	1,01 eg	10,72 a
11- NPK2+OPM       112,27 b       2,92 h       42,53 b       26,13 g       1,03 df       10,05 c         12- NPK3+OPM       89,87 de       3,55 ef       38,28 d       29,03 f       0,70 j       9,18 h         13- NPK0+PGPR+OPM       104,29 c       4,85 a       51,43 a       50,42 a       1,31 b       10,59 ab         14- NPK1+PGPR+OPM       78,09 f       2,57 j       33,68 fg       40,54 b       0,64 k       8,78 i	9- NPK0+OPM	85,67 e	3,51 f	40,13 c	39,92 b	0,99 fg	9,44 fg
12- NPK3+OPM       89,87 de       3,55 ef       38,28 d       29,03 f       0,70 j       9,18 h         13- NPK0+PGPR+OPM       104,29 c       4,85 a       51,43 a       50,42 a       1,31 b       10,59 ab         14- NPK1+PGPR+OPM       78,09 f       2,57 j       33,68 fg       40,54 b       0,64 k       8,78 i	10- NPK1+OPM	90,12 de	3,81 b	39,46 c	36,31 c	0,83 h	9,12 h
13- NPK0+PGPR+OPM       104,29 c       4,85 a       51,43 a       50,42 a       1,31 b       10,59 ab         14- NPK1+PGPR+OPM       78,09 f       2,57 j       33,68 fg       40,54 b       0,64 k       8,78 i	11- NPK2+OPM	112,27 b	2,92 h	42,53 b	26,13 g	1,03 df	10,05 c
14- NPK1+PGPR+OPM         78,09 f         2,57 j         33,68 fg         40,54 b         0,64 k         8,78 i	12- NPK3+OPM	89,87 de	3,55 ef	38,28 d	29,03 f	0,70 j	9,18 h
	13- NPK0+PGPR+OPM	104,29 c	4,85 a	51,43 a	50,42 a	1,31 b	10,59 ab
	14- NPK1+PGPR+OPM	78,09 f	2,57 j	33,68 fg	40,54 b	0,64 k	8,78 i
<b>15- NPK2+PGPR+OPM</b> 90,83 d 3,50 f 35,78 e 35,04 d 0,83 h 8,66 i	15- NPK2+PGPR+OPM	90,83 d	3,50 f	35,78 e	35,04 d	0,83 h	8,66 i
<b>16- NPK3+PGPR+OPM</b> 88,63 de 2,33 k 34,00 f 26,59 g 0,75 i 7,94 k	16- NPK3+PGPR+OPM	88,63 de	2,33 k	34,00 f	26,59 g	0,75 i	7,94 k

Table 3. Fe.	Cu. Mn. Zn.	Mo and B	contents of	cauliflower leaves
I uble of I c,		, mo una D	contents of	cuuliilo wel leuves

\* Differences between the means indicated by different letters in the same column are statistically significant (p≤0.05).

#### Conclusions

As a result, all PGPR treatments increased nutritional values of cauliflower. Moreover, combination of PGPR and organic manure remarkably increased macro and micro nutrients among the treatments. Cauliflower producers could utilize the current PGPR combinations with organic manure in order to obtain higher nutritional values in cauliflower production.

Furthermore, fertilizers were found more effective when used as combination instead of alone. The use of biofertilizers and organic manures could reduce chemical fertilizer utilization that prevents environmental pollution caused by agrochemicals.

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# Investigation of the Relationship Between Morphological Features of Different Hyacinth Cultivars by Path Analysis

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#### Abstract

Path analysis is included in the group of alternative multivariate statistical methods within a multivariate structure. Path analysis is an extension of the regression model, where we can analyze not just direct effect of the predictors to the response variable, also the indirect effects of whole predictors to the response variable. The purpose of the path analysis is to estimate the importance and amount of hypotheses created for causal relationships between variables. This is well explained by the path diagram. The main objective of the current study investigation of the effect of morphological parameters of the four hyacinth (*Hyacinthus orientalis* L.) cultivars ("Blue Jacket", "Carnegie", "City of Haarlem", and "Jan Bos") grown in the field conditions to the first flowering time by means of path analysis. The data set contains of 123 observations.

According to the results of the study, the leaf length and stalk thickness had the positive significant direct and indirect effects to the first flowering time. Floret length, flower diameter, flower length and floret diameter had the negative significant direct and indirect effects to the first flowering time.

**Keywords:** *Hyacinthus orientalis* L., correlation, morphological parameters, first flowering. time.

### Introduction

Path analysis begins with a model that consists of a system of equations and assumptions about the relationship between a set of variables. The variables can be observed or latent. Path analysis was invented by Sewall Wright over 50 years ago. It makes a distinction between the direct, indirect and total effects of one variable on another. The direct effect is the influence of one variable on another that is not mediated by any other variable that is part of the model. The indirect effect is the effect of one variable on another that is mediated by, or passes through, at least one other variable in the system. The total effect is the sum of the direct and indirect effects [1]. The method of path coefficients has long been a simple,

highly productive method for analysis of genetic and environmental variables in formally closed breeding-population systems [2].

Hyacinth flowers attract attention in terms of form and color, because of easy to grow and production, opening with snow or snow melting at the end of winter, have become one of the most used plants in various countries' parks and gardens. In addition, the sale of cut flowers and bulbs on the market contributes to the economy. Flowering time is one of the most important traits with respect to crop yield [3].

In this study, leaf emergence time (day), leaf number, leaf length (mm), flower length (mm), flower diameter (mm), floret number, floret length (mm), floret diameter (mm), stalk thickness (mm), leaf diameter (mm) parameters were used to calculate direct and indirect effects to the first flowering.

# Material

The data used in the paper consisted of 123 observations that belongs to the four hyacinth (*Hyacinthus orientalis* L.) cultivars ("Blue Jacket", "Carnegie", "City of Haarlem", and "Jan Bos") grown in the field conditions [4].

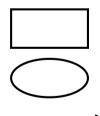
# Method

The primary components of path analysis are

- the path diagram;
- the estimation of path coefficients.

The path diagrams are most useful for the analysis of two or more equations that link observed or latent variables and residual terms to each other in a system. The path diagram is usually easy to interpret and it readily reveals relationships that might be missed if only the equations of a model were examined.

Standard Symbols:



Observed variable

Latent variable

Influence (path) from variable at base to variable at head of arrow

Covariance between a pair of exogenous variables or between a pair of disturbances or errors

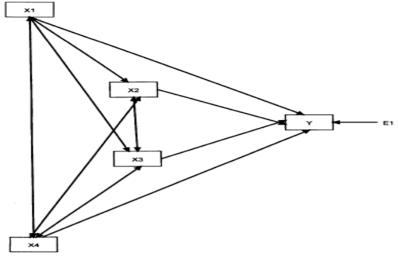


Figure 1. Path diagram for a multiple regression model

Fig.1 shows the standard notational conventions in path diagrams. A basic distinction between the variables is whether they are latent, observed or residual variables. Each observed variable is enclosed in a box. Latent variables appear in ellipses or ovals. Residuals are not usually enclosed in either, although occasionally they are placed in ovals to signify that they are latent [1].

In the path analysis, indirect effect of independent variables plays an important role on the dependent variable [5]. Indirect effect of  $X_i$  (IE<sub>YXi</sub>) on Y can be calculated by the following equation:

$$IE_{YX_i} = r_{X_iX_k}P_{YX_k}$$

where  $r_{X_iX_k}$  is correlation coefficient between X and X<sub>i</sub> variables, P<sub>YXk</sub> is path coefficient of X<sub>i</sub> variable [6]. MINITAB statistical programme was used for analysis [7].

#### **Results and Discussion**

Descriptive statistics concerning all parameters were given in Table 1.

Table 1	Descriptive	statistics	of all	morphole	orical	narameters
I ADIC I.	Descriptive	statistics	or an		Jeicar	parameters

Parameter	Ν	Mean	Std. Deviation	Minimum	Maximum
Leaf Emergence Time (day) (LET)	123	132.93	10.25	33	149
First Flowering Time (day) (FFT)	123	158.49	5.24	149	171
Leaf Number (LN)	123	6.46	0.74	5	9
Leaf Length (mm) (LL)	123	96.46	25.72	44.86	190.86
Flower Length (mm) (FWL)	123	133.95	9.15	62.15	211.86

ciences (ICOFAAS 2019) Antalya, Turkey, 8-11 November 2019							
Flower Diameter (mm) (FWD)	123	61.16	7.98	42.44	77.63		
Floret Number (FN)	123	23.41	9.14	10	59		
Floret Length (mm) (FL)	123	17.78	2.53	11.15	29.19		
Floret Diameter (mm) (FD)	123	24.57	5.93	0	42.60		
Stalk Thickness (mm) (ST)	123	9.02	2.23	3.95	15.23		
Leaf Diameter (mm) (LD)	123	17.28	3.49	9.78	27.30		

*Proceedings of the* 2<sup>nd</sup> **International Conference on Food, Agriculture and Animal Sciences (ICOFAAS 2019)** *Antalya, Turkey, 8-11 November 2019* 

Correlation coefficients between all parameters were given in Table 2.

Parameters	LET	LN	LL	FWL	FWD	FN	FL	FD	ST	LD
LN	-0.15									
LL	0.12	-0.20*								
FWL	0.17	-0.10	0.70**							
FWD	0.09	0.02	0.07	0.14						
FN	0.21*	-0.18*	0.41**	0.54**	-0.10					
FL	-0.04	0.08	-0.10	-0.01	0.52**	-0.25**				
FD	-0.01	0.01	0.17	0.15	0.57**	-0.08	0.31**			
ST	0.16	-0.08	0.60**	0.48**	0.29**	0.59**	-0.06	0.32**		
LD	-0.11	-0.01	0.49**	0.28**	0.27**	-0.04	0.05	0.41**	0.39**	
FFT	0.01	0.01	0.21*	-0.04	-0.50**	0.23**	-0.55**	-0.37**	0.18	-0.02

 Table 2. Correlation matrix between parameters

\*:p<0.05; \*\*:p<0.01(LN-Leaf Number, LL-Leaf Length, FWL-Flower Length, FWD-Flower Diameter, FN-Floret Number, FL-Floret Length, FD-Floret Diameter, ST-Stalk Thickness, LD-Leaf Diameter, FFT-First Flowering Time)

Path analysis results were illustrated in Table 3.

Parameters	LET	LN	LL	FWL	FWD	FN	FL	FD	ST	LD
LET	0.0011	-0.0124	0.0338	-0.0480	-0.0278	-0.0014	0.0105	0.0017	0.0468	0.0017
LN	-0.0002	0.0828	-0.0548	0.0288	-0.0052	0.0012	-0.0253	-0.0024	-0.0226	0.0001
LL	0.0001	-0.0167	0.2720*	-0.1955	-0.0197	-0.0028	0.0301	-0.0326	0.1779	-0.0077
FWL	0.0002	-0.0085	0.1898	-0.2801**	0.0429	-0.0037	0.0018	-0.0297	0.1415	-0.0044
FWD	0.0001	0.0014	0.0180	-0.0405	-0.2967**	0.0007	-0.1559	-0.1134	0.0885	-0.0041
FN	0.0002	-0.0151	0.1115	-0.1518	0.0308	-0.0067	0.0743	0.0151	0.1742	0.0007
FL	-0.0000	0.0069	-0.0271	0.0017	-0.1533	0.0017	-0.3019**	-0.0617	-0.0168	-0.0008
FD	-0.0000	0.0010	0.0449	-0.0422	-0.1703	0.0005	-0.0943	-0.1976*	0.0947	-0.0063
ST	0.0002	-0.0063	0.1626	-0.1332	-0.0883	-0.0039	0.0170	-0.0629	0.2975**	-0.0060
LD	-0.0001	-0.0007	0.1343	-0.0793	-0.0788	0.0003	-0.0154	-0.0804	0.1148	-0.0156

**Table 3.** Direct and indirect effects of investigated traits to the First Flowering Time(FFT)

\*\*:p<0.01;\*:p<0.05; R<sup>2</sup>=0.51(LET-Leaf Emergence Time, LN-Leaf Number, LL-Leaf Length, FWL-Flower Length, FWD-Flower Diameter, FN-Floret Number, FL-Floret Length, FD-Floret Diameter, ST-Stalk Thickness, LD-Leaf Diameter)

According to the regression analysis results the prediction model can be written as follows:

# FFT=0.0011LET+0.0828LN+0.272LL-0.2801FWL-0.2967FWD-0.0067FN-0.3019FL-0.1976FD+0.2975ST-0.0156LD

Direct effects of LET, LN, FN and LD were found non-significant. The correlation coefficient between LL and FFT was positive and significant (p<0.05). The direct effect of LL on FFT was also positive and significant. The correlation coefficients of FWD, FL and FD with FFT were found negative and statistically significant. The direct effects of these parameters to the FFT also were found negative and statistically significant. However, the correlation coefficient of FN with FFT was found positive and statistically significant (p<0.01), the direct effect of FN on the FFT was found negative and non-significant. The correlation coefficient between ST and FFT was found non-significant, however ST had the statistically significant (p<0.01) direct effect on FFT.

### Conclusions

Correlation coefficients have shown that first flowering time was positively and significantly correlated with leaf length and floret number, also negatively and significantly correlated with FWD, FL and FD. The path analysis has shown that LL and ST had the

positive significant direct and indirect effects to the first flowering time. FL, FWD, FWL and FD had the negative significant direct and indirect effects to the first flowering time. These parameters may be useful as predictors of the best first flowering time.

Acknowledgement: This study was prepared by using the figures of some of the data obtained from the master thesis and was supported by Van Yüzüncü Yıl University Scientific Research Projects Coordination (ZF-YL20, 2005).

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# The Effect of Some PGPBs on Post-Harvest Weight, Length and Width of Hyacinth Bulbs Planted in Different Areas

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### Abstract

The aim of this study is to determine the effect of PGPB and chemical NPK 20:20:20 fertilizer applications on the weight, length and width of the bulbs of hyacinth (Hyacinthus orientalis cv. "Delft Blue" plant grown in the laboratory and research-application gardens of Siirt University Faculty of Agriculture. When half and full doses of commercial fertilizer NPK are used, as PGPB, nitrogen-fixing bacteria: Cellulomonas turbata (TV54A), phosphate dissolving bacteria: Bacillus-GC Group (TV119E) and nitrogen fixingphosphate dissolving bacteria: Kluvvera cryocrescens (TV113C) were used. In addition to the individual use of these bacteria, a dual combination of the bacteria TV54A and TV119E was applied. At the end of the study, the highest values of weights and lengths of hyacinth bulbs grown in laboratory conditions in control (37.09 g and 47.67 mm), <sup>1</sup>/<sub>2</sub> (33.79 g and 48.12 mm) and full dose NPK (32.43 and 47.24 mm) applications were obtained while the highest values of the width in the control pots (44.67 mm) were determined and all the average values were found to be statistically significant; When the bacteria were evaluated, all applications were in the same statistical group, but the highest values were obtained from dual combination application on bulb weight, length and width as 23.45 g, 44.03 mm and 36.31 mm (P<0.001), respectively. Weight and length of hyacinth bulbs grown in field conditions at P<0.01 level; widths were statistically significant at P<0.05 level. When the highest values were examined, on the bulb weight, there was no statistically significant difference between control (34.77 g), ½ NPK (32.33 g) and full dose NPK (31.99 g). On the bulb length, the highest values were found on control (47.57 mm) and full dose NPK (46.47 mm). In terms of bulb width, which is the other parameter examined, the highest value was obtained in control (41.54 mm) application. When bacterial applications were evaluated, the highest bulb weight value was determined on phosphate-soluble bacteria: Bacillus-GC Group (TV119E) (28.47 g); the highest bulb length (43.69 mm) and width (40.12 mm) values were found on the Kluyvera cryocrescens (TV113C) applications. The lowest values were observed on the combination of TV54A and TV119E bacteria combination applications in all properties.

**Keywords:** Bulb growth, *Hyacinthus orientalis* L., nitrogen fixing bacteria, ornamental plant, PGPB, phosphate dissolving bacteria.

# Introduction

The population of microorganisms is located around the root zone of the plant, which is usually identified in the soil as the rhizosphere. Physicochemical activities are carried out entirely by these microorganisms in the soil. The majority of microorganisms in the soil are composed of bacteria [1].

In the microorganisms (mycorrhiza, fungus, protozoa, actinomycetes, algae, nematodes, bacteria) that operate in the rhizosphere, the effect of bacteria is high and some of them have beneficial and some of them have harmful effects. Some of the root bacteria have beneficial effects and act as stimulants for plant growth, act as biocontrol agents or act as both in some cases [2]. PGPRs are carried out in laboratory, greenhouse and field environments. However, formation of unfavorable environments in studies conducted in the field such as soil pH differences, temperature rises, decrease in precipitation rates, lack of moisture and nutrient deficiencies lead to a decrease in the colonization of microorganisms [3-4]. The use of plant growth-promoting bacteria (PGPB), which is used as bio-fertilizer in plant nutrition, as a bioagent in plant protection against diseases and for its resistance properties against stress factors with different effect mechanisms, is not as common as in other plant species. These bacteria are used for various purposes and thus new data are gathered. The effect of bacterial inoculation on flower and plant quality is generally investigated in ornamental plants, but there are few studies on bulb quality in bulbous-tuberous ornamental plants other than the studies on flower yield. Flower quality and size of bulbous-tuberous plants depends on the healthy, large and high-quality bulbs.

Hyacinth is a plant used in landscaping and commercially produced as a cut flower. If the development in flowers and bulbs are in the desired levels, the market value of hyacinths increases. In this context, this study is important to reveal how hyacinth bulbs are affected by biological fertilizers made with bacteria or inorganic fertilizers.

### **Material and Method**

This study was carried out in two places: Siirt University Faculty of Agriculture Research Application Garden and Horticulture Laboratory. Commercial hyacinth (*Hyacinthus orientalis* L. Delft Blue) bulbs were used as plant material in the study. Bulbs planted in pots in the laboratory were applied as five bulbs in one repetition for 3 replications. The ones planted in the field were used as 3 replications with 12 bulbs in one repetition. The control group bulbs were not treated in any way. NPK fertilizer as inorganic fertilizer is given to the planting soil as 20:20:20, in full and half-doses, with the irrigation water. PGPBs were selected as nitrogen-fixing bacteria: *Cellulomonas turbata* (TV54A), phosphate-dissolving bacteria: *Bacillus*-GC Group (TV119E), nitrogen-fixing and phosphate-dissolving bacteria: *Kluyvera cryocrescens* (TV113C). These bacteria were used

individually as well as in combination with the TV54A and TV119E bacteria. In the experiment, within the scope of the TÜBİTAK project numbered TOVAG 1080147, nutrient agar (Merck-VM71680604), which was isolated from Van Lake basin and was previously diagnosed with MIS system, was used as a feed-lot for the multiplication of

bacteria, PGPB activity of which was exposed to greenhouse and field conditions. 20 g of nutrient agar was added to one liter of distilled water, adjusted to pH 7.0, and the mixture was sterilized by autoclave for 15 minutes at 121°C. After sterilization, the feed-lots were cooled to 50°C, then transferred to petri plates and allowed to solidify. The stock cultures of the bacteria were planted in nutrient agar medium with the help of the needle and incubated at  $26 \pm 2$ °C for 24 hours [5].

The nutrient broth (Merck-VM775843711) was used as the liquid feed-lot. 8 g of nutrient broth feed-lot was added to one liter of distilled water and pH was adjusted to 7.0. The mixture was sterilized by autoclave for 15 minutes at 121°C and then allowed to cool. A single colony was taken from the bacteria developed in nutrient agar feed-lot and was transferred to nutrient broth feed-lot in aseptic conditions. The bacteria transferred to the liquid feed-lot were incubated at  $26 \pm 2$ °C for 24 hours and at 120 rpm in the horizontal shaker. After incubation, the bacteria concentrations were turbidimetrically adjusted to ~  $10^8$ cfu / ml. The isolates were transferred to hyacinth bulbs which were previously passed through tap water and detergent water and kept for 20 minutes in 5% (v/v) sodium hypochlorite and washed 3 times with pure water. Dried bulbs were planted on the field one day later. The bulbs planted in November 2018 were removed from the place they were planted after the flowering had ended and the vegetative parts had completely dried. The bulbs were weighted with a scale, their lengths and widths were measured by calipers and recorded.

The analysis of the data was done in the SAS 9.1 statistical package program according to randomized plot trial design in laboratory experiments. But field experiments were conducted according to randomized block trial design. LSD multiple comparison test was used for comparing the averages. Tests were performed at  $\alpha = 0.05$  importance level. Descriptive statistics in terms of the traits emphasized; given as average and standard error. The difference between the applications was determined according to independent sample t test [6].

### Results

According to the results of our study, the highest value of hyacinth bulb weight in laboratory applications was obtained in control application with 37.09 gr, while the difference between full dose NPK and 1/2 NPK applications was statistically insignificant (Table 1). The lowest value was obtained with the application of *Cellulomonas turbata* (TV54A) with 21.34 g, while it was in the same group with other bacterial applications. According to the results of the research, inorganic fertilization and bacterial applications tended to decrease bulb weight. In bacterial applications, *Cellulomonas turbata* (TV54A) + *Bacillus*-GC Group (TV119E) showed higher results (23.45 g) than other bacteria (Fig. 1). In a previous study using hyacinth bulbs bacteria have been reported to have different effects on the weight of hyacinth bulbs [7]. Similar results were also observed in this study. These results are thought to be due to the fact that phytohormone and enzymes produced by bacteria result in a decrease in bulb weight.

Laboratory Applications / Properties	Bulb weight*** (g)	Bulb length*** (mm)	Bulb width*** (mm)
Control	37.09 A	47.67 A	44.67 A
Full dose NPK	32.43 A	47.24 A	41.08 B
<sup>1</sup> / <sub>2</sub> NPK	33.79 A	48.12 A	41.17 B
Cellulomonas turbata (TV54A)	21.34 B	41.34 B	34.90 C
Bacillus-GC Group (TV119E)	22.60 B	42.67 B	36.06 C
TV54A + TV119E	23.45 B	44.03 B	36.31 C
Kluyvera cryocrescens (TV113C)	21.57 B	42.08 B	35.24 C
Significant degree	P<0.001	P<0.001	P<0.001

**Table 1.** Effects of npk fertilizer and bacteria applications on bulb weight, length and width on laboratory conditions

\*\*\*: In the same letter, there is no statistically significant 0.1% difference between the averages

Bulb length, which is another characteristic examined under laboratory conditions, was found to be the highest in control and inorganic fertilizer application, but the difference between them was not statistically significant. The highest value was obtained from 1/2 NPK application with 48.12 mm (Fig. 2). The lowest value was obtained from *Cellulomonas turbata* (TV54A) bacterial application with 41.34 mm, whereas the difference between other bacterial applications was statistically insignificant. Among the bacterial applications, the highest value was obtained from the combination of double bacteria (44.03 mm) of *Cellulomonas turbata* (TV54A) + *Bacillus*-GC Group (TV119E). It is thought that the partially high value obtained from the paired combination may be due to the synergistic effect of the two bacteria. Similar results were also reached in the study conducted in the tulip [8].

For the width of hyacinth bulb grown in laboratory conditions, which is another value examined, the highest value was found in the control application with 44.67 mm. The lowest value was obtained from *Cellulomonas turbata* (TV54A) bacterial application (34.90 mm) (Fig. 3). Among the bacteria, the highest value was found in the combination of double bacteria. A similar result was observed in the study conducted by [8] (2019). In light of this result, it would be useful to try a combination of binary and more bacteria rather than single application of bacteria in the future.

Field Applications / Properties	Bulb weight** (g)	Bulb length** (mm)	Bulb width* (mm)
Control	34.77 A	47.58 A	41.54 A
Full dose NPK	31.99 A	46.47 A	40.68 AB
<sup>1</sup> / <sub>2</sub> NPK	32.33 A	43.25 B	40.47 AB
Cellulomonas turbata (TV54A)	26.95 B	42.62 BC	39.40 AB
Bacillus-GC Group (TV119E)	28.47 B	42.78 BC	38.35 AB
TV54A + TV119E	26.41 B	40.68 C	37.28 B
Kluyvera cryocrescens (TV113C)	28.32 B	43.69 B	40.12 AB
Significant degree	P<0.01	P<0.01	P<0.05

**Table 2.** Effects of NPK fertilizer and bacteria applications on bulb weight, length and width on field conditions

\*: In the same letter, there is no statistically significant 5% difference between the averages; \*\*: In the same letter, there is no statistically significant 1% difference between the averages

In the field study, the highest hyacinth bulb weight, length and width values were obtained from the control application, respectively as 34.77 g, 47.58 mm and 41.54 mm (Table 2). The lowest values were obtained from the double bacteria combination of *Cellulomonas turbata* (TV54A) + *Bacillus*-GC Group (TV119E) with 26.41 g, 40.68 mm and 37.28 mm respectively. In terms of bacterial applications, the highest hyacinth bulb weight was obtained in *Bacillus*-GC Group (TV119E) application with 28.47 g (Fig. 1). The highest bulb length and width values were 43.69 mm and 40.12 mm respectively in *Kluyvera cryocrescens* (TV113C) bacteria application (Fig. 2-3). Considering these data, unlike laboratory conditions, the synergistic effect of the double combination under field conditions was not observed and the lowest results were obtained. In addition, higher results were found in single applications. With these results, it is concluded that single bacterial applications are more successful in interaction with other microorganisms in the soil. However, it would be more beneficial to carry out such studies under different ambient conditions and with different bacteria combinations.

*Cellulomonas turbata* (TV54A), *Bacillus*-GC Group (TV119E), TV54A+TV119E and *Kluyvera cryocrescens* (TV113C) were found to have more positive effects on bulb weight according to t test. When the bulb length is considered ½ NPK and TV54A + TV119E bacterial applications in the laboratory while *Cellulomonas turbata* (TV54A) *and Kluyvera cryocrescens* (TV113C) bacteria were found to have significant effects on the field conditions. The width of the control group bulbs, where nothing was applied, was found important in laboratory conditions (Table 3).

Table 3. Effects of applications on bulb weig	t, length and width in field and laboratory
conditions according to t-test	

Applications	ns Areas		Bulb weight(g)	Bulb length(mm)	Bulb width(mm)
	E: -1.1	Average	34.77	47.58	41.54 b
Control	Field	Std. Deviation	1.279	0.345	1.341
Control group	T all a sectors	Average	37.09	47.67	44.67 a
	Laboratory	Std. Deviation	1.976	1.665	0.462
-		t value	-1.700 ns	-0.89 ns	0.262*
		n	6	6	6
	Field	Average	31.99	46.47	40.68
	Field	Std. Deviation	1.345	0.510	0.552
Full Dose NPK	Laboratory	Average	32.43	47.24	41.08
	Laboratory	Std. Deviation	3.061	1.548	1.256
-		t value	-0.231 ns	-0.812 ns	-0.499 ns
		n	6	6	6
	Field	Average	32.33	43.25 b	40.47
	Tielu	Std. Deviation	1.709	1.618	2.747
1/2 NPK	Laboratory	Average	33.79	48.12 a	41.17
	Laboratory	Std. Deviation	1.668	0.897	0.947
-		t value	-1.055 ns	-4.557*	-0.615 ns
		n	6	6	6
	Field	Average	26.95 a	42.62 a	39.40
Cellulomonas turbata	rielu	Std. Deviation	1.336	0.429	4.204
(TV54A)	Laboratory	Average	21.34 b	41.34 b	34.90
	Laboratory	Std. Deviation	2.484	0.177	1.008
-		t value	3.445*	4.812*	1.807 ns
		n	6	6	6
	Field	Average	28.47 a	42.78	38.35
Bacillus-GC	Field	Std. Deviation	0.205	0.745	1.380
Group (TV119E)	Laboratore	Average	22.60 b	42.67	36.06
-	Laboratory	Std. Deviation	2.043	1.075	0.919

_		t value	4.960*	0.153 ns	2.400 ns
		n	6	6	6
	<b>F</b> '.11	Average	26.41 a	40.68 b	37.28
TV54A + TV119E	Field	Std. Deviation	0.357	0.585	0.590
	<b>T 1</b>	Average	23.45 b	44.03 a	36.31
	Laboratory	Std. Deviation	1.205	0.336	1.145
-		t value	4.092*	-4.356*	1.315 ns
		n	6	6	6
	<b>F</b> ' 11	Average	28.32 a	43.69 a	40.12
Kluyvera	Field	Std. Deviation	0.828	0.358	3.034
cryocrescens (TV113C)	<b>T 1</b>	Average	21.57 b	42.08 b	35.24
	Laboratory	Std. Deviation	2.124	0.635	0.524
-		t value	5.133*	3.820*	2.749 ns
		n	6	6	6

Proceedings of the 2 <sup>nd</sup> International Conference on Food, Agriculture and Animal
Sciences (ICOFAAS 2019) Antalya, Turkey, 8-11 November 2019

\*: In the same letter, there is no statistically significant 5% difference between the averages; ns: no significant

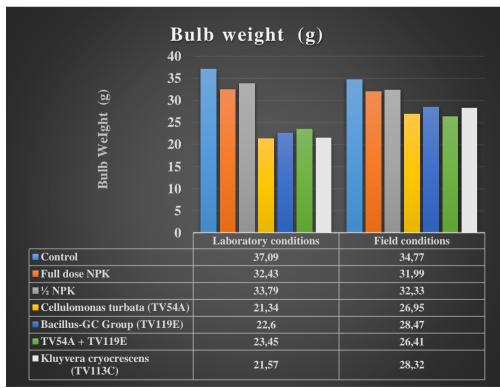


Figure 1. Effects of NPK fertilizer and bacteria applications on bulb weight

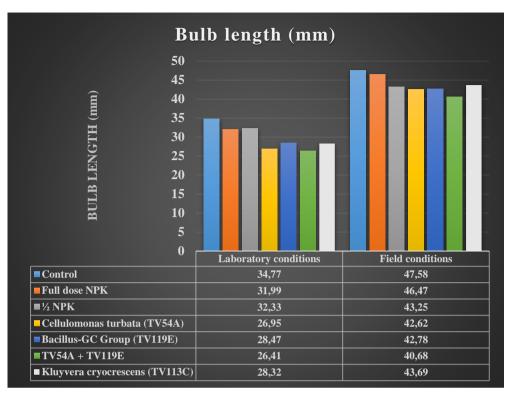


Figure 2. Effects of NPK fertilizer and bacteria applications on bulb length

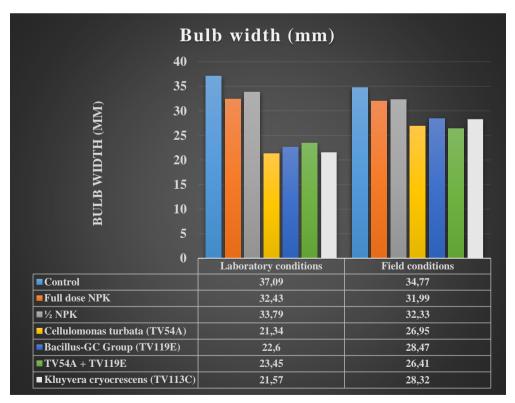


Figure 3. Effects of NPK fertilizer and bacteria applications on bulb width

In the study carried out with natural flower bulbs (*Lilium candidum, Galanthus elwesii* and *Leucojum aestivum*) in Yalova ecological conditions; 0, 5, 10 and 20 kg N applications were performed for each decare in order to determine the effects of different amounts of nitrogen application on bulb size. In the application, it was observed that 10 kg da<sup>-1</sup> N fertilizer had statistically significant effect on bulb weight of lily (*L. candidum*). However, in snowdrops (*G. elwesii*) and loddon lily (*L. aestivum*), nitrogen applications on bulb size were found to be ineffective [9].

Three different types of *Tulipa gesneriana* L. tulip bulbs were planted by being coded (inoculated) with different bacterial formulations. These formulations are: Formulation A (*Pantoea agglomerans* RK-79 + *Pantoea agglomerans* RK-92), Formulation B (*Pantoea agglomerans* RK-79 + *Pantoea agglomerans* RK-92), Formulation TV-91C + *Bacillus subtilis* TV-17C), Formulation C (*Pantoea agglomerans* RK- 79 + *Pantoea agglomerans* RK-79 + *Pantoea agglomerans* RK-79 + *Pantoea agglomerans* RK-79 + *Pantoea agglomerans* RK-79 + *Pantoea agglomerans* RK-79 + *Pantoea agglomerans* RK-79 + *Pantoea agglomerans* RK-79 + *Pantoea agglomerans* RK-92 + *Bacillus megaterium* TV-3D + *Paenibacillus polymyxa* TV-12E) and Formulation D (*Pantoea agglomerans* RK-79 + *Pantoea agglomerans* RK-92 + *Bacillus megaterium* TV-6D + *Pseudomonas putida* TV-42A). In terms of average number of baby bulbs, formulation C was determined as the most successful application. It has been stated that the number and quality of bulbs can be increased with bacterial formulation applications depending on the variety factor and that bacterial applications have benefits to the amount of macro-micro nutrients in soil, tulip bulb and leaf contents [10].

In a study, *Bacillus subtilis* FZB24 bacteria strain was inoculated into saffron (*Crocus sativus* L.) corms. The effect of bacterial inoculation on plant growth and chemical components of stigma was investigated and compared with the control group. During the comparison, it was found that application of *Bacillus subtilis* FZB24 significantly increased leaf length, number of flowers per corm, stigma weight of first flower and total stigma biomass. In addition, it was observed that bulbs treated with PGPR sprouts faster. According to the results obtained from the research, it was found that *B. subtilis* FZB24 application could contribute to saffron cultivation by accelerating corm development (earlier sprouting) and increasing stigma biomass by 12% [11].

A total of ten applications were performed on saffron grown under greenhouse conditions, and the baby corm diameter (mm), baby corm length (mm) and baby corm weight (g) were determined. These applications are: (1) *Achromobacter xylosoxidans* strain TV-42A, (2) *Brevibacillus choshinensis* strain TV-53D, (3) *Myroides odoratus* strain TV-85C, (4) *Bacillus megaterium* strain TV-87A, (5) *Colwellia psycrerytreae* strain TV-108G, (6) *Kluyvera cryocrescens* strain TV- 113C, (7) *Bacillus* GC group B strain TV119E, (8) Control (without bacteria and hormone application) (9) Control 2 [100 mg L<sup>-1</sup> IBA (indole-3 butyric acid)] and (10) Control 3 [100 mg L<sup>-1</sup> GA<sub>3</sub> (gibberellic acid)]. Saffron growth and yield values were higher or equal in some bacterial applications compared to hormone applications. It has been concluded that biofibers used in organic agriculture have a positive effect on the increase of plant growth and development of saffron [12].

In the field study conducted with tulips (*Tulipa gesneriana* L. cv. 'Clear Water') in Faisalabad, *Burkholderia phytofirmans* (PsJN), T2*Bacillus* sp. (MN-54), T3*Enterobacter* sp. (MN-17) and *Caulobacter* sp. (FA-13) isolates were used as foliar fertilizer. The results showed that tulip responds well to bacterial strains and a significant improvement is observed in morphological properties, bulb properties and other quality parameters [13].

The amount of substances stored in the bulb and the size of the bulb, determine the size and quality of the flowers. Criteria that increase the commercial value of bulbs: Their appearance, being disease-free and their size. In terms of development, large bulbs are made possible by vegetative growth, irrigation that promotes bulb development and cultural processes such as eliminating weeds, diseases, pests and providing fertilization. However, the bulbs that are not picked up from the flowers and the bulbs growing from the plants left to the seed remain smaller and the quality of the flowers decreases [14-15].

The size and quality of the flower is directly related to the size and weight of the bulb. The size of the bulb is therefore very important. A good fertilization program ensures the efficiency of the growing environment with irrigation and the effective control of weeds and diseases.

### Conclusions

As a result, it was determined that bacterial applications had negative effects on the bulb weight, length and width in both laboratuary and field conditions, while the highest values were observed in control (without bacterial application and fertilization) plants.

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# Determination of Morphological Properties of Some Long Type Pepper Genotypes

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### Abstract

In this study, 20 long pepper genotypes were used as plant material. In order to determine the morphological characteristics of pepper genotypes, 60 seedlings of each genotype were planted in a plastic greenhouse of Serik (Antalya) Anamas Agriculture Production Company on February 25, 2019. In the study, one characteristic for seedlings (anthocyanin coloration of hypocotyl), one characteristic for flowers (attitude of peduncle), three characteristics for plants (attitude, type of culture and growing season), five characteristics for leaves (leaf length, leaf width, leaf color, leaf blistering and petiole length) and 13 characteristics for fruits [fruit attitude, fruit length (without stalk), fruit diameter, fruit shape of longitudinal section, fruit shape of cross section, fruit glossiness, fruit texture of surface, fruit taste, stalk length, fruit color before maturity, fruit color at maturity, fruit number of locules, main use of fruit] were investigated. In the study, it was determined that anthocyanin coloration of hypocotyl of seedling was present, plant attitude was erect, fruit texture of surface was smooth, stalk length was medium, fruit color before maturity was green, fruit color at maturity was red and the main use was fresh market for all of the 20 pepper genotypes.

# Introduction

In Turkey, 30 032 827 tons of vegetables were produced on an area of 820 668 ha. In this production, pepper vegetables are in the 3rd place with a production share of 2 554 974 tons (1). Pepper is an important vegetable species in the Solanaceae family such as tomato and eggplant (2). Pepper is a type of vegetable belonging to the genus Capsicum which is a one-year plant in warm climates and is a perennial plant in tropical climates (3). The homeland of pepper is the continent of America (4) and Bolivia, Peru, Paraguay, Northern Argentina, South Brazil and Mexico are the gene centers of many pepper species (5).

100 g sweet green pepper contains 92.6-93% water, 1.1-1.2 g protein, 1.4 g cellulose, 0.2 g fat, 9-11 mg calcium, 22-25 mg phosphorus, 0.4-0.7 mg iron, 13 mg sodium and 213 mg potassium (6). The caloric value of pepper is very low (22%). The main importance of pepper in human health and nutrition is due to the vitamin C. 100 g pepper contains 530 IU

of vitamin A, 0.06 g of vitamin B1, 0.02 mg of vitamin B2, 0.31 mg of vitamin B6 and 160 mg of vitamin C (7). In addition, pepper has an effect on human health such as opening appetite, facilitating digestion, strengthening the stomach, preventing bleeding and positively affecting the nervous system (8). Pepper is a food that can be consumed as fresh or cooked and as well as can be used as food and pharmaceutical industry (5).

Pepper fruits differ in shape, color, size, flavor, crust and wall thickness (3). Among pepper types, the long pepper type is widely produced and consumed in Turkey (9). The aim of this work is to determine some morphological characteristics of 20 pepper genotypes which can be used as parent in future for long type pepper breeding.

# Material and Method

20 pepper genotypes were used as plant material in this study. This work was carried out in plastic greenhouse of Anamas Agricultural Production Company (Serik -Antalya) in 2019.

Seeds were planted on 10 January 2019 in seedling viols filled with mixture of peat and perlite. Seedlings were planted on 25 February 2019 with a planting frequency of 100x40 cm with 60 plants from each genotype. After planting, necessary cultural maintenance procedures were suitable performed according to the technique. Morphological observations were carried out on 25 May 2019 in pepper seedlings by selecting 20 plants belonging to each genotype.

Disease tests of genotypes (Resistance to Tobamovirus-Tobacco mosaic virüs Pathotype 0 (TMV:0), Resistance to Tobamovirus-pepper mild mottle virüs Pathotype 1.2 (pMMoV:1.2), Resistance to Tobamovirus-pepper mild mottle virüs Pathotype 1.2.3 (pMMoV:1.2.3), Resistance to Potato Y virüs Pathotype 0 (PVY:0) and Resistance to Tomato spotted wild virüs Pathotype 0 (TSWV:0)) were performed in Akdeniz University, Faculty of Agriculture, Department of Agricultural Biotechnology.

Morphological characterization of pepper genotypes was performed by using the Technical Question Form and the property document for the Differences, Uniformity and Clarity tests (10).

In the study, one characteristic for seedlings (Seedling: Anthocyanin colaration of hypocotyl), one characteristic for flowers (Flower: Attitude of peduncle), three characteristics for plants (Plant: Attitude, type of culture and Growing season), five characteristics for leaves (Leaf: Length, Leaf: Width, Leaf: Color, Leaf: Blistering and Petiole: Length) and 13 characteristics for fruits [Fruit: Attitude, Fruit: Length (without stalk), Fruit: Diameter, Fruit: Shape of longitudinal section, Fruit: Shape of cross section, Fruit: Glossiness, Fruit: Texture of surface, Fruit: Taste, Stalk: Length, Fruit: Color before maturity, Fruit: color at maturity, Fruit: Number of locules, Main use] were investigated.

### **Results and Discussion**

In the study, it was determined that anthocyanin coloration of hypocotyl of seedling was present, plant attitude was erect, fruit texture of surface was smooth, stalk length was medium, fruit color before maturity was green, fruit color at maturity was red and the main use was fresh market for all of the 20 pepper genotypes (Table 1, 2 and 3). 15 of the genotypes used in the study are suitable for greenhouse production and 5 of them are suitable for greenhouse and open field production. In terms of growing season, while 7 genotypes were found suitable for spring, autumn and winter production; 4 genotypes were found as dropping and 16 genotypes were found as semi-dropping in regard to the attitude of peduncle of flower (Table 1).

10 of the genotypes used in the study are seen to be have short, 5 to be have long and 5 to be have medium leaf length. Also in the study, it was determined that 11 genotypes had narrow, 6 genotypes had medium and 3 genotypes had large leaf width. 14 genotypes used in the experiment had green and 6 genotypes had dark-green leaf color. In addition, 14 pepper genotypes had medium leaf blistering and 13 genotypes had medium petiole length (Table 1).

In terms of fruit attitude, 19 genotypes were found to be dropping and in terms of fruit shape of longitudinal section 19 genotypes were found to be narrow triangular. It was determined that 11 tomato genotypes were long in fruit length and 16 genotypes were medium in fruit diameter. In terms of fruit shape of cross section, 14 genotypes were found circle and 6 genotypes were found oval. In terms of fruit glossiness, 10 genotypes were determined as strong, 8 genotypes as medium and 2 genotypes as small characteristics. It was determined that in terms of fruit taste of 20 genotypes used in the study, 18 genotypes had sweet and 2 genotypes had hot characteristics. In addition, 19 genotypes had be equally two and three and 1 genotype had predominantly two fruit number of locules (Table 2 and 3).

It was determined that 14, 4, 4, 4 and 16 genotypes were resistant to Tobamovirus-Tobacco mosaic virüs Pathotype 0 (TMV:0), Tobamovirus-pepper mild mottle virüs Pathotype 1.2 (pMMoV:1.2), Tobamovirus-pepper mild mottle virüs Pathotype 1.2.3 (pMMoV:1.2.3), Potato Y virüs Pathotype 0 (PVY:0) and Tomato spotted wild virüs Pathotype 0 (TSWV:0), respectively (Table 3).

# Conclusion

Morphological characteristics of 20 long type pepper genotypes were identified in the study. According to the results, it was determined that the pepper genotypes can be used as parent in future breeding studies. While the planned works will be terminated as intended, hybrid varieties that can provide the demands of producers and consumers will be offered to the agriculture of the country.

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**Table 1.** Some characteristics of pepper genotypes

Genotype	Seedling:	Plant:	Type of	Growing	Flower:	Leaf:	Leaf:	Leaf: Color	Leaf:	Petiole:
No	Anthocyanin colaration of	Attitude	culture	season	Attitude of	Length	Width		Blistering	Length
	hypocotyl				peduncle					
B1	Present	Erect	G	SAW	SD	Short	Narrow	Green	Weak	Medium
B2	Present	Erect	G	W	SD	Long	Medium	Green	Weak	Medium
B3	Present	Erect	GOF	SSAW	SD	Long	Narrow	Dark-Green	Strong	Short
B4	Present	Erect	GOF	SSAW	SD	Short	Narrow	Dark-Green	Medium	Short
B5	Present	Erect	G	SAW	SD	Short	Medium	Green	Medium	Medium
<b>B6</b>	Present	Erect	G	SAW	SD	Short	Narrow	Green	Medium	Medium
B7	Present	Erect	G	AW	SD	Medium	Narrow	Green	Medium	Long
<b>B8</b>	Present	Erect	G	W	D	Short	Narrow	Green	Weak	Long
<b>B9</b>	Present	Erect	G	SAW	SD	Medium	Large	Green	Medium	Medium
B10	Present	Erect	G	SA	SD	Medium	Narrow	Green	Medium	Medium
B11	Present	Erect	G	SA	SD	Medium	Medium	Green	Medium	Medium
B12	Present	Erect	G	W	SD	Long	Large	Green	Medium	Medium
B13	Present	Erect	G	SSA	SD	Short	Medium	Green	Medium	Long
B14	Present	Erect	G	SAW	SD	Long	Narrow	Green	Medium	Medium
B15	Present	Erect	GOF	SSA	SD	Long	Large	Dark-Green	Medium	Long
B16	Present	Erect	GOF	SSAW	SD	Short	Narrow	Dark-Green	Medium	Medium
B17	Present	Erect	GOF	SSAW	D	Short	Narrow	Dark-Green	Strong	Medium
B18	Absent	Erect	G	SAW	D	Short	Narrow	Dark-Green	Strong	Short
B19	Present	Erect	G	AW	D	Medium	Medium	Green	Medium	Medium
B20	Present	Erect	G	SAW	SD	Short	Medium	Green	Medium	Medium

Abbreviations in the table: G: Greenhouse, GOF: Greenhouse and open field, SAW: Spring Autumn Winter, W: Winter, SSAW: Spring Summer Autumn Winter, AW: Autumn Winter, SA: Spring Autumn, SSA: Spring, Summer Autumn, SD: Semi-Dropping, D: Dropping,

**Table 2.** Some characteristics of pepper genotypes

Genotype No	Fruit: Attitude	Fruit: Length (without stalk)	Fruit: Diameter	Fruit: Shape of longitudinal section	Fruit: Shape of cross section	Fruit: Glossiness	Fruit: Texture of surface	Fruit: Taste	Stalk: Length
B1	Erect	Medium	Medium	Narrow Triangular	Circle	Medium	Smooth	Sweet	Medium
B2	Dropping	Long	Medium	Narrow Triangular	Circle	Strong	Smooth	Sweet	Medium
B3	Dropping	Medium	Medium	Narrow Triangular	Oval	Strong	Smooth	Hot	Medium
B4	Dropping	Medium	Medium	Narrow Triangular	Circle	Strong	Smooth	Sweet	Medium
B5	Dropping	Long	Medium	Narrow Triangular	Circle	Strong	Smooth	Sweet	Medium
B6	Dropping	Long	Small	Narrow Triangular	Circle	Medium	Smooth	Hot	Medium
B7	Dropping	Medium	Big	Narrow Triangular	Oval	Medium	Smooth	Sweet	Medium
B8	Dropping	Long	Medium	Narrow Triangular	Oval	Little	Smooth	Sweet	Medium
B9	Dropping	Long	Medium	Narrow Triangular	Circle	Medium	Smooth	Sweet	Medium
B10	Dropping	Medium	Medium	Narrow Triangular	Circle	Strong	Smooth	Sweet	Medium
B11	Dropping	Long	Medium	Narrow Triangular	Circle	Strong	Smooth	Sweet	Medium
B12	Dropping	Long	Medium	Narrow Triangular	Oval	Little	Smooth	Sweet	Medium
B13	Dropping	Long	Medium	Narrow Triangular	Circle	Medium	Smooth	Sweet	Medium
B14	Dropping	Medium	Medium	Narrow Triangular	Circle	Strong	Smooth	Sweet	Medium
B15	Dropping	Long	Small	Narrow Triangular	Circle	Strong	Smooth	Sweet	Medium
B16	Dropping	Long	Medium	Narrow Triangular	Oval	Medium	Smooth	Sweet	Medium
B17	Dropping	Long	Big	Moderately Triangular	Circle	Strong	Smooth	Sweet	Medium
B18	Dropping	Short	Medium	Narrow Triangular	Oval	Medium	Smooth	Sweet	Medium
B19	Dropping	Medium	Medium	Narrow Triangular	Circle	Medium	Smooth	Sweet	Medium
B20	Dropping	Medium	Medium	Narrow triangular	Circle	Strong	Smooth	Sweet	Medium

**Table 3.** Some characteristics of pepper genotypes

Genotype	Fruit:	Fruit:	Fruit: Number of	Main use	Resistance to	Resistance to	Resistance to	Resistance	Resistance to
No	Color	color at	locules		Tobamovirus-	Tobamovirus-	Tobamovirus-	to Potato Y	Tomato spotted
	before	maturity			Tobacco	pepper mild	pepper mild	virüs	wild virüs
	maturity				mosaic virüs	mottle virüs	mottle virüs	Pathotype	Pathotype 0
					Pathotype 0	Pathotype 1.2	Pathotype 1.2.3	0 (PVY:0)	(TSWV:0)
					(TMV:0)	(pMMoV:1.2)	(pMMoV:1.2.3)		
B1	Green	Red	Equally two and three	Fresh Market	Present	Absent	Absent	Absent	Present
B2	Green	Red	Equally two and three	Fresh Market	Present	Present	Present	Present	Present
B3	Green	Red	Equally two and three	Fresh Market	Absent	Absent	Absent	Absent	Absent
B4	Green	Red	Equally two and three	Fresh Market	Present	Not tested	Not tested	Not tested	Present
B5	Green	Red	Predominantly two	Fresh Market	Present	Absent	Absent	Absent	Absent
B6	Green	Red	Equally two and three	Fresh Market	Present	Not tested	Not tested	Not tested	Present
B7	Green	Red	Equally two and three	Fresh Market	Present	Present	Present	Present	Present
B8	Green	Red	Equally two and three	Fresh Market	Present	Present	Present	Present	Present
B9	Green	Red	Equally two and three	Fresh Market	Absent	Absent	Absent	Absent	Present
B10	Green	Red	Equally two and three	Fresh Market	Present	Not tested	Not tested	Not tested	Present
B11	Green	Red	Equally two and three	Fresh Market	Present	Not tested	Not tested	Not tested	Present
B12	Green	Red	Equally two and three	Fresh Market	Present	Not tested	Not tested	Not tested	Present
B13	Green	Red	Equally two and three	Fresh Market	Present	Not tested	Not tested	Not tested	Present
B14	Green	Red	Equally two and three	Fresh Market	Present	Not tested	Not tested	Not tested	Present
B15	Green	Red	Equally two and three	Fresh Market	Present	Not tested	Not tested	Not tested	Present
B16	Green	Red	Equally two and three	Fresh Market	Absent	Absent	Absent	Absent	Present
B17	Green	Red	Equally two and three	Fresh Market	Absent	Absent	Absent	Absent	Absent
B18	Green	Red	Equally two and three	Fresh Market	Absent	Absent	Absent	Absent	Absent
B19	Green	Red	Equally two and three	Fresh Market	Present	Present	Present	Present	Present
B20	Green	Red	Equally two and three	Fresh market	Absent	Absent	Absent	Absent	Present

# **Evaluation of Isparta Province in Terms of Horticulture**

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# Abstract

Horticulture has an important place in terms of agricultural production of Turkey. Isparta, which has a transition climate between the Mediterranean and Central Anatolia regions, has an ecology suitable for the cultivation of many of the horticultural species. Horticulture is produced in 23% of Isparta's total agricultural land. Approximately 20% of apple production and 5.7% of cherry production in Turkey are performed in Isparta region. The highest table grapes are produced in Yalvaç district and the highest raisin are produced in Senirkent district. In terms of vegetable and cut flower production, the Isparta province plays an important role in eliminating the production deficit in the summer months when greenhouses in the Mediterranean coastline are empty. In recent years, production increases have been observed especially in greenhouse tomatoes, cucumbers and cut flowers in Isparta. In this respect, Isparta has the potential to do much more than the existing production.

Keywords: Horticulture, Isparta, Fruit, Grape, Ornamental Plants, Vegetable

## Introduction

Turkey has ecological conditions that allows the cultivation of a large majority of horticultural species. Because Turkey is surrounded by seas on three sides, it is a country where some tropical climate fruit species can be grown alongside horticultural plants grown in subtropical and temperate climatic conditions [1]. It is seen that the production amount of the species in horticultural plants increases with the development of production techniques and breeding of more yielding new varieties [2] [3]. In this respect, especially the Mediterranean Region take over in terms of horticultural plants growing. In addition to subtropical fruit species, the coastline of the Mediterranean Region is gaining importance in terms of early cultivation of some other temperate fruit species. This region provides a large part of the needs of Turkey's vegetables and ornamental plants. In addition, the horticulture cultivation of our country, especially vegetable and ornamental plants, occupies an important place in exportation and provides considerable foreign exchange input. Isparta, located in the Mediterranean Region, has a transition climate between subtropical climate and temperate climate zone. Isparta province, with the effect of its lakes, allows many horticultural plants to be grown. Indeed, Eğirdir and Gelendost districts located around the Eğirdir lake have the most favorable ecology of Turkey in terms of apple cultivation. Approximately 20% of Turkey's apple production is provided from this region. In terms of cherry cultivation, Uluborlu and Senirkent districts have significant production potential. As well as, it is appear that Isparta ecology is not very suitable for fruit species especially almonds and apricots which are damaged by late spring frosts. However, there are also areas where a significant amount of apricot production can be made with the effect of Eğirdir Lake. In recent years, Isparta has important advances in terms of undergrowth vegetable and ornamental plants. Isparta has become an important production

center in terms of elimination the production deficit of vegetables and ornamental plants, especially in the summer months when greenhouses in the Mediterranean coastline are empty. In this respect, Isparta province and its districts have the potential to produce much more than the current production of the vegetable and the ornamental plants. In later years, production is expected to increase much more. Within the scope of this study, the current status and the potential for development in the following years of Isparta province in terms of horticulture cultivation were evaluated on the basis of districts.

# Agricultural Area and Horticulture Production of Isparta Province

Turkey has approximately 23.2 million ha of cultivable agricultural fields and about 0.8% of this area is located in Isparta (Table 1). Horticultural species are grown on about 18% of Turkey's agriculture area. Approximately 23% of Isparta agricultural areas are reserved for horticulture. It can be said that Isparta province is above the average of Turkey in terms of horticulture production area. While the area covered by fruit orchard in Isparta is 34 827 ha, the area allocated for vegetable cultivation is 5 600 ha. Grapes and ornamental plants are cultivated on 6 747 ha and 65 ha, respectively. Of the total horticultural area of Isparta province, approximately 74% is composed of fruit (including spice plants), 12% is vegetables, 14% is vineyard and 0.1% is ornamental plants. Although the province of Isparta is an important center in terms of fruit cultivation, apples constitute the majority of the production areas.

	Fruit <sup>x</sup>	Vagatabla	Vinyond	Ornamental	Total
	Fruit	Vegetable	Vinyard	Plants	Horticulture
Agriculture Area of Turkey (ha)	3 045 346	820 668	417 041	51 744	4 251 193
Agriculture Area of Isparta (ha)	34 827	5 600	6 747	65	47 239
Isparta's Share in Production Area (%)	1.14	0.7	1.6	1.2	1.1
Production Quantity in Turkey (tons)	16 561 028	30 032 827	3 933 000	1 711 773 663 <sup>y</sup>	50 526 855 <sup>z</sup>
Production Quantity in Isparta (tons)	802 767	162 906	89 220	77 520 000 <sup>y</sup>	1 054 893 <sup>z</sup>
Isparta's Share in Production (%)	4.8	0.5	2.2	4.5	2.1

Table 1.	Horticulture	Production	Area a	and Production	n Ouantit	v in Tur	kev and Is	parta [4].
	1101010000	110000000000000000000000000000000000000			~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~	J		

x Including spice plants

<sup>Y</sup> Production amount of ornamental plants is given in pieces.

<sup>z</sup> Fruit, Vegetable and Grape production amounts.

Turkey has a total horticulture production of 50 526 855 tons consisting fruit, vegetables and grapes. Approximately 2% of this production is provided from Isparta. 76% of the total horticulture production in Isparta is composed of fruit, 15% of vegetables and 9% of grapes. Turkey's total fruit production has been recorded as 16 561 028 tons according to data for 2018. Isparta has a share of 4.8% of this production with 802 767 tons. With regard to vegetable production, Turkey's total vegetable production is about 30 million tons and 0.5% of this production is provided from Isparta. Isparta province has an important production amount in terms of grape production. Turkey's total grape production with 89 220 tons. In recent years, there have been remarkable developments in the production of ornamental plants in Isparta. In 2018, 77 520 000 pieces of cut flowers were produced in Isparta. In this regard, it is seen that

about 4.5% of the Turkey's total production of cut flowers was supplied from Isparta. Isparta has the potential to produce much more in this area. It is thought that Isparta may be an important production center in the elimination of the cut flower production deficit in the summer months when greenhouses are empty especially in the Mediterranean coastline. Climatic conditions of Isparta province are also an important advantage from this perspective.

### **Evaluation of Fruit Production in Isparta Province**

Fruit production values of Isparta province are given in Table 2. Isparta province has ecological conditions that allows the cultivation of temperate climates fruit species. In this respect, production of apple, cherry, peach, apricot, sour cherry, plum and walnut is important in Isparta. Especially apple and cherry production seems to be more remarkable than other fruits. Except these species, there are also orchard where almond, pear, quince and jujube are cultivated at low rates. It is also known that there are olive and pomegranate cultivation in the low altitude areas (Sütcüler). The most cultivated fruit species in Isparta is apple. According to 2018 data, with a 717 401 tons apple production, it provides approximately 20% of Turkey's apple production. In this regard, the most striking areas are Eğirdir and Gelendost (364 703 and 182 736 tons respectively) which are located around Eğirdir Lake. Particularly Boğazova plain located in districts of Eğirdir provides approximately half of Isparta apple production. Yalvaç and Şarkıkaraağaç districts also are remarkable in terms of apple production. The most important reasons of the apple production in Eğirdir and Gelendost districts are suitable ecology, suitable irrigation facilities and storage facilities. It is seen that the producers of Isparta province use modern techniques in apple production. However, the reasons why apple does not take its rightful place especially in exportation should be discussed. In this respect, it is important to introduce new varieties in apple cultivation and to establish orchards with preferred varieties in export. In addition to new varieties in the development of apple cultivation, there are benefits to start new breeding programs for the solution of current problems in our country [5].

	Apple	Cherry	Peach	Apricot	Sour Cherry	Plum	Walnut
DISTRICTS							
Aksu	15 532	65	113	28	16	409	66
Atabey	10 793	1 942	949	282	203	290	138
Eğirdir	364 703	920	2 083	74	182	307	105
Gelendost	182 736	1 536	933	732	504	2 031	70
Gönen	4 508	2 580	150	80	206	231	100
Keçiborlu	3 225	878	140	190	311	294	197
Senirkent	75 001	15 850	1 062	2 584	2 170	958	345
Sütçüler	1 501	292	259	-	137	275	363
Şarkıkaraağaç	19 600	1 367	265	147	663	179	469
Uluborlu	10 381	5 215	14	33	1 688	104	133

Table 2. Fruit production values of Isparta Province (tons) [4].

*Proceedings of the* 2<sup>nd</sup> **International Conference on Food, Agriculture and Animal Sciences** (**ICOFAAS 2019**) *Antalya, Turkey, 8-11 November 2019* 

Yalvaç	19 648	1 083	334	5 018	886	330	196
Yenişarbademli	2 527	808	61	-	98	18	139
Central	7 246	3 739	3 457	132	2 040	577	452
ISPARTA	717 401	36 275	9 820	9 300	9 104	6 003	2 773
TURKEY	3 625 960	639 564	789 457	769 927	184 167	296 878	215 000

Another important species is cherry for Isparta fruit growing. According to 2018 data, cherry production of Isparta province was 36 275 tons. With this amount of production, Isparta supply the 5.7% of Turkey cherry production. Approximately 60% of the cherry production in Isparta province is realized in Senirkent and Uluborlu districts (15 850 and 5 215 tons respectively). Isparta Central and Gönen districts are other areas which are taking attention in cherry cultivation. Especially in orchards located in the high altitude areas of Isparta Yakaören village, late season cherry production provides important advantages. The most important problems of Isparta cherry producers are the insufficiency export and low prices. Also, the high input costs in cherry growing are among the major problems [6]. Increasing export opportunities and incentives is the most important factor for the development of Isparta cherry production [7]. Isparta province provides the 1.2% of Turkey peach production with 9 820 tons of peach production. Approximately half of the peach production in Isparta is produced in Central and Eğirdir districts. Peach production in other districts is quite limited. One of the most important reasons for low peach production in Isparta is that it is not an alternative to apple and cherry cultivation. In addition, the fact that late spring frosts are effective in most parts of Isparta is another factor that limits peach cultivation. Apricot is another species that late spring frosts limit its cultivation. Apricot production was realized as 9 300 tons in Isparta [4]. The majority of the apricot production in Isparta province (82%) was realized in Yalvac and Senirkent districts. Apricot orchards are generally intensive in villages located on the shore of Eğirdir Lake of Yalvaç, Senirkent and Gelendost districts which are not affected by late spring frosts. One of the important fruit species of Isparta province is sour cherry. Isparta supply about 5% of Turkey sour cherry production with 9 104 tons. Senirkent, Uluborlu and Central districts constitute about 65% of the total sour cherry production. According to 2018 data, plum production in Isparta province was 6003 tons. Isparta constitutes a part of 2% of Turkey plum production. About half of this production is carried out in Gelendost and Senirkent districts. It is seen that early flowering plum cultivars are affected by late spring frosts at some years in Isparta province. In Isparta province, early

leafing walnut cultivars are also suffer from late spring frosts in some years. Also, it is seen that winter colds cause damage to young walnut trees in Isparta. However, Isparta province produces 2 773 tons of walnuts. These values constitute approximately 1.2% of the walnuts production of Turkey. It is important to evaluate areas where winter and late frosts are not effective in walnut growing.

### **Evaluation of Grape and Ornamental Plants Production in Isparta Province**

Isparta province grape and ornamental plants production values are given in Table 3. Isparta provide 2.2% of total grape production of Turkey with 89 220 tons. 68% of Isparta grape production evaluated as table, 31% is raisin and 1% is wine. While more than half of the table grapes are produced in Yalvaç, more than half of the grapes for raisin and wine are produced in Senirkent. Apart from Yalvaç, Central, Keçiborlu and Şarkıkaraağaç are other districts that produce significant amounts in table grape production. In recent years, Isparta province realize a significant progress in ornamental plants production. However, all of the ornamental plants production in Isparta province consist of carnation. In this respect, 77 520 000 pieces of carnation flowers are produced in Isparta province. Isparta provide a significant portion of Turkey's carnation production (approximately 13%). It is remarkable that all carnation production is produced in Central district. It can be said that Isparta has the potential to produce much more amount than current ornamental plant production. Isparta province plays an important role in elimination the production deficit in the summer months when the greenhouses are empty in Antalya. The factor that Isparta is close to Antalya is an important advantage for the export of cut flowers.

	Table Grape	Raisin	Vine Grape	Total Grape Production	Carnation	Ornamental Plants
DISTRICTS						
Aksu	-	-	-	-	-	-
Atabey	538	-	-	538	-	-
Eğirdir	182	84	-	266	-	-
Gelendost	529	-	-	529	-	-
Gönen	826	1 105	26	1 957	-	-
Keçiborlu	5 839	37	364	6 240	-	-
Senirkent	2 156	23 970	762	26 888	-	-
Şütçüler	70	-	-	70	-	-
Şarkıkaraağaç	4 262	2 077	-	6 339	-	-
Uluborlu	93	182	-	275	-	-
Yalvaç	35 036	-	-	35 036	-	-
Yenişarbademli	323	-	-	323	-	-
Central	10 759	-	-	10 759	77 520 000*	77 520 000*
ISPARTA	60 613	27 455	1 152	89 220	77 520 000*	77 520 000*
TURKEY	1 945 262	1 524 091	463 647	3 933 000	607 070 350*	1 711 773 663*

Table 3. Grape and ornamental plants production values of Isparta province (tons) [4].

\* Production amount of ornamental plants is given in pieces.

### **Evaluation of Vegetable Production in Isparta Province**

The total vegetable production values of Isparta province are given in Table 4, and the vegetable production values under protected conditions of Isparta province are given in Table 5. The most cultivated vegetable species in Isparta province is tomato. A total of 86 466 tons of tomatoes are produced in Isparta province and 39 176 tons of this production is protected cultivation. In this respect, Isparta Central and Yalvaç districts supply more than half of Isparta's total production with 30 272 and 20 017 tons, respectively. The tomato production values under protected conditions of Central and Yalvaç districts are 21 000 and 12 015 tons, respectively. Approximately 84% of tomato production under protected conditions is carried out in Isparta Central and Yalvaç districts. Another most cultivated species of vegetable in Isparta province is bean. Total bean production is 22 044 tons and 16 120 tons of this production is realized in Senirkent district. The bean production values of Aksu and Yalvaç districts (2 303 and 1 037 tons, respectively) are remarkable. Only 98 tons of bean production in Isparta province (Sütçüler and Atabey districts) carried out under protected conditions.

DISTRICTS	Tomato	Bean	Cucumber	Watermelon	Onion	Melon
Aksu	1 320	2 303	26	-	267	-
Atabey	4 664	240	293	2 800	81	2 250
Eğirdir	1 960	109	216	255	453	200
Gelendost	1 902	52	286	207	87	122
Gönen	5 233	42	24	112	17	80
Keçiborlu	5 966	234	310	1 250	100	1 600
Senirkent	1 890	16 120	255	1 200	45	400
Şütçüler	5 645	267	236	294	174	6
Şarkıkaraağaç	7 360	589	390	1 514	4 850	986
Uluborlu	49	11	3	-	8	-
Yalvaç	20 017	1 037	13 752	864	533	275
Yenişarbademli	188	419	25	45	40	30
Central	30 272	621	345	1 750	327	625
ISPARTA	86 466	22 044	16 161	10 291	6 982	6 574
TURKEY	12 150 000	668 973	18 48 273	4 031 174	2 073 549	1 753 942

Table 4. Vegetable production values of Isparta province (tons) [4].

16 161 tons of cucumber is produced in Isparta province. 85% of this production is realized in Yalvaç (Table 4). As evaluating the cucumber production values, 95% (11 265 tons) of the total 11 890 tons of provincial production is carried out in Yalvaç (Table 5). Watermelon, onion and melon production in Isparta province is also seen to be made in low rates. In this respect, a watermelon production of 10 291 tons is realized and more than half of this production is performed in Atabey, Şarkıkaraağaç and Isparta Central districts (Table 4). On the other hand,

4 850 tons of onion production, which is 6 982 tons in the province, is realized in Şarkıkaraağaç district. It is seen that Isparta has a total melon production of 6 574 tons. Atabey and Keçiborlu districts attracts attention in terms of melon production. Melons, watermelons and onions are seen to remain at very low levels in Isparta compared to the production values of Turkey.

DISTRICTS	Tomato	Cucumber	Lettuce	Pepper	Watermelon	Bean
Aksu	675	-	-	-	-	-
Atabey	2 070	48	140	75	-	40
Eğirdir	210	-	-	-	-	-
Gelendost	1 020	225	-	-	-	-
Gönen	710	-	-	-	-	-
Keçiborlu	45	10	-	5	100	-
Senirkent	-	-	-	-	-	-
Şütçüler	771	192	291	96	-	58
Uluborlu	-	-	-	-	-	-
Yalvaç	12 015	11 265	-	181	-	-
Yenişarbademli	-	-	-	-	-	-
Şarkıkaraağaç	660	-	-	-	-	-
Central	21 000	150	-	-	-	-
ISPARTA	39 176	11 890	431	357	100	98
TURKEY	3 675 950	941 466	50 256	418 627	844 418	48 230

 Table 5. Vegetable production values under protected conditions in Isparta (tons) [4].

In terms of protected vegetable cultivation, there is not a significant cultivation except to tomatoes and cucumbers in Isparta province. According to data from 2018, the protected cultivation of pepper, watermelon and bean in Isparta remained at very low levels compared with the production of Turkey (Table 5). However, when it is considered that there is no protected cultivation in the Mediterranean coastline where July and August are very hot, plateau vegetable production becomes more important in cooler regions such as Isparta [8]. In this respect, Isparta province is also an important potential for the cultivation of other vegetable species except tomatoes and cucumbers.

## Conclusion

Horticulture is produced in 23% of total agricultural areas in Isparta. 76% of horticultural production is made up of fruit, 15% of grape and 8% of vegetable. It is seen that apple and cherry cultivation among fruit species and tomato and cucumber cultivation among vegetable species are remarkable. Carnation production has also increased significantly in recent years in terms of cut flower cultivation. It is foreseen that other ornamental plant species can be grown

in Isparta in the future. The production potential of Isparta province should be evaluated in terms of both vegetable and cut flower cultivation.

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## Determination of Malondialdehyde and Chlorophyll Amounts by Applying Potassium to Salt Stressed Pepper Plant

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#### Abstract

Salt stress, which is one of the abiotic stress factors, is one of the important stress factors that affect the growth of the plant in arid and semi-arid regions and cause decreases in product yield. The saline-exposed plant is unable to perform metabolic activities and its chances of survival may be difficult. While some plants are susceptible to this stress, some varieties can survive by tolerance mechanisms induced by physiological, biochemical and molecular responses. Some measures to be taken against salinity; In addition to the use of plant genotypes with high tolerance to salt stress and improvement of new varieties, some applications can be made to reduce the harmful effect of salinity. Potassium (K) application is one of them.

The aim of this study is to determine whether the plants will get rid of stress by applying different doses of Potassium (K +) to the pepper plant under salt stress. The study was carried out under controlled conditions in 16/8 hour light / dark photoperiod, 25 °C and 70% humid climate room. After the seeds sown in pumice germinated, seedlings consisting of 2 real leaves were taken to hydroponic culture. Hoagland nutrient solution was used in culture in hydroponic solution. The K + in the current Hoagland solution was calculated to be 136 ppm and this was used as control. Other doses were K1 = 116 ppm, K2 = 136 ppm, K3 = 156 ppm, K4 = 176 ppm. The plants were also treated with 100 mM NaCl salt. Sampling for measurements and analyzes was performed on the 20th day of salt application. In these samples, chlorophyll and salt levels of malendialdehyde (MDA) were determined from some biochemical parameters with salt resistance scale. In the results obtained, it was found that K + applications K3 = 156 ppm and K4 = 176 ppm doses were effective in saving the plant from the harmful effect of salt, decreases in MDA amounts of plants and increases in chlorophyll amounts compared to control.

Keywords: Potassium, Scale, MDA, Chlorophyll.

#### Introduction

Stress, plant production, abiotic (low and high temperatures, deficiency or excess of nutrients, air pollution, heavy metals, drought, salinity and radiation) and biotic (virus, bacteria, fungus, etc.) it may cause growth, development and decrease in yield depending on them [1]. Natural and culture-shaped plants can remain under the influence of various stress factors throughout their lives. Salinity is an important problem that negatively affects product yield and quality. In saline conditions, with the decrease in the osmotic potential of the soil solution, the water potential also decreases, thus reducing the water intake of the plant. The decrease in yield in salt stressed plants is due to the direct toxin effect of sodium (Na) and similar cations in medium, but another reason is the deterioration of the ion balance. n soils with high levels of salt stress, Na and Cl ion may lead to deterioration of the ion balance of plants by reducing K,

Ca and N intake [2], [3], [1] [4], [5], [6]. There is a positive effect between the amount of K on the leaf of the plant and the increase in resistance of the plant in salty conditions and the high K + / Na + ratio is proportional to salt [7], [1]. In the case of potassium or calcium deficiency, ozmoregulation in the plant is impaired and enzyme activation is inhibited and metabolism is negatively affected. In such a case, it was stated that the negative effect of the salt was removed and less affected by stress by making potassium supplementation from the outside [8].

#### **Material and Method**

This research was carried out in the climatic room of Van Yuzuncu Yıl University, Faculty of Agriculture, Department of Horticulture, Physiology Laboratory. Demre sharp pepper variety was used in the study.

Elements	App. 1	App. 2	App. 3	App. 4	App. 5
	Control	K1+ NaCl	K2+ NaCl	K3+NaCl	K4+ NaCl
	(ppm)	(ppm)	(ppm)	(ppm)	(ppm)
Nitrogen (N)	186	186	186	186	186
Phosphorus (P)	31	31	31	31	31
Potassium (K)	136	116	136	156	176
Magnesium (Mg)	49,28	49,28	49,28	49,28	49,28
Calcium (Ca)	200	200	200	200	200
Sulfur (S)	66	66	66	66	66
Iron (Fe)	3.3	3.3	3.3	3.3	3.3
Manganese (Mn)	0.031	0.031	0.031	0.031	0.031
Boron (B)	0.205	0.205	0.205	0.205	0.205
Copper(Cu)	0.015	0.015	0.015	0.015	0.015
Zinc (Zn)	0.023	0.023	0.023	0.023	0.023

Table 1. Contents of the nutrient solution used (ppm).

The nutrient solution used was prepared according to [9].

The experiment was conducted in a split air-conditioned climate room and water culture, where the normal atmosphere was achieved. For this purpose, the pepper seeds were sown in 40x25x5 cm foamy germination pots filled with small-grain pumice sieved. The germination containers were placed in a climate chamber with a temperature of  $25 \pm 2^{\circ}$ C and a humidity of 70-80%. The first real leaves of the cotyledon leaves to be horizontal and begin to be seen in the seedlings of irrigation, seedlings to be developed with the Hoagland nutrient solution to be developed better [9]. In the pumice medium, the second real leaves were seeded in water trays in 25x25x18 cm plastic tubs filled with Hoagland nutrient solution. In the pumice environment, the growing seedlings were taken into the water culture in 25x25x18 cm plastic tubs filled with Hoagland nutrient solution. Seedlings were grown in water culture for 14 days and started to be applied when they have 4-5 real leaves. NaCl was added to the Hoagland nutrient solution to provide a

100 mM salt concentration. During the replenishment of the repeated solutions every week, the salt application was maintained at the same concentration. In addition to the salt application, different doses of K were applied in addition to the Hoagland nutrient solution. The amount of K used in the normal Hoagland nutrient solution is 136 ppm. However, in our application, starting from 20 ppm lower dose, Control = 136 ppm, K1 = 116 ppm, K2 = 136 ppm, K3 = 156 ppm, K4 = 176 ppm was applied. Table 1 shows the ppm values of all nutrients in the nutrient solution.

### Assessment with 1-5 Scale

In order to determine the degree of morphological damage in plants, a scale was created. According to the degree of damage in the salt stress test, pepper plants were rated from 1 to 5 based on the following symptoms [10].

- 1: Plants are not affected by salt stress at all (control plants)
- 2: Local yellowing and curling of leaves
- 3: Yellowing of leaves and 25% necrotic spotting
- 4: 50-75% necrotic stain on leaves and deaths
- 5: Severe necrosis of 75-100% in leaves and complete death of plant

### Malondialdehyde analysis

The method defined by Lutts et al. [11] was employed for measuring the amount of malondialdehyde, which is produced as a result of the lipid peroxidation that causes stress-induced damage to cellular membranes. Malondialdehyde (MDA) concentration was determined by using an "extinction" coefficient, which is 155 mM -1 cm -1, expressed as  $\mu$ mol/g fresh weight. The following equation was used in the calculation: MDA= (A 523 - A 600) x volume of the extract (ml) / (155 mM/cm x sample amount)

### Chlorophyll analysis.

Leaf segments, either fresh or frozen at -80°C, were placed in 5 ml of 80% ethanol and heated in a water bath at 80°C for 20 min. Total chlorophyll was evaluated in the alcohol extracts from absorbance readings, using the appropriate extinction coefficient. Chlorophyll content (mg/g fr wt) was calculated as  $1000 \times A654/(39,8 \text{ x sample fr wt})$ , according to Luna et al. [12]

### Assessments

According to the experiment, randomized plots based on the chance of the experiment were 3 replications and each plant was established as 15 plants. Statistical analysis of the data obtained as a result of biochemical analysis of the plant growth parameters were performed by using the SAS Insitue [13] package program according to Duncan multiple comparison test (P<005).

### Results

The pepper plants grown under salt stress was applied at the same time with different doses of potassium.

Application	Scale values
Control	1
K1+Salt	4
K2+ Salt	3
K3+ Salt	2.5
K4+ Salt	2

**Table 2.** Salt resistance scale according to the symptoms in the leaves (1-5 points)

In order to determine the morphological damage that occurs in plants, seedlings were given a score of 1 to 5 as indicated in the scale forming method. When the scale values are examined, the most affected by salt was observed K1 Salt application of in demre pepper plants. This was followed by K2 Salt, K3 Salt, K4 Salt applications.

Lipid peroxidation product that determines stress-induced and cell membrane damage MDA amount and chlorophyll amounts were shown in Table 3. The amount of MDA in the leaves of pepper plants increased in K1 + Salt and K2 + Salt applications. By decreasing according to other applications K3+Salt and K4+ Salt applications was in the same range as the control. In terms of chlorophyll, while K1 + Salt application decreased chlorophyll amount, K2 + Salt and K4 + Salt applications increased compared to control, the amount of chlorophyll in K3 + Salt application was similar to control.

Table 3. MDA and chlorophyll content of leaves taken from plants after applications ( $\mu$  mol / g T.A.)

Application	MDA	Chlorophyll	
Control	3.626C	5.939B	
K1+ Salt	9.162A	3.610C	
K2+ Salt	5.323B	7.430A	
K3+ Salt	3.993C	5.744B	
K4+ Salt	3.402C	7.317A	

### **Discussion and Conclusion**

Pepper plants treated with different K + doses with 100 mM NaCl were fed with nutrient solution. In the present study, the most damaged application was observed at the K1 dose according to the degree of damage caused by salt application in the scale evaluation, which is a morphological observation. K2, K3 and K4 doses respectively. The highest dose of potassium was the least affected by salt damage than the control. Scale assessments based on the degree of damage are a kind of control by comparing the morphological symptoms of salt damage and their grading with other parameters which are shown and examined according to all applications. Aktas (2002) [14], pepper, Yaşar (2003)[1] made use of the scale they created in eggplant studies.

In environmental stress conditions, potassium is vital in protecting chloroplasts against oxidative damage.

ROS formed in stressed plants cause peroxidation of membrane lipids and lead to cell membrane damage [15,16] used as an indicator. In addition, the determination of the amount of malondialdehyde, a product of lipid peroxidation, is used as the simplest indicator of oxidative damage [1,5,16,18,19]. In our study, the amount of MDA in the other parameters in parallel to the amount of MDA salt of plants in which the amount of MDA increased compared to control, but it was observed that the decrease in K +doses. Similar conditions were observed

in chlorophyll deposits. As K + doses increased, there was an increase in chlorophyll levels in the leaves of plants. It was found that there was a correlation between the level of salt effect of the plant and the amount of MDA measured in the leaves. The increase in MDA amount was directly proportional to the decrease in K + dose and the decrease in MDA amount. The amount of chlorophyll was inversely related to MDA. According to the control, salt stress was applied and chlorophyll amount decreased at low K + dose while K2 and K4 doses increased. As it is seen in our study, it is possible to protect the plant from the toxic effect of salt by providing ion balance in cell even under salt stress of appropriate doses of potassium. It also causes an increase in chlorophyll pigments which are other protective factors. Under environmental stress conditions, potassium is vital in protecting chloroplasts against oxidative damage.

Acknowledgement: This work was supported by FAP-2019-8516 (Scientific Research Projects Coordination Unit or Supporting Institution).

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## The Effect of Different Commercial Fertilizer Applications on the Pomological Properties and the Amount of Vitamin C, Water Soluble Dry Matter of Pepper

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### Abstract

In the Study Demre pepper variety was used. The study was carried out in the Physiology Laboratory, Faculty of Agriculture, Van Yuzuncu Yıl University, in a climate room where normal atmosphere was provided. The main purpose of the normal atmosphere in the experiment was to ensure that salt stress effects occur under normal conditions. . The study was carried out under controlled conditions in a light / dark photoperiod of 16/8 hours, climate room of 25 °C and 70% humidity. The study aimed to investigate the on some kinds of-quality criteria of some commercial fertilizer having a big market share in the world and Turkey. To this end, nine different commercial fertilizer applications were made to the pepper plants. Base fertilizers were applied to the groups to be applied to the soil before planting seedlings. Other fertilizer applications were applied in 10-15 days intervals according to the instructions of the company. In addition, the group watered with distilled water only (control) and second groups with only Hoagland nutrient solution of irrigation, fertilizer application constituted control of these commercial applications. The fertilization plan was made in accordance with the size of the company said. In this study; fruit stalk length, fruit diameter, fruit weight, fruit length, and vitamin C contents, the amount of Dry Matter were examined. When the data obtained were evaluated in terms of fruit feature; It was remarkable that Bestline fertilizer had the highest values in terms of all the developmental parameters examined except for the plant nodes distance.

The highest values in terms of fruit diameter and weight were measured from 13-24-12 + 10 (SO<sub>3</sub>+Fe+Zn application and the lowest values were measured from 10-30-10 application.Potassium and phosphorus-weighted fertilizers (TSP, Potasmag, Bestline, 10-30-10) were determined to cause a decrease in vitamin C. According to the control applications, 13-24-12 + 10 (SO<sub>3</sub>) + Fe + Zn, Potasmag, 20-20-20, 10-5-40 fertilizers were also found to have significant decreases in Dry Matter amounts.

Keywords: Commercial fertilizer, fertilizer, pomologic feature, vitamin C.

### Introduction

Fertilizer is one of the most important inputs in agricultural production. If it is not applied sufficiently, it causes significant losses in yield and quality, but if it is applied more, it causes pollution of bottom and surface waters, especially with washing of nitrogen and phosphorus

fertilizer, nitrogen oxide (NO, N<sub>2</sub>O, NO<sub>2</sub>) emission and air pollution [1]. In order to obtain the expected benefit from the applied fertilizer, it is necessary to know the data such as the time, method, plant type, soil character, climate and amount of plant nutrients in the soil [2]. In some studies, it was determined that increases in plant development, fruit number, fruit height, fruit weight and vitamin C increased with chemical fertilizer application [3,4].

The rapid increase in human population necessitates the increase in yield and quality in crop production. Correct fertilization is very important in order to increase yield and quality in crop production. Today, agriculture is still maintained in many places with traditional fertilizer use habits [5]. Unconsciously one-way and large amounts of fertilizer applications without relying on soil analysis bring some yield and quality problems. In addition, this situation has reached the dimensions that threaten human health through nutrition. There are many commercial fertilizers on the market and are being used. Made in this study, which has a large market share in the world and in Turkey, these fertilizers; effects on fruit stalk length, fruit diameter, fruit weight, fruit length, and vitamin C contents, the amount of Dry Matter of pepper plant.

### Material and Method

The experiment was carried out in a split air-conditioned climate room where normal atmosphere was provided. Demre pepper variety was used in the study. A mixture of perlite, peat and barn manure (1: 2: 4) was used as growing medium.

Structure	Ph	Lime	Organic material	EC	Sand	Clay	Silt
	1.2.4.	%	%	mS/cm	%	%	%
Loamy	8.21	11.34	2.23	2284	49.8	24	26.2

**Table 1**. Some physical and chemical analysis results of the growing environment

Applied fertilizers
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	1
10-30-10. Gübresi:	Fosfor ağırlıklı ve iz elementli NPK gübresidir.
10-5-40 Gübresi:	Nitrat azodu(%10), fosfor pentaoksit(%5), potasyum oksit(%40),
	bakır( $\%0.02$ ), demir( $\%0.05$ ), mangan( $\%0.02$ ), çinko( $\%0.02$ )
	içermektedir.
	içermektedir.
Potasmag:	Potasyum oksit(%25) ve magnezyum oksit(%10) içermektedir.
TSP :	Fosfor penta oksit(%42) içermektedir.
Uan:	Amonyak azotu(%8), nitrat azotu(%8), üre azotu(%16),
Uan.	
	bor(%0.01), bakır(%0.02), demir(%0.02), mangan(%0.01),
	molibden(%0.001), çinko(%0.002) içermektedir.
10-25-	Amonyak azotu(%8), üre azotu(%2), fosfor penta oksit(%25),
20+20(SO <sub>3</sub> )+Zn:	potasyum oksit(%20), kükürt trioksit(%20), çinko(%1)
20 / 20(003) / 2m	
	içermektedir.
Bestline:	Amonyum azotu(%5.9), nitrat azotu(%6.6), üre azotu(%2.5),
	fosfor penta oksit(%8), potasyum oksit(%23), magnezyum
	oksit(%2), bakır(%0.02), demir(%0.02), mangan(0.02),
	çinko(%0.02) içermektedir.

13-24- 12+10(SO <sub>3</sub> )+Fe+Zn:	Azot(%13), amonyum nitrat(%12), fosfor pentaoksit(%24), potasyum oksit(%12), kükürt trioksit(%10), çinko(%1), demir(%1) içermektedir.
20-20-20 Gübresi:	Azot(%13), amonyum nitrat(%12), fosfor pentaoksit(%24), potasyum oksit(%12), kükürt trioksit(%10), çinko(%1), demir(%1) içermektedir.

### Table 2. Hogland nutrient solution content

Macro elements	g/lt	Micro elements	g/lt
Ca(NO <sub>3</sub> ) <sub>2</sub> .4H <sub>2</sub> 0	1.180	$C_6H_5FeO_7$ .5 $H_2O$	0.02
KNO <sub>3</sub>	0.252	Mn Cl <sub>2</sub>	0.00072
KH <sub>2</sub> PO <sub>4</sub>	0.136	H <sub>3</sub> BO <sub>3</sub>	0.00116
MgS0 <sub>4</sub>	0.246	$ZnCl_2$	0.000048
-		CuCl <sub>2.</sub> 2H <sub>2</sub> O	0.00004

Pepper seeds were placed in plastic germination containers filled with pumice, after which 100 seeds were planted and watered with tap water. Seeds were planted in the pumice and covered with paper at a temperature of 24-26 °C and left to germinate in the air conditioning room and watered at intervals of 1 day. After germination started, the papers were removed and grown until they had 2 true leaves in a 16/8 hour light-dark photo period. After having 2 real leaves; 1: 2: 4: ratio of perlite, peat and manure growing medium with a size of 9x15 cm 570 ml volume of large six-hole and each cup of 1 seedling in each cup of 12 glasses were confused for each application.

The fertilization plan is based on the dimensions specified by the company. 9 different commercial fertilizer applications were applied to the plants. Base fertilizers applied to the groups were applied to the groups to be applied to the soil before planting seedlings. Other fertilizer applications were applied in 10-15 days intervals according to the instructions of the company. In addition, there are two different groups that constitute the control application of the study. The control group (the group irrigated with pure water only) and the groups made with only 1/2 Hoagland nutrient solution [6] of irrigation were the control application of these commercial fertilizer applications.

In this study, fruit stalk length (cm), fruit diameter (cm), fruit weight (g), fruit length (cm), and vitamin C contents (mg/100g), the amount of Dry Matter (%) were examined.

## **Determination of Fruit Properties**

fruit stem length (cm) and fruit length (cm), with the help of a ruler, fruit diameter (cm) with the help of calipers, and fruit weight (g) was measured with precision scales

## **Dry Matter**

Dry matter content was measured by refractometer.

## Analysis of ascorbic acid (Vitamin C)

Ascorbic acid content was determined according to the method suggested by Cemeroglu (2007) [7]. Samples of 5 mL were mixed with % 2.5 (w/v) metaphosphoric acid (Sigma, M6285,

33.5%), and centrifuged at 6500 rpm for 10 min at 4 °C. 0.5 mL of the solution was completed to 10 mL with % 2.5 (w/v) metaphosphoric acid. Supernatants were passed through 0.45  $\mu$ m membrane filter. Ascorbic acid detection was made with HPLC device using C18 column (Phenomenex Luna C18, 250 x 4.60 mm, 5  $\mu$ ) and the temperature was adjusted to 25 °C. Mobile phase consisted of ultra distilled water with 1 mL/min flow rate at 2.2 pH acidified with H2SO4. DAD detector was used and spectral measurements were made at 254 nm wavelength. Different levels of L-ascorbic acid (SigmaA5960) (50, 100, 500, 1000, and 2000 ppm) were used for ascorbic acid readings. Results are presented as mg/100 g fresh mass

### Statistical analysis

For all the properties studied, a randomized trial design (Random Plots) was used to test the differences between the groups. In case of significant differences between the groups (p < 0.05), Duncan multiple comparison test was applied to the relevant data to test which group had significant difference from the other. For this purpose, SAS (1985) statistical package program [8] was used.

### **Result and Discussion**

Fruit weight, fruit diameter and fruit length were found to be significantly affected from the applications (Table 3).

The average fruit weight of the application is the highest 13-24-12 + 10 (SO<sub>3</sub>) + Fe + Zn and this is the same statistical group; respectively, Hogland, 20-20-20, Bestline, 10-25-20 + 20 (SO3) + Zn applications followed (Table 3). According to control applications, fertilizer applications with significant decreases in average fruit weight were determined as 10-30-10, Potasmag and TSP, respectively.

The fruit diameter ranges from 1.135 cm (Hogland) to 1.384 cm  $(13-24-12 + 10 (SO_3) + Fe + Zn)$ . It was seen that these changes are statistically significant. The length of the fruit stalk, was not statistically significant with the applications made. The length of fruit stalk ranged from 1.993 to 2.167 cm. Fertilizer applications were caused statistically significant changes in fruit length. Fruit length was obtained from 20-20-20 application with 9.691cm measurement. This was followed by data from Bestline (9.182 cm) and Hogland (9.098 cm) applications, respectively. The shortest fruit length was taken from the plants where TSP fertilizer application was made with a measurement of 7.932 cm (Table 3).

**Table 3.** Effects of fertilizer applications on fruit weight (g), fruit diameter (cm), fruit stalk length (cm), fruit length (cm).

Grup		Fruit weight	Fruit diameter	Fruit stalk length	Fruit length
	Ν	$(\overline{X} \pm S_{\overline{X}})$	$(\overline{X}\pm S_{\overline{X}})$	$(\overline{X}\pm S_{\overline{X}})$	$(\overline{X} \pm S_{\overline{X}})$
10-25-20+20(SO <sub>3</sub> )+Zn	35	7.726±2.142 <sup>a-c</sup>	1.302±0.214 <sup>a-c</sup>	2.017±0.404 <sup>a</sup>	8.291± 1.858 <sup>b-d</sup>
13-24-12+10(SO <sub>3</sub> )+Fe+Zn	24	$8.401 \pm 2.422^{a}$	1.384±0.231ª	2.077±0.294ª	$8.842 \pm 1.456^{a-d}$
TSP	39	6.919±2.462 <sup>b-d</sup>	1.222±0.166°	2.154±0.300 <sup>a</sup>	7.932±2.209 <sup>d</sup>
Potasmag	32	6.599±1.947 <sup>c-d</sup>	1.239±0.200°	1.998±0.315ª	$8.713 \pm 1.577^{a-d}$
20-20-20	30	7.855±2.766 <sup>a-c</sup>	1.294±0.206 <sup>a-c</sup>	2.068±0.412ª	9.691±1.647 <sup>a</sup>

Р		0.0048	0.0012	0.371	0.0016
Control	34	7.437±2.752 <sup>a-d</sup>	1.378±0.231ª	$2.153 \pm 0.299^{a}$	8.833±1.958 <sup>a-d</sup>
Hogland	26	$8.141 \pm 2.090^{ab}$	$1.135 \pm 0.146^{ab}$	2.043±0.249ª	9.098±1.716 <sup>a-c</sup>
10-30-10	40	$6.234{\pm}2.318^{d}$	1.196±0.158°	2.167±0.463ª	$8.048 \pm 2.170^{cd}$
10-5-40	33	$7.240 \pm 2.657^{a-d}$	1.293±0.219 <sup>a-c</sup>	2.057±0.311ª	$8.011 \pm 1.433^{d}$
UAN	35	$7.347 \pm 1.730^{a-d}$	1.303±0.245 <sup>a-c</sup>	2.039±0.485ª	$8.807 \pm 2.087^{a-d}$
Bestline	35	7.822±2.090 <sup>a-c</sup>	1.259±0.165 <sup>bc</sup>	1.993±0.288ª	9.182±1.660 <sup>ab</sup>

The difference between the means indicated by different letters in the same column is significant at p <0.05 level.

Koca (2013) [9], potassium sulfate application, plant vegetation growth character (the longest plant, the highest number of leaves, fresh dry weight of the whole plant, average weight, average weight, as well as the total onion yield per unit onion diameter) was found to be the highest. In a similar study El Bassiony (2006) [10], the effect of potassium fertilizer on the growth, yield and quality of onion plants in the study of the best growth (plant height, number of leaves / plants) highest yield and onion quality 20 kg / ha by applying potassium sulfate to the soil obtained. Again in Aisha and Taallab (2008) [11], the highest vegetation growth character (longest plant, maximum number of leaves, maximum fresh dry weight of the whole plant, average weight as well as total onion) using chemical forms of K as potassium sulfate. yield and onion diameter) were determined. Bozköylü (2008) [12] in the study carried out by the effect of chemical fertilizers on plant development and organic fertilizers were found to be more effective than chemical fertilizers. Similarly, in the study of Gulser et al. (2014) [13], the number of fruits, fruit height, fruit weight and vitamin C were found to increase in chemical fertilizer applications.

It was found that the fertilizers used in the study make a significant difference on the amount of dry matter (Table 4). The highest SCF application amount of 5.333% with 10-25-20 + 20 (SO3) + Zn fertilizer, the lowest amount of dry matter with 3.233% 13-24-12 + 10 (SO3) + Fe + Zn fertilizer.

According to fertilizer applications, it was determined that there is a statistically significant difference in the content of vitamin C (Table 4). The highest vitamin C content was obtained from the fruits of plants treated with Hogland nutrient solution, while the lowest amount of vitamin C was measured in the fruits of plants treated with Bestline fertilizer.

		Dry matter (%)	Vitamin C (mg/100g)
Application	N	$(\overline{X}\pm S_{\overline{X}})$	$(\overline{X}\pm S_{\overline{X}})$
10-25-20+20(SO3)+Zn	3	5.333±0.219 <sup>a</sup>	168.260±1.406 <sup>cd</sup>
13-24-12+10(SO3)+Fe+Zn	3	3.233±0.677°	174.981±0.739 <sup>cd</sup>
TSP	3	$5.033{\pm}0.504^{ab}$	134.3675±5.745 <sup>e</sup>
Potasmag	3	3.533±0.393 <sup>bc</sup>	137.783±0.917°
20-20-20	3	3.633±0.233 <sup>bc</sup>	211.244±3.339 <sup>b</sup>

Table 4. Dry matter, vitamin C values of pepper fruits

Bestline	3	4.133±0.797 <sup>bc</sup>	108.518±1.693 <sup>f</sup>
UAN	3	4.900±0.681 <sup>ab</sup>	180.661±4.794°
10-5-40	3	3.900±0.265 <sup>bc</sup>	217.590±4.677 <sup>b</sup>
10-30-10	3	4.933±0.243 <sup>ab</sup>	166.564±4.816°
Hogland	3	4.567±0.291 <sup>a-c</sup>	257.707±4.599ª
Kontrol	3	5.100±0.361 <sup>ab</sup>	$168.000 \pm 5.744^{cd}$
Р		0.018	0.001

In a study conducted by Yoldaş et al. (2009) [14], the amount of dry matter in tomato fruit increased with the use of cattle and chemical fertilizer. The highest values were found to be 5.2% with the use of cattle manure at 6 t / da. They stated that there was no statistically significant difference between the dry matter values obtained with this application and the values obtained with the use of SG and chemical fertilizer (full,  $\frac{1}{2}$  dose) at 9 t / da. They determined that there was a 26.8% increase in dry matter compared to the control with the application of 6 t / da of cattle manure. In the present study, the amounts of TSSC varied from fertilizer to fertilizer and according to control applications, it was determined that there were significant decreases in TSS amounts of 13-24-12 + 10 (SO<sub>3</sub>) + Fe + Zn, Potasmag, 20-20-20, 10-5-40 fertilizers.

Koca (2013) [9] did not observe significant changes in vitamin C content according to the applications of the study. In a study by [15], it was found that nitrogenous fertilization increased the vitamin contents, carotene especially Vitamin B1 contents of the plants and also decreased the Vitamin C content. Bayram Erdoğan and Elmaci (2013) [16] stated that, in addition to soil applications, potassium applications from leaves decreased the symptoms of deficiency and increased functional quality parameters such as taste, texture, color, Vitamin C,  $\beta$  - carotene and folic acid contents. In a study conducted by Yoldas et al. (2009) [14], they found that vitamin C values of tomato fruit ranged between 13-23 mg / 100g. Cattle manure 3, 6 t / da and full application of chemical fertilizers have reported increased amounts of vitamin C. However, these increases were not found to be statistically significant. Similar to these results, Zennie and Ogzewalla (1977)[17], reported the amount of vitamin C in tomato as 23 mg / 100g. Al-Harthi and Al-Yahyai. (2009) [18], in a study comparing the effects of fertilizer forms (K<sub>2</sub>SO<sub>4</sub>, KCl) used as a source of potassium on fruit quality; found that both K sources have positive effects on banana fruit quality. In the same study, while K<sub>2</sub>SO<sub>4</sub> fertilizer has more beneficial effects than KCl fertilizer, this result is in line with the results obtained in the study on the sensitivity of melon plant to different K sources (Jifon and Lester, 2009) [19]. In our study, it was determined that potassium and phosphorus-weighted fertilizers (TSP, Potasmag, Bestline, 10-30-10) caused a decrease in vitamin C.

Acknowledgement: This work was supported by FAP-2019-8516 (Scientific Research Projects Coordination Unit or Supporting Institution).

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## Classification of Triticale Cultivars Using Common Vector Approach and Support Vector Machine

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### Abstract

In this study, six triticale cultivars (Melez, Presto, Sorti, Karma, Tatlıcak, Mikham) with 80 doses of nitrogen (N) fertilizer is classified by using Common Vector Approach (CVA) and Support Vector Machine (SVM). Nine yield characters or features (spike length, spikelet number per spike, grain number per spike, grain weight per spike, grain thickness, grain length, grain width, plant height and grain protein content) belonging to thirty plants of the same triticale cultivar were considered in the classification stage. Very satisfactory results are obtained for the training stage by using both classifiers. In the testing stage, performance of classifiers is low because characters or features belonging to the different cultivars are close to each other.

Keywords: Triticale cultivars, classification, common vector approach, support vector machine.

### Introduction

Triticale is the first successful human-made cereal grain. Triticale takes place in the diets of poor population of some developing countries and is a valuable crop for livestock rations due to its several nourishing characteristics. Besides these properties, triticale is one of the potential plants that can grow easily in marginal agricultural areas (plant nutrient deficiency, hot stress etc.) [1]. Triticale has been used mostly for animal feeding, however, it can be consumed as biscuits, cookies, and unleavened breads such as tortillas and chapatis for human feeding. Because of its grains contain high amino acids concentration and valuable nutritional components; it is aimed to improve its bread-making quality [2]. In addition to, triticale is more resistant than other cereal to unfavorable soil and climatic conditions. It can be achieved to high yield in even marginal areas thanks to this feature. Triticale's acreage is 35.000 hectares; its production is 110.000 tones and yield per hectares 31.5 kg in Turkey [3].

Classification of plant varieties with computer algorithms has been become popular in recent years. Computer-based algorithms have been extensively used in agriculture in order to classify various plants and their characters or samples. The common vectors representing the invariant features of the plants can be extracted by eliminating the differences in each class of plants [4]. Then these common vectors are used for the identification of varieties and characters of plants. Different methods were used in order to derive features or parameters from plant varieties [5-10]. Some classifications were analyzed for characters of rapeseed. Ali et al. [11] analyzed for near infrared spectroscopy and principal component grain of rapeseed. Jankulovska et al. [12]

presented the use of different multivariate approaches to classify rapeseed genotypes based on quantitative traits. Some of these traits using by researchers were plant height, number of primary branches per plant, number of pods per plant, pod length, number of seeds per pod, seed weight per pod, 1000 seed weight, seed weight per plant and oil content.

In this study, six triticale cultivars (Melez, Presto, Sorti, Karma, Tatlıcak, Mikham) with 80 doses of nitrogen fertilizer is classified by using Common Vector Approach (CVA) and Support Vector Machine (SVM). Nine yield characters or features (spike length, spikelet number per spike, grain number per spike, grain weight per spike, grain thickness, grain length, grain width, plant height and grain protein content) belonging to thirty plants of the same triticale cultivar were considered. Yield characters belonging to two of these plants are combined. Thus 15 feature vectors with 18 characters or features are constructed for each cultivar or class. In the classification stage, leave-one-out strategy is used, that is, 14 feature vectors are used for the training stage and remaining one is used for testing stage in each step of strategy. Very satisfactory results are obtained for the training stage by using both classifiers. In the testing stage, performance of classifiers is low because characters or features belonging to the different cultivars are close to each other. Also the number of characters and the number of samples taken from these characters are insufficient in order to classify triticale cultivars efficiently.

### **Material and Methods**

The field experiment was conducted in the year of 2007 at Eskisehir Osmangazi University, Eskisehir (39°48' N; 30°31' E; 789 m elevation) in Central Anatolia Region of Turkey. The experiment included six hexaploid winter triticale (x *Triticosecale* Wittmack) cultivars; Tatlicak 97, Melez 2001, Mikham 2002, Presto 2000, Karma 2000, Sorti. A completely randomized design was assigned. Triticale was planted with 450 seed m<sup>-2</sup> and 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> was applied to all plots. 80 kg ha-1 N as ammonium sulfate applied one-half at planting and one-half at tillering stage. The sowing was done in October. Each plot (7.5 m<sup>2</sup>) had six rows and the space between the rows was 25 cm. The plants were harvested in July.

A 0.54 m<sup>2</sup> (0.18m by 3m) portion at the center of each triticale plot was sampled for 30 spikes of triticale genotypes (spike length, spikelet number per spike, grain number per spike, spike weight) and grain characters (length, thickness, width) at harvest time. The 30 plant height was measured from soil to top of plants. The N content of grain was determined by the Kjeldahl digestion method because protein content was calculated by multiplying 6.25 with N concentration in grain. Nine yield characters (spike length (SL), spikelet number per spike (SPN), grain number per spike (GNS), grain weight per spike (GWS), grain thickness (GT), grain length (GL), grain width (GWI), plant height (PH) and grain protein content (PC) were used. The samples of nine characters of the cultivar Karma are given in Table 1.

Sample	SL	SPN	GNS	GWS	GT	GL	GW	PH	GP
no	(cm)	(#)	(#)	(g)	(cm)	(cm)	(g)	(cm)	(%)
1	12	31	59	2.5	0.15	0.55	0.16	105	10.004
2	12	30	60	1.8	0.17	0.65	0.15	103	10.004
3	12	27	54	2.1	0.16	0.75	0.15	110	12.716
4	11.5	25	49	1.5	0.14	0.65	0.18	103	12.821
5	10.5	25	50	1.2	0.19	0.63	0.12	108	13.400
6	11	27	57	3.1	0.18	0.72	0.21	109	14.188

Table 1. Samples belonging to nine characters of Karma cv.

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171330621.960.180.620.1711111.35418927412.10.170.630.1910712.365191028541.20.160.740.1810511.321201128591.70.150.780.1911211.658
18927412.10.170.630.1910712.365191028541.20.160.740.1810511.321201128591.70.150.780.1911211.658
191028541.20.160.740.1810511.321201128591.70.150.780.1911211.658
20 11 28 59 1.7 0.15 0.78 0.19 112 11.658
21 10 29 40 1.7 0.13 0.63 0.17 109 12.324
22 9 28 42 3.3 0.19 0.62 0.21 93 11.325
23 9.5 27 43 1.06 0.21 0.64 0.22 95 12.630
24 8.5 26 50 1.3 0.25 0.66 0.14 96 11.245
25 10.5 22 41 1.22 0.17 0.55 0.15 97 10.960
26         10         28         42         1         0.19         0.71         0.18         95         11.000
27 11 24 39 2 0.13 0.73 0.19 94 11.359
28 8.5 31 54 1.8 0.16 0.69 0.17 95 10.452
29 9 21 53 1.17 0.17 0.68 0.17 98 12.365
30 9.5 22 55 1.5 0.19 0.64 0.16 96 10.248

CVA is a successful subspace method that has been previously proposed [4, 13-15]. Therefore, CVA was used for the classification of six triticale cultivars (Melez, Presto, Sorti, Karma, Tatlıcak and Mikham). As in all classification methods, CVA has training and testing stages. In the training stage, a common vector which represents common or invariant properties of each class is calculated and an in difference subspace for each class is constructed. Let the vectors  $a_1^c, a_2^c, ..., a_m^c \in \mathbb{R}^n$  be the feature vectors for a variety-class C in the training set where  $m \le n$ . Then each of these feature vectors which are assumed to be linearly independent can be written as

$$\mathbf{a}_{i}^{c} = \mathbf{a}_{i,dif}^{c} + \mathbf{a}_{com}^{c} + \varepsilon_{i}^{c} \qquad \text{for } i=1.2....m$$
(1)

where the vector  $\mathbf{a}_{i,dif}^{c}$  indicates the differences resulting from climatic effects and alienpollination, and the vector  $\mathbf{a}_{com}^{c}$  is the common vector of the variety or character class C, and  $\varepsilon_{i}^{c}$  represents the error vector [13]. The common vector can be obtained from the following procedure. Let us define the covariance matrix of the feature vectors belonging to a variety or character class as

$$\boldsymbol{\Phi} = \sum_{i=1}^{m} (\mathbf{a}_{i}^{c} - \mathbf{a}_{ave}^{c}) (\mathbf{a}_{i}^{c} - \mathbf{a}_{ave}^{c})^{T}$$
<sup>(2)</sup>

where  $\mathbf{a}_{ave}^{c}$  is the average feature vector of  $c^{th}$  class whose covariance matrix is to be calculated and *T* indicates the transpose of a matrix.

The eigenvalues of the covariance matrix  $\Phi$  are non-negative and they can be written in decreasing order:  $\lambda_1 \ge \lambda_2 \ge ... \ge \lambda_n$ . Let  $u_1^c, u_2^c, ..., u_n^c$  be the orthonormal eigenvectors corresponding to these eigenvalues. The first (*m*-1) eigenvectors of the covariance matrix corresponding to the nonzero eigenvalues form an orthonormal basis for the difference subspace B [13]. The orthogonal complement, B<sup>⊥</sup>, is spanned by all the eigenvectors corresponding to the zero eigenvalues. This subspace is called the indifference subspace and has a dimension of (n-m+1). The direct sum of two subspaces B and B<sup>⊥</sup> is the whole space, and the intersection of them is the null space. The common vector can be shown as the linear combination of the eigenvectors corresponding to the zero eigenvalues of  $\Phi$  [13], that is,

$$\mathbf{a}_{com}^{c} = \left\langle \mathbf{a}_{i}^{c}, \mathbf{u}_{m}^{c} \right\rangle \mathbf{u}_{m}^{c} + \dots + \left\langle \mathbf{a}_{i}^{c}, \mathbf{u}_{n}^{c} \right\rangle \mathbf{u}_{n}^{c} \qquad \forall i=1.2.\dots.m$$
(3)

From here, the common vector  $\mathbf{a}_{com}^c$  is the projection of any feature vector onto the indifference subspace  $B^{\perp}$ . The common vector represents the common properties or invariant features of the variety or character class C. The common vector is independent from index *i*. Therefore,

the common vector is unique for each class and all the error vectors  $\varepsilon_i^c$  would be zero.

During the classification stage, the following decision criterion is used:

$$distance = \underset{1 \le C \le S}{\operatorname{argmin}} \left\| \sum_{j=m}^{n} \left\{ \left[ \left( \mathbf{a}_{x} - \mathbf{a}_{i}^{c} \right)^{T} \mathbf{u}_{j}^{c} \right] \mathbf{u}_{j}^{c} \right\} \right\|^{2}$$
(4)

where  $\mathbf{a}_{\mathbf{x}}$  is an unknown or test vector and S indicates the total number of classes. If the distance is minimum for any class C, the feature vector  $\mathbf{a}_{\mathbf{x}}$  is assigned to class C.

Classification algorithm given above can be summarized as follows:

Step 1: Construct feature vectors by using samples taken for each character belonging to any class (cultivar). Be sure that number of samples in each feature vector (or dimension of each feature vector) is greater than number of feature vectors for each class.

Step 2: Find the covariance matrix (Eq. (2)) for each class by using feature vectors belonging to that class.

Step 3: Find the eigenvalues  $\lambda_i$  and corresponding eigenvectors  $u_i$  for each covariance matrix.

Step 4: Find the common vector (Eq. (3)) for each class by using the (n-m+1) eigenvectors corresponding to zero eigenvalues.

Step 5: When an unknown feature vector  $\mathbf{a}_{\mathbf{x}}$  is given, classify this vector by using Eq. (4).

In a briefly explained manner, SVM method [16-20] is originally proposed as a binary classifier and it determines the optimal hyperplane which maximizes the distance between the optimal hyperplane and the nearest sample to this hyperplane. So it is also called the maximum margin classifier [21]. Support vectors correspond to the data samples which are nearest to the optimal hyperplane. If a two-class problem is investigated, the training set is denoted as  $TS = \{(\mathbf{x}_1, L_1), (\mathbf{x}_2, L_2), \dots, (\mathbf{x}_M, L_M)\}$ . In the set.  $\mathbf{x}_i$  ( $i = 1, 2, \dots, M$ ) is the data sample and  $L_i$  ( $L_i \in \{-1,1\}$ ) is the class label (either negative or positive class.). Any vectorial test data (  $\mathbf{x}_{test}$ ) can be classified using the decision function as follow:

$$f(\mathbf{x}_{\text{test}}) = \sum_{i=1}^{M} \left\{ \alpha_i L_i \left( \mathbf{x}_i^{\mathsf{T}} \mathbf{x}_{\text{test}} \right) + b \right\}$$
(9)

where  $\alpha_i (i = 1, 2, ..., M)$  are the nonzero coefficients that are solution of the quadratic programming problem,  $(|b|/||\mathbf{w}||)$  is the perpendicular distance from the optimal hyperplane to the origin and  $\mathbf{w}$  is the normal vector of the hyperplane. The sign of this decision function gives the label of the class to which the test data  $(\mathbf{x}_{test})$  is assigned and this decision function assumes the linearly separable case. SVM classifier explained above is a two-class classifier. For dealing with multi-class problems (with *S* classes), it is possible to construct "S(S-1)/2" classifiers [22]. In this paper, the linear SVM classifier is preferred and it is implemented by using Pattern Recognition Toolbox (PRTools) [23]. All the experiments regarding with the linear SVM classifier are conducted in MATLAB.

#### Results

In the experimental study, six triticale cultivars (Melez, Presto,i Sorti, Karma. Tatlıcak and Mikham) are classified. Each of six cultivars forms one class in the classification methods. Each class has nine characters or features. 30 samples for each character are taken. Two sets of these characters are combined and 15 feature vectors with 18 features are constructed. The "leave-one-out" strategy are used in the testing stage, that is, 14 feature vectors are used in the training stage and remaining one feature vector is tested in each step of the strategy. Therefore, testing stage has 15 steps because each class has 15 feature vectors. The correct recognition rates obtained by using the CVA and SVM methods are given in Table 2 as an average of 15 steps.

		Recognitio	n Rates (%)	
	Traini	ing Set	Testi	ng Set
Cultivars	CVA	SVM	CVA	SVM
Karma	100	92.00	86.67	93.33
Melez	100	80.00	60.00	53.33
Mikham	100	75.55	53.33	26.67
Presto	100	84.4	60.00	40.00
Sorti	100	88.44	66.67	86.67
Tatlıcak	100	87.11	73.33	73.33
Average	100	84.58	66.67	62.22

Table 2. The classification results of six triticale cultivars

When the feature vectors used in the training stage were tested by using CVA all classes were correctly classified. i.e., 100% correct recognition rate is obtained. SVM gave average recognition rate of 85%. But, the scores obtained in the test set are very low for the CVA and SVM methods. The reason of low results is closeness of the samples in the same characters belonging to different cultivars.

#### Conclusions

In this paper, triticale cultivars are classified during the experimental study. It can be concluded that CVA and SVM methods are very successful in the training stage of the classification process. But they are unsuccessful in the test stage because characters or features belonging to

the different cultivars are close to each other. Also, the number of characters and the number of samples taken from these characters are insufficient in order to classify triticale cultivars efficiently.

When more specific characters are extracted for each cultivar, and more samples are taken from characters, good performance can be achieved from the classification process. Also, dissimilarity between the samples in the same character belonging to different doses of fertilizer and different cultivars are very crucial for high performance of classification.

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## Effects of Applied Nitrogen and Potassium on Seed Yield and Oil content of Black Cumin (Nigella sativa L.)

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### Abstract

This study was carried out to determine the efficiency of nitrogen (N) doses (0, 30, 60, and 90 kg N ha<sup>-1</sup>) under supplemental potassium (K) application (50 kg K<sub>2</sub>O ha<sup>-1</sup>) on black cumin in 2011 and 2012. The significant effects (p<0.01) of N and K doses on seed yield and seed oil content were found. The highest seed yield was obtained from 60 kg N ha-1 in the first year and 90 kg N ha<sup>-1</sup> in the second year. The seed yield was increased at a K application dose of 50 kg ha<sup>-1</sup> compared to the control. The highest seed yield (1245.1 kg ha<sup>-1</sup>) in the first year was obtained from 50 kg K ha<sup>-1</sup> with 60 kg N ha<sup>-1</sup>, whereas the highest seed yield (991.4 kg ha<sup>-1</sup>) in the second year was obtained from 50 kg K ha<sup>-1</sup> with 90 kg N ha<sup>-1</sup>. Furthermore, the seed oil content ranged from 36.5 - 40.3 % through N in two-year means. The results showed significant effects of K and N applications on seed oil content. The seed oil content increased with 50 K kg ha<sup>-1</sup> compared to control and decreased with N doses in both years. The highest seed oil content (40.3 %) was obtained from 50 kg K ha<sup>-1</sup> with 30 kg N ha<sup>-1</sup> when both years were combined.

Keywords: Black cumin, nitrogen, potassium, seed oil.

## Introduction

The seeds of *Nigella sativa* L., commonly known as black seed, have been used in traditional medicine especially in many Asian, Middle Eastern and Far Eastern countries to treat headache, coughs, abdominal pain, diarrhea, asthma, rheumatism and other diseases [1-3]. In spite of the fact that *Nigella sativa* L. is consumed very highly throughout the World, low yielded local population of Nigella sativa were cultivated due to lack of registered varieties belong to the species. There are few studies to obtain desired yield and quality using the population. Nitrogen, which is one of the most important plant nutrients, consists a large part of the dry matter in plants [4]. In order to ensure high quality and yielded agricultural products, it is important that the plant nutrients in the soil should be in balance with each other in addition to their quantities in the soil. Unless the plant nutrients in the soil are in sufficient quantities, negative relationships will be appeared during the uptake of plant nutrients from the soil.

Nitrogen takes place in the structure of vitamins, proteins, enzymes and chlorophyll of plants. 15-18% of the protein is composed of N. Generally, K uptake of plants is closely related to N uptake. The plants need K uptake to maintain water availability, especially in dry conditions with increased N fertilization. The element K is also one of the plant nutrients that has important

functions in plants [5]. It has a vital importance in plants for physiological, metabolic and biochemical aspects. Regulates turgor. Prevents water loss and fading in plants. It helps to transport plant nutrients and photosynthesis products. It is known to increase protein content, production and quality. In this study, it is aimed to determine the effects of increased N doses on yield and oil content of black cumin with two different K applications.

### Materials and Methods

This research was carried out during the two years of 2011 and 2012 at the Faculty of Agriculture, Eskisehir Osmangazi University. A local population (Dereyalak) of black cumin was used as seed material. The soil of the experimental area was alkaline (soil/water-1/2.5 pH 8.09), loam textured and calcareous with a low content (0.91%) of organic matter. Phosphorus content (6.4 mg kg<sup>-1</sup>) of the soil was insufficient but the K content (160 mg K kg<sup>-1</sup>) of the soil was sufficient.

Total precipitation in 2011 (223.9 mm) was higher than that in 2012 (168.4 mm); however, the average temperature was lower in the first year (14.8°C) than in the second year (15.3°C). The long-term average temperature (15.5°C) and the total precipitation (160.1 mm) were approximately equal to those of the second year.

The experiment was arranged in a split plot design with three replications (K to main plots and N doses to subplots), and the plot size was  $5.0 \text{ m} \times 1.8 \text{ m}$ , consisting of 6 rows. Seeds were sown at the rate of 15 kg ha<sup>-1</sup> at the end of March in both years using rows with 30 cm spacing on a well-prepared seedbed. Nitrogen fertilization was applied as ammonium nitrate (33% N) at four doses (control, 30, 60, and 90 kg N ha<sup>-1</sup>), and all of the plots received half of N before sowing with the remaining being applied at flowering stage. Phosphorus fertilization was applied at sowing (40 kg ha<sup>-1</sup>) in the form of triple superphosphate (TSP 42% P<sub>2</sub>O<sub>5</sub>).

The plots fertilized with K were not fertilized with TSP because the K fertilization was applied in the form of monopotassium phosphate (0-52-34) at a dose of 50 kg K<sub>2</sub>O ha<sup>-1</sup>. Potassium and N fertilizers were not applied to the control plots. The weeds were controlled manually by hoeing. Harvest and handling were performed manually at the beginning of August. The seeds after harvesting were ground for oil content. The samples were dried at 105°C for 3 hours in a drying oven. After drying, it was treated with ether and degreased and weighed again at 105°C for 2 hours. The differences between the dry samples were determined and % oil ratios were found [6].

#### **Results and Discussion**

A significant two-way interaction between N and K applications was found for seed yield (Table 1) and seed oil content (Table 2). The highest seed yield was obtained from the application of 50 kg K  $ha^{-1}$  with 60 kg N  $ha^{-1}$  as 1245 kg  $ha^{-1}$  in 2011, whereas seed

yield was higher in K application with 90 kg N ha<sup>-1</sup> in 2012. Tulukçu [7] stated that the highest seed yield was obtained with 80 kg N ha<sup>-1</sup> application in which N application increased yield in black seed. In this study, 90 kg N ha<sup>-1</sup> alone increased the yield but the highest yield was obtained in 50 kg K ha<sup>-1</sup> and 60 kg N ha<sup>-1</sup> application compared to the combination of two years.

	Nitrogen	(N) application			
$(kg ha^{-1})$					
0	30	60	90	-	
	2011				
976.5	1052.8	1036.7	1114.7	1045.1	
1000.7	1027.4	1245.1	1182.7	1114.0	
988.6b	1040.1ab	1140.9a	1148.7a		
K: 329.2	N: 127.7*	KxN: 180.5			
	2012				
797.1	851.0	887.5	884.7	855.1	
863.5	859.3	949.4	991.4	915.9	
830.3b	855.2b	918.4a	938.0a		
K: 8.84	N: 5.84*	KxN: 8.26			
	Means of	years			
886.7	951.9	962.1	999.7	950.1	
932.1	943.3	1097.2	1087.0	1014.9	
909.4	947.6	1029.7	1043.3		
	976.5 1000.7 988.6b K: 329.2 797.1 863.5 830.3b K: 8.84 K: 8.84 886.7 932.1	0       30         0       30         2011         976.5       1052.8         1000.7       1027.4         988.6b       1040.1ab         K: 329.2       N: 127.7*         2012       2012         797.1       851.0         863.5       859.3         830.3b       855.2b         K: 8.84       N: 5.84*         Means of 1         932.1       943.3	0306020112011976.51052.81036.71000.71027.41245.1988.6b1040.1ab1140.9aK: 329.2N: 127.7*KxN: 180.5201220122012797.1851.0887.5863.5859.3949.4830.3b855.2b918.4aK: 8.84N: 5.84*KxN: 8.26Means of years886.7951.9962.1932.1943.31097.2	(kg ha <sup>-1</sup> )030609020112011976.51052.81036.71114.71000.71027.41245.11182.7988.6b1040.1ab1140.9a1148.7aK: 329.2N: 127.7*KxN: 180.520122012797.1851.0887.5884.7863.5859.3949.4991.4830.3b855.2b918.4a938.0aK: 8.84N: 5.84*KxN: 8.26Means of years886.7951.9962.1999.7932.1943.31097.21087.0	

Table 1. Effect of N and K fertilization on average of yield (kg ha<sup>-1</sup>) in black seed.

\* and \*\* indicate significance at < 0.05 and < 0.01, respectively.

After N, K is the most needed nutrient by plants. Yield decreases when especially K concentration is low in soil. Saadul [8] and Nataraja et al. [9] reported that 90 kg N ha<sup>-1</sup> and 50 kg K ha<sup>-1</sup> and 50 kg N ha<sup>-1</sup> and 3 kg K ha<sup>-1</sup> applications increased the yield and yield parameters of black seed, respectively. Increased K and N applications have been reported to increase seed yield of cotton [10] and rosemary [11]. In this study, although there is sufficient K content in soil, it has been observed that additional K application increases seed yield.

The highest seed oil content was determined as 45.5% in 2011 and 34.6% in 2012 by 50 kg K  $ha^{-1}$  without N application (Table 2). These results were in accordance with those of Ashraf et al. [12], who found that seed oil content was significantly higher at the control and at the lowest N dose (30 kg N  $ha^{-1}$ ). Moreover, Ozguven and Sekeroglu [13] and Shah [14] did not find any significant differences among N doses on the seed oil content of black cumin. Many studies on other oily seeds have shown that increasing the N dose resulted in reduction in seed oil content [15-18].

Potassium (K)		Nitrogen (N	N) application		
application		(kg	ha <sup>-1</sup> )		Means
(kg ha <sup>-1</sup> )	0	30	60	90	-
		2011		1	1
0	44.323	42.563	41.870	40.833	42.398 b
50	45.463	44.960	43.077	42.823	44.081 a
Mean	44.893 a	43.762 b	42.474 c	41.828 c	
LSD 0.05		K: 0.27**	N: 1.08**	KxN: 1.53	
		2012			
0	34.547	32.590	31.377	34.347	33.215
50	34.247	35.723	32.597	30.247	33.204
Mean	34.397	34.157	31.987	32.297	
LSD 0.05		K: 5.22	N: 3.32	KxN:	4.69
		Means of	years		
0	39.435	37.577	36.624	37.590	37.807
50	39.855	40.342	37.837	36.535	38.643
Mean	39.645	38.960	37.231	37.063	

\*\* indicates significance at < 0.05 and < 0.01, respectively.

## Conclusions

This study revealed that the highest seed yield of black cumin was obtained from the applications of 60 kg N ha–1 with supplemental K in both years, while oil content reduced by increasing N doses. It was concluded that K application should be advised for enhancement of seed yield and oil content of black cumin and the effective N dose was determined as 60 kg N ha–1.

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## **Comparison of Non-Linear Growth Curve Models on Broilers**

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### Abstract

This study was conducted to find the best fitted non-linear model for broilers. Eight non-linear models including Von Bertalanffy, Brody, Gompertz, Logistic, Morgan-Mercer-Flodin, Negatif exponential, Richards and Weibull were fitted on body weight-age data and compared adjusted coefficient of determination ( $R^2$ ), mean square error (MSE), iteration number (IN), Akaike's information criterion (AIC) and Bayesian information criterion (BIC) for determining the most appropriate model describing the growth curve for broilers. Based on all the criteria used for comparing these models in broilers, it can be established that the Richards and Gompertz function gave the best fit for the age-body relationship although Bertalanffy, Logistic, MEF and Weibull functions were equally good in predicting the growth curves of the chickens. Richards and Gompertz models have the highest  $R^2$ , and the lowest MSE, AIC and BIC values. However, it was observed that the Richards model had higher iteration values (64.9) than Gompertz model (9.6). Brody and Negatif exponential functions were not good in fitting chicken growth data in this study with respect to parameter estimates,  $R^2$ , MSE, IN, AIC and BIC values.

Keywords: Growth curve, nonlinear functions, broiler.

## Introduction

Growth is one of the most important biological features of living things. Many definitions of growth have been made to date. Although there is no standard definition, growth can also be expressed as a change in body weight over time. The growth characteristics of animals are shaped by very complex metabolic activities. For this reason, researchers have tried to analyze growth using various mathematical models in order to express the complex biological event more easily. The use of mathematical growth models provides a good way of summarizing the information contained in such data into a few parameters with biological meaning (Fitzhugh 1976; Bilgin et al., 2004).

Growth varies by species and each species has its own growth curve. Lifetime weight-age relationships have become subjects of major interest of animal scientists and producers, due to the economic importance of mature weight, rate of maturing and related characteristics. These models are useful because they summarize time series data into a few parameters to enable an objective comparison of the growth efficiencies. When these functions are expressed graphically, irregular fluctuations in weight caused by random environmental effects are usually eliminated (Al-Samarai, 2015; Adenaike et al., 2017; Kaplan and Gurcan 2018).

The aim of the present study was to estimate growth curve parameters using different non-linear growth models to determine the age-body weight relationships of broilers.

### Materials and methods

The growth functions were carried out from the weekly body weight. Eight functions were used (Von Bertalanffy, Brody, Gompertz, Logistic, Morgan-Mercer-Flodin, Negatif Exponential, Richards and Weibull) to identify the better function that describe the growth curve in the broiler. Table 1 shows the functions considered, Von Bertalanffy, Brody, Gompertz, Logistic, Morgan-Mercer-Flodin, Negatif Exponential, Richards and Weibull.

Von Bertalanffy	$W(t) = A [1 - \beta exp(-kt)]^{3}$
Brody	$W(t) = A [1 - \beta exp(-kt)]$
Gompertz	$W(t) = A \exp \left[-\beta \exp \left(-k t\right)\right]$
Logistic	
	$W(t) = A / [1 + \beta exp(-kt)]$
Morgan-Mercer-Flodin	
	$W(t) = (A\beta + kt^{m}) / (\beta + t^{m})$
Negatif Exponential	$W(t) = A \exp \left[-\beta \exp \left(-\mathbf{k} t\right)\right]$
Richards	$W(t) = A \left[ 1 - \beta \exp(-kt) \right]^m$
Weibull	$W(t) = A - \beta \exp(-kt^{m})$

 Table 1. Growth curve models

Biologically, parameters from the different functions can be in interpreted as follows: *W*t is observed weight at age t. The parameter *A* is weight at maturity, that is, asymptotic limit of the weight when age (t) approaches infinity.  $\beta$  is the initial weight; k is the growth rate. The parameter m is the shape parameter defining the inflection point, which occurs where the estimated growth rate changes from an increasing to a decreasing function. All parameters estimated by the Levenberg-Marquardt iteration method using NLIN procedure in SPSS program (SPSS, 2010). Several criteria are used to determine the goodness of fit: coefficient of determination (R<sup>2</sup>), mean square error (MSE), iteration number (IN), Akaike's information criterion (AIC) and Bayesian information criterion (BIC). To compare the models, 50-day-old body weight data from 20 broilers were used. The body weight of the chickens was measured weekly (1, 7, 14, 21, 28, 35, 42, 50 days of age).Calculations was made on the average means of the weight variable.

## **Results and Discussion**

The model parameters and standard errors estimated from different models are presented Table 2. Eight non-linear models including Von Bertalanffy, Brody, Gompertz, Logistic, Morgan-Mercer-Flodin, Negatif exponential, Richards and Weibull were fitted on body weight-age data and compared adjusted coefficient of determination (R<sup>2</sup>), mean square error (MSE), iteration number (IN), Akaike's information criterion (AIC) and Bayesian information criterion (BIC) for determining the most appropriate model describing the growth curve for broilers. The R<sup>2</sup> values were high for all growth models except for Brody and Negatif exponential indicating a significant relationship between age and weight in broilers.

Modes	А	β	k	m
	***	***	***	***
Von Bertalanffy	6967.9	0.90	0.023	-
Brody	2095851.3	1.00	0.000026	-
Gompertz	4495.7	5.05	0.044	-
Logistic	3071.2	39.18	0.105	-
Morgan-Mercer-Flodin	5948.3	0.000052	61.29	-2.36
Negatif Ex.	847514.9	-	0.000054	
Richards	3735.5	-22.93	0.072	-9.05
Weibull	3879.5	3818.19	0.00014	2.29
SEM	12437.3	73.11	0.808	1.791

Table 2. Means and standard errors of parameters of growth curve models

SEM:Standart error

Concerning with the mature weight (A), except for Brody and Negatif exponential, the value of Morgan-Mercer-Flodin was the highest (5948.3) whereas the lowest (3071.2) was found in Logistic. For the  $\beta$  value, the highest values were found in Weibull (3818.19) and the lowest values were found in Richards (-22.93). On the other hand, the highest value of k was found in Morgan-Mercer-Flodin (61.29) and the lowest in Brody (0.000026).

Table 3. Goodness of fit criteria results for models

Modeller	$\mathbb{R}^2$	MSE	IN	AIC	BIC
	***	***	***	***	***
Bertalanfy	0.998a	2627.3c	13.8e	31.1bc	27.8b
Brody	0.962b	48077.4b	131.8a	41.8a	38.5a
Gompertz	0.999a	1686.6c	9.6e	29.3d	25.9cd
Logistic	0.998a	1869.9c	11.0e	30.1cd	26.8bcd
Morgan-Mercer-Flodin	0.999a	2314.1c	38.0d	31.8b	27.4bc
Negatif Ex.	0.927c	75960.4a	88.6b	42.0a	39.8a
Richards	0.999a	1459.4c	64.9c	29.9cd	25.6d
Weibull	0.999a	1964.1c	29.5d	30.9bc	25.6bcd
SEM	0.001	890.3	3.03	0.377	0.377

Based on all the criteria used for comparing these models in broilers, it can be established that the Richards and Gompertz function gave the best fit for the age-body relationship although Bertalanfy, Logistic, MEF and Weibull functions were equally good in predicting the growth curves of the chickens. Richards and Gompertz models have the highest  $R^2$ , and the lowest

MSE, AIC and BIC values. However, it was observed that the Richards model had higher iteration values (64.9) than Gompertz model (9.6). Brody and Negative exponential functions were not good in fitting chicken growth data in this study with respect to parameter estimates,  $R^2$ , MSE, IN, AIC and BIC values. Similar results obtained by Kuhi et al., (2003) who found that growth functions, the Richards and the Gompertz gave a suitable fit to broilers.

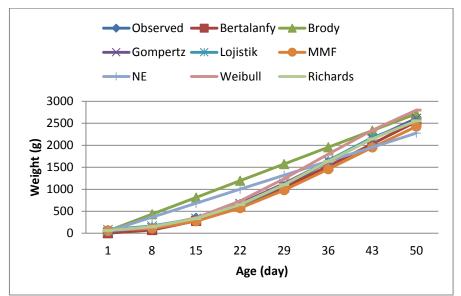


Figure 1. Estimates of growth according to age, in weeks, obtained by the models and observed mean weight.

In conclusion, the comparison of eight growth functions in terms of the goodness of fit criteria revealed that flexible growth functions (especially Richards function) were the most appropriate functions to describe the age-related changes of bodyweight in broilers. Although it was not easy to give a composite description of growth characteristic by interpreting and relating to the biology of the animal, considering all the results of the analysis, Richards and Gompertz models were more appropriate to describe the growth curve in the broiler compared with other functions.

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# The Effect on Plant Growth and Yield in Curly Leaf Lettuce (*Lactuca sativa var. Crispa*) of Different Nutritional Recipes

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#### Abstract

This study was carried out in the Physiology laboratory of Van Yuzuncu Yil University Faculty of Agriculture, Department of Horticulture, in the climate and in the hydroponic culture. This study was carried out in order to determine the yield and plant growth differences which will be produced as a result of applications of different nutrient solutions applied to Caipira variety curly lettuce prepared in a hydroponic culture.

Seven different applications were made in the study. Control group 1 used standard hoagland solution. By modifying the standard Hoagland nutrient solution, in 2<sup>nd</sup>, Magnesium (Mg) and Phosphorus (P) were increased by 10%. In the 3<sup>rd</sup> application, Mg and Potassium (K) were increased by 10%. In the 4<sup>th</sup> application only Nitrogen (N) was increased by 10%. In 5<sup>th</sup> practice, micro elements and N are increased by 10%. On the 6th application, micro elements and K are increased by 10%. In the seventh application, Mg and N contents are increased by 10%. At the end of the study, the weight of chronos, root weight, number of marketable leaves, number of leaves taken were analyzed.

As a result of the analyzes made, the difference between the plant growths in terms of crown weight and crown height was found to be statistically significant ( $\alpha = 0.05$ ). The fourth application has been the best development in terms of both crown weight and crown height and hence yield.

Keywords: Hydroponic culture, curly leaf lettuce, Hoagland solution, nutritional recipes.

### Introduction

Soilless agriculture; stagnant or flowing nutrient solutions, nutrient mist or nutrient solution fed solid environments are defined as the realization of plant production [1]. In this respect, soilless cultivation is divided into water culture (hydroponics) and medium culture (substrate culture).

In both cultivation techniques, nutrient and water requirements of plants are met by using nutrient solutions containing all the necessary elements for their development [2]. Today, one of the main reasons for the rapid spread of landless agriculture in greenhouse cultivation is soil-based problems. Soilless culture, soil fatigue, soil-borne diseases and pests, such as the absence of problems, fertilizer and water control by controlling the development of plants, eliminating the factors that reduce the quality of soil and has advantages such as increasing yield [3]. In addition, as a result of controlled nutrition of plants in soilless agriculture, plant growth and product quality can be kept under control, increasing interest in soilless agriculture.

Today, the importance of cultivation in soilless agriculture is increasing day by day. In recent years; The need for ready-made fertilizer solutions has increased with the widespread use of water-grown vegetables in water culture method. In this study, it was aimed to determine the most appropriate recipe and quality and yield differences that will occur with the macro and micro nutrients added to the nutrient solution in certain amounts on the curly leaf salad group which has the most growing area in water culture.

## Material and Method

This study was carried out in a split air-conditioned climate room in which the normal atmosphere ( $15\pm2^{\circ}C$  13 hours night -  $22\pm2^{\circ}C$  11 hours day and 70% humidity) was provided. The study was conducted in hydroponic culture.

In the study, Caipira curly salad seedlings in the three to four leaf period obtained from the seedling company in Antalya were used as plant material. In addition, 45x32x9 (length x width x depth) cm plastic containers and styrofoam plates of 3 cm thickness were used as growing area material to provide a stagnant aquaculture environment.

Seedlings were harvested 40 days after planting. The effects of 7 different food recipes on yield and quality criteria of Caipira (*Lactuca sativa* var. Crispa) curly lettuce were investigated. Caipira curly leaf lettuce seedlings were placed in circles of 4 cm diameter, also opened in styrofoam plates placed on cuvettes. Seedlings, 15 cm above the row and 18 cm between the rows of these flats are fixed with cotton.

The study was established according to the split plot design. The applications were 3 replications and 15 curly leaf lettuce seedlings were used for each application with 5 plants in each parcel. Table 1 shows the nutrient solution recipe used in the study.

Elements	App.1	App. 2	App. 3	App. 4	App. 5	App. 6	App. 7
Nitrogen (N)	186	186	186	205	205	186	205
Phosphorus (P)	31	34	31	31	31	31	31
Potassium (K)	135	135	149	135	135	149	135
Magnesium (Mg)	49	54	54	49	49	49	54
Calcium (Ca)	217	217	217	217	217	217	217
Sulfur (S)	66	72	72	66	66	66	72
Iron (Fe)	3,3	3,3	3,3	3,3	3,7	3,7	3,3
Manganese (Mn)	0,031	0,031	0,031	0,031	0,034	0,034	0,031
Boron (B)	0,205	0,205	0,205	0,205	0,225	0,225	0,205
Copper (Cu)	0,015	0,015	0,015	0,015	0,016	0,016	0,015
Zinc (Zn)	0,023	0,023	0,023	0,023	0,025	0,025	0,023

**Table 1**. Nutrient solution recipe used (ppm)

Seven different applications were made in the study. In the first application, which is the control group, standard hoagland solution was used. Standard Hoagland nutrient solution [4] was modified, Magnesium (Mg) and Phosphorus (P) increased by 10% in the 2nd application, Mg and Potassium (K) increased by 10% in the third application, only Nitrogen in the fourth application. (N) increased by 10%. In the 5th application, micro elements and N were increased by 10%, in the 6th application micro elements and K were increased by 10% and in the 7th application Mg and N contents were increased by 10%.

The pH was adjusted to 5.8 in nutrient solution solutions renewed every week and the EC values were maintained around 1.35-1.50 mS / cm.

The styrofoam was placed on the cuvettes and the plant roots were made available in the nutrient solution. The aeration is done through thin plastic hoses connected to the aquarium pump. Plants were subjected to illumination for 16 hours per day.

Until the harvest period, the solutions in the cuvettes were renewed with new solutions at intervals of one week. At the end of the study, the weight of crown, the height of crown, root weight, number of marketable leaves, discarded leaves and yield taken were analyzed.

All results were the means of three replicates, and each replicate consisted of fifteen plants. Data were analysed statistically and treatment means were separated by Duncan's Multiple Range Test using SAS [5] software.

## Result

Lettuce seedlings which were transferred to stagnant water culture when three or four real leaves were harvested were harvested 40 days after planting. Maboko and Du Plooy [6] reported that for curly lettuce in water culture, this period varies between 35 and 42 days according to the varieties. On the other hand, Uygunsoy [7] reported that the harvest time was 48 days; this finding is consistent with the literature and the time required from planting to harvest.

To determine the effects of different nutrient solutions on yield and quality criteria of curly lettuce grown in stagnant aquatic culture, the end of this study, crown height (cm), crown weight (g), root collar diameter (mm), root weight (g), discard for each application. number of leaves (number), number of marketable leaves (number), yield (g / m2) were examined. (Table 1; Table 2; Table 3).

Applications	Crown Length (cm)	Crown Weight (g)	Root Throat Dia. (cm)	Root Weight (g)	Num. of Discard Leaves (adet)	Number of Discard Leaves (adet)
Application 1	22.200 B	128.467 C	0.846 AB	11.6 A	3.66 A	22.667 AB
Application 2	22.500 B	130.133 BC	0.810 B	11.2 A	2.66 B	24.667 A
Application 3	25.133 A	152.400 A	0.866 AB	12.0 A	2.66 B	24.333 A

Table 2. Some growth and development parameters of plants

Proceedings of the 2<sup>nd</sup> International Conference on Food, Agriculture and Animal Sciences (ICOFAAS 2019) Antalya, Turkey, 8-11 November 2019

Application 4	25.233 A	162.933 A	0.866 AB	12.6 A	3.00 AB	23.667 AB
Application 5	24.833 A	129.800 BC	0.796 B	10.6 A	2.66 B	21.667 B
Application 6	24.533 A	155.400A	0.986 A	12.0 A	3.00 AB	22.667 BA
Application 7	24.366 A	149.200 AB	0.930 AB	14.1 A	2.33 B	21.333 B

The difference between the averages receiving the same capital letter in the same column is insignificant compared to  $p \le 0.005$ .

In the study, the effects of different nutrient applications on crown height, crown weight, root collar diameter, root weight, discard leaves number, marketable leaf number were statistically significant ( $\alpha = 0.05$ ) (Table 2).

In terms of plant crown height, the highest mean crown height was determined in application 4 (25.233 cm / plant), followed by application 3 (25.133 cm). The minimum crown height average was measured at application 1 (22.200 cm / plant).

Maboko and Du Plooy [8], in their study with curly lettuce varieties in aquatic culture, plant length and plant diameter in curly lettuce is generally indicative of marketable plant size and reported that the plant length varies between 17.21 cm and 14.38 cm. These researchers also investigated the effects of plant frequency on yield and quality of curly lettuce cultivars in water culture in 2009, and grown plants at 10x20, 10x25, 15x20, 20x20 and 20x25 cm intervals and reported that plant length increased with increasing plant density. They have explained that this may be due to competition between plants in terms of photosynthetically active radiation (PAR). Ercan and Bayyurt [9] investigated the effects of two different planting densities on yield and quality of Campania curly lettuce varieties in stagnant water culture, and reported that lettuce grown in 8x8 cm row spacing was longer than lettuce plants grown at 12x12 cm intervals. Uygunsoy [7], Lollo Rossa and Lolo Bionda varieties of curly salad type 12x15 cm intervals (55 plants / m2) in the study of plant lengths measured 35.75 cm and 28.61 cm respectively. Our results are in accordance with the literature.

In terms of plant crown weight, the maximum plant crown weight average was determined in application 4 (162.933 g / plant), followed by application 6 (155.400 g / plant) in the same statistic group, followed by the least plant crown weight average application 1 (128.467). g / plant).

Uygunsoy [7] 's stagnant aquatic culture in the study of four different types of lettuce used in the study for the difference between the varieties of plant rootless weight was statistically significant ( $\alpha = 0.05$ ). Lolo Bionda and red leaf Lollo Rossa varieties with curly salad type have 148.89 g and 132.92 g plant rootless weight respectively. Ercan and Bayyurt [9], stagnant water culture to provide oxygen to the solution ozone generator, air pump and air pump + air stone applications in their study investigated the plant rootless weight, 202 g to 144.9 g have determined. Okudur and Ercan [10], in their study investigating the effect of different fertilizer applications in stagnant aquatic culture, found the plant rootless weight to be 178.69 g in full fertilizer application, and 129.39 g in ready fertilizer application and 98.23 g in 50% fertilizer application. Ercan and Bayyurt [9], Campania curly lettuce cultivated in stagnant aquatic culture grown in lettuce plants 8x8 cm intervals grown in the rootless weight of 282.8 g, 12x12

cm intervals were found to be 178 g. Differences in plant weight may occur due to variety, growing season, fertilizer applications and planting distances.

In terms of plant root collar diameter, the highest root collar diameter average was measured at application 6 (0.986 mm / plant) among applications, while the least root collar diameter average was measured at application 5 (0.796 mm / plant).

Uygunsoy [7], in the study using four types of lettuce, the difference between the varieties in the statistical analysis for stem diameter was found to be statistically significant ( $\alpha = 0.05$ ). Lolo Bionda and red-leaf Lollo Rossa varieties with curly lettuce type are 1.3 cm and 1.3 cm in diameter and are in the same group. Maboko and Du Plooy [6], 16 curly lettuce varieties in their study to determine the performance of water culture, the lowest and highest values for stem diameter measured 2.43 cm and 3.11 cm respectively. It may be that the results obtained are lower than the literature, the harvest time is shorter than the given studies and because of the variety feature

In terms of plant root weight, the highest root weight average was measured at application 7 (14.1 g / plant), while the minimum root weight average was measured at application 5 (10.6 g / plant).

Okudur and Ercan [10], in their study investigating the effect of different fertilizer applications in stagnant water culture, reported that the root weight of Confeti curly lettuce cultivar was 23.55 g in full fertilizer application, 24.12 g in 50% fertilizer application and 29.62 g in ready fertilizer application.

Uygunsoy [7], in his study of stagnant water culture, the difference between the applications in the statistical analysis for root weight was found to be statistically insignificant ( $\alpha = 0.05$ ). Curly lettuce type Lolo Bionda green leaf variety, 77.07 g and Yedikule type Little Gem variety has a root weight of 59.03 g, they formed separate groups. In their study, the reasons of plant roots to be heavier than Okudur and Ercan [10]; varieties, the diversity of the growing season and the later harvest of plants associated with.

In our results, it is possible to say that the root weight average is lower than the other studies, besides the variety feature, it is possible to say that the water temperature used and the growing time are short.

In terms of the number of discard leaves, the highest average number of discard leaves among the applications was measured in application 1 (3.66 units / plant), while the lowest average number of discarded leaves was measured in application 7 (2.33 units / plant).

Gül et al. [11], curly-leaf salads discarded and marketable number of leaves showed significant differences depending on the varieties and growing periods; Topaklı Solak [12] states that cultivation patterns and planting times have a significant effect on the number of discard leaves, and that salads grown in high tunnels have a lower number of discard leaves. In the same study, it was explained that the low number of discarded leaves affected the market value and thus the quality. Considering the literature, it is seen that the number of discarded leaves is quite low. It can be said that this situation stems from aquaculture.

In terms of the number of marketable leaves, the highest average number of marketable leaves was recorded in application 2 (24.667 plants / plants), while the least marketable leaves were recorded in application 7 (21.333 plants / plants).

Ercan and Bayyurt [9], in their study investigating the effects of different applications in the stagnant aquaculture providing oxygen to the solution, reported that the number of leaves per plant in Bohemia and Delight lettuce varieties was between 28.48 and 21.30 and Bohemia varieties formed more leaves. Uygunsoy [7], in his study, the number of leaves of Lolo Bionda variety of curly lettuce type 40.07 units / plant as stated.

Maboko and DuPlooy [6] reported that Tango 20 / plant and NIZ-44-675 cultivar forms 18 leaves / plant leaves in terms of number of leaves per plant in lettuce varieties grown in different plant densities in water culture. Uygunsoy [7] reported that; Bitez lettuce varieties of the type of yedikule reported that the number of leaves varies between 25.10 and 37.30 according to fertilizer applications. The results of the data obtained in the light of these literature are similar to those of Uygunsoy [7]. According to Uygunsoy [7], we can conclude that the reason for the low number of leaves is that the harvest time is shorter than the given studies and the variety is different.

After the weights of the harvested plants were taken, the total marketable yield value in the area covered by the plants was calculated by converting to  $g/m^2$ . In the study, the effects of different nutrient applications on yield were statistically significant ( $\alpha = 0.05$ ) (Table 3).

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Applications	Yield (g/m <sup>2</sup> )	% change	difference (g)
Application 1	3854 C	0	0
Application 2	3904 BC	1.297	50
Application 3	4572 A	18.629	718
Application 4	4888 A	26.829	1034
Application 5	3894 BC	1.037	40
Application 6	4662 A	20.965	808
Application 7	4476 AB	16.139	622

**Table 3.**Yield averages of applications and % changes according to control (Application 1), yield differences

The difference between the averages receiving the same capital letter in the same column is insignificant compared to  $p \le 0.005$ .

Among the applications in terms of yield, the highest yield average was recorded in application 4 (4888 g / m2). This was followed by application 6 (4662 g / m2) in the same statistics group. The minimum yield was determined at application 1 (3854g / m2). It was calculated that there was a difference in yield between 1,037 kg / m2 in m2 compared to application 4 with the highest yield and application 1 with the lowest yield. When this figure is proportional to decare, it is remarkable that there is a big difference of about 1 ton / decare. The greatest difference

between applications with respect to control (Application 1) is determined in application 4, application 6 and application 3, respectively.

Toth et al. [13] examined the yields of five lettuce cultivars grown in stagnant aquatic culture at three different plant densities, and 4.07 kg from 17 plants per m2 of Bonaly cultivar, and 4.17 kg from 13 plants per m2 of Tourbilon. reported that they have the highest yield. Maboko and Du Plooy [6], five different frequency (20, 25, 30, 40 and 50 m2) 4 curly lettuce varieties in their study with the increase in the number of plants per square meter of leaf age and dry weight tend to increase, due to rapid growth rate reported that they came at a significant early harvest. The same researchers reported that the optimal distance for non-head lettuce varieties was 20x20 cm (50 plant m2) and that yield-related parameters gave the best results. Salad-lettuce cultivation yield values vary depending on many factors such as variety, climatic conditions, growing period, growing method, number of plants per unit area, but it is seen that yield is obtained in the range specified in the literature.

As a result of the widespread use of lettuce cultivation in water culture, we found that the best nutrient recipe for the aquaculture was better than the fourth application (Nitrogen (N) increased by 10% only). However, variety characteristics used in aquaculture studies, temperature of the solution used as growing medium, elements present in the water used and EC and pH values of the solution due to the effect of change over time, preparing the formulations by taking them into consideration will result in more successful results and will provide great convenience to manufacturers.

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## Investigation of Macro Element Uptake of Magnesium (Mg<sup>+2</sup>) and Salt Stress Applied Pepper Plants

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### Abstract

This study was carried out in order to determine whether the element of magnesium would be effective in the uptake of macronutrients of the pepper plants under salt stress. In the study, Demre long pepper type was used as material. The study was carried out in 16/8 hour light / dark photoperiod, 25 °C and 70% humid climate room in controlled conditions. After germinating the seeds planted in the pumice, the seedlings formed in 2 real leaves were cultured in hydroponics. Hoagland nutrient solution was used in culture in hydroponic solution. The current Hoagland solution  $Mg^{+2}$  was calculated as Mg = 149,28 ppm and was used as the control. Other doses were Mg 2 = 24,64 ppm, Mg3 = 73,92 ppm, Mg4 = 98,56 ppm, Mg5 = 123,20 ppm. In addition, 100 mM NaCl salt was applied to the plants. Sampling for measurements and analyzes was performed on the 20th day of salt application. In these samples,  $(K^+)$ , zinc  $(Ca^{+2})$ , and magnesium (Mg<sup>+2</sup>) contents from macro nutrients were determined. At the same time, Na<sup>+</sup> and Cl<sup>-</sup> uptake were also investigated because of the salt application. Increasingly added magnesium in saline has generally had a positive effect on the amount of macro elements in the plant, reducing the harmful effect of NaCl and / or lifting it. It was determined that the accumulation of Na and Cl ions dropped in leaves as Mg concentration increased. In the results obtained, Mg4 = 98,56 ppm dose were effective in saving the plant from the detrimental effect of salt.

Keywords: Salt stress, magnesium, macro element, NaCl, pepper.

## Introduction

When the salinity problem is mentioned; It is understood that Na and Cl ions, which make the most harmful effect and are the most common ions, are found in high levels in the soil [1]. Some applications can be made to reduce the harmful effect of salinity, to reverse the loss of productivity caused by salt accumulation, to obtain revitalized soils. These applications consist mainly of large quantities of quality water, energy and careful soil management components. The first method to be used is to wash the sodium chloride from the plant root area using plenty of clean water. The amount and quality of the washing water, the structure of the soil, the type and concentration of the salt, the soil permeability, the efficiency of the drainage system are important for a complete wash. Despite the expenses; The fact that the salinity problem is mostly seen in arid and semi-arid areas makes it clear that a solution in the form of washing with water is not practical. In addition to washing the salt from the soil profile with water; some methods such as increasing the amount of soil humus by using organic fertilizers, avoiding

excessive inorganic fertilization, the use of fertilizers containing soil salt-enhancing elements such as high filler and chlorine, the use of soilless cultivation in greenhouses or changing the top layer of the soil at regular intervals, can be applied to avoid damages. But, these processes are sometimes time-consuming and often expensive. In addition, saline soils can be re-formed in areas where improved irrigation methods are not used [2,3]. However, the most important and most definite way to eliminate these negativities is to develop plant species and varieties that are tolerant to salinity and to make applications to eliminate the negative effects of salinity.

Based on this information, the aim of this study was to determine the relationship between Mg uptake, macro element uptake and Na, Cl accumulation in stressed plants by applying different doses of Mg in nutrient medium together with salt stress to Demre peppers.

## **Materyal and Method**

This research was carried out in the climatic room of Van Yüzüncü Yıl University, Faculty of Agriculture, Department of Horticulture, Physiology Laboratory. Demre sharp pepper variety was used in the study.

The experiment was carried out in a split air-conditioned climate room with normal atmosphere and water culture. The main objective of the experiment is to ensure that the effects of salt stress occur as normal under normal conditions. In this way, the stress level to which the plants are exposed in the outdoor environment will be measured with the least margin of error and the results obtained as a result of the application of the results obtained will show greater consistency with the study results.

Elements	App. 1	App.2	App.3	App.4	App.5	App.6
	Control	Mg1+NaCl	Mg2+NaCl	Mg3+NaCl	Mg4+NaCl	Mg5+NaCl
Nitrogen (N)	186	186	186	186	186	186
Phosphorus (P)	31	31	31	31	31	31
Potassium (K)	135	135	135	135	135	135
Magnesium(Mg)	49,28	24,64	49,28	73,92	98,56	123,20
Calcium (Ca)	217	217	217	217	217	217
Sulfur (S)	66	66	66	66	66	66
Iron (Fe)	3.3	3.3	3.3	3.3	3.3	3.3
Manganese (Mn)	0.031	0.031	0.031	0.031	0.031	0.031
Boron (B)	0.205	0.205	0.205	0.205	0.205	0.205
Copper(Cu)	0.015	0.015	0.015	0.015	0.015	0.015
Zinc (Zn)	0.023	0.023	0.023	0.023	0.023	0.023

**Table 1.** Contents of the nutrient solution used (ppm).

The nutrient solution used was prepared according to Hoagland and Arnon, [4].

In the study, first of all, pepper seeds were sown in 40x25x5 cm plastic germination cups filled with pumice sieved and then watered with fountain water. This irrigation method has been used since it is not possible to use normal irrigation water or water in the environment where pepper is grown for internal irrigation. The bottom surface of the germination vessels has 9 holes with a diameter of 0.5 cm and the irrigation water is drained by the plants. After the pumice was thoroughly wetted and the irrigation water was drained, the germination pots were placed in the climate room with  $25\pm2$  °C temperature 70% humidity, covered with A4 paper and the containers were regularly checked and the pumice was continued to be irrigated with tap water. For the better development of the seedlings, the cotyledon leaves coming horizontally and the first true leaves (3-4) began to be seen, irrigation was started with Hoagland nutrient solution [4].

### Mineral element analysis

Three leaves from tip to downward were taken and they were kept in deep freezer at  $-84^{\circ}$ C. About 200 g samples were taken from the deep freezer and samples were supplemented with 10 ml 0.1 N HNO3 (Nitric acid). They were then kept in plastic boxes at dark and room temperature for a week. The samples were shaken in the shaker for 24 hours and Na<sup>+</sup>, K<sup>+</sup>, Ca<sup>+2</sup>, Mg<sup>+2</sup> contents were read in Atomic Absorption Device according to Kacar (1994) [5]. Cl<sup>-</sup> ion was measured by an automated chloridometer (Buchler – Cotlove chloridometer) which was analyzed by colorimetric amperometric titration with silver ions. At the end of these measurements, the amount of ions in the fresh leaf sample was determined as  $\mu$ g/mg fresh weight [6].

All results were the means of three replicates, and each replicate consisted of fifteen plants. Data were analysed statistically and treatment means were separated by Duncan's Multiple Range Test using SAS (1985) [7] software.

#### Result

At the end of the application of 100 mM NaCl salt stress for 20 days, the data obtained in terms of the amount of Na, K, Cl, Ca and Mg ion in the leaf is given in Table 2.

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Application	Na <sup>+</sup>	Cl	$\mathbf{K}^+$	Ca <sup>+2</sup>	Mg+
Control	1.2306 D	0.2840 C	148.92 A	6.2996 A	15.698 A
Mg1+NaCl	7.3513 A	2.3633 AB	74.58 B	3.7722 B	9.677 B
Mg2+ NaCl	6.4585 AB	2.6877 A	106.65 B	4.8389 AB	11.808 AB
Mg3+ NaCl	4.6489 BC	1.8870 AB	103.60 B	5.1168 AB	12.435 AB
Mg4+ NaCl	3.5498 C	1.5130 B	108.99 AB	5.8755 A	14.033 AB
Mg5+NaCl	4.1539 C	1.8870 AB	71.02 B	4.7948 AB	12.510 AB

**Table 2.** Ion deposits in leaf parts after application ( $\mu$  g / mg F.W.)

The difference between the means taking the same capital letter in the same column is insignificant according to P $\leq$ 0.05. (F.W. Fresh Weight)

Na accumulation in the leaves of salt treated plants as Mg doses increased decreased due to the increase in magnessium doses (until Mg5+NaCl). The highest Na accumulation was observed in Mg1 + NaCl application while the lowest Na accumulation was observed in Mg4 + NaCl application.

In terms of Cl accumulation in the leaves of the plants treated with salt, the difference between the other treatments except the control was found to be statistically significant. There was an increase in Cl ion accumulation in other applications compared to the control group. Cl

accumulation in the leaf organ increased compared to control. The highest increase was seen in Mg2 + NaCl application, while the lowest Cl accumulation was observed in the application of Mg4 + NaCl. As the Mg dose increased, the accumulation of Cl in the leaf decreased until Mg5+NaCl.

Similar to the control, the highest K accumulation in the leaves was observed at Mg4 +NaCl dose. The lowest K accumulation was observed in the application of Mg5 + NaCl.

It was seen that there were differences between the applications in terms of Ca accumulation in the leaves in the salt treated plants. Mg4 + NaCl application was in the same statistical value range as the control, while the lowest Ca accumulation was observed in the application of Mg1 + NaCl.

In the study of salt application, it was found that there are differences in Mg + accumulation in leaves. Mg accumulation was a decrease in lowest doses (Mg1 + NaCl) of magnessium before leaf control, there was an increase in high doses. Similar to the control, the highest Mg accumulation in the leaves was observed at Mg4 +NaCl dose.

#### **Discussion and Conclusion**

One of the most important reasons for the decrease in the growth of salt stressed pepper plants is the amount of Na they accumulate in their bodies in excess and toxic levels. Na + entering the plant cell disrupts membrane potential and facilitates passive entry of extracellular Cl into the cell via anion channels [8,9]. High NaCl uptake leads to an increase in the accumulation of Na <sup>+</sup> and Cl<sup>-</sup> ions in the cell and a decrease in Ca <sup>+2</sup>, K <sup>+</sup> and Mg <sup>+2</sup> concentrations [10]. Yasar et al. (2006a) [11], Aktaş [2] (2002), Turhan [12] (2002), in their studies, under the stress of salt in the leaves of plants increased the accumulation of Na and Cl ions. In our study, it was observed that increasing doses of Mg to the plants under salt stress reduced the accumulation of Na and Cl ions in the leaves of the plants. In addition, Levitt [13] (1980), in the case of more than NaCl in the environment where plants receive more than the Na ion and the competition reported that there is a decrease in K ion. Likewise, Yaşar et al. [14] (2006b) in their study of watermelon stated that under the stress of salt K and Ca ions decreased in the leaves of plants. In our study, with increasing doses of Mg applications to plants under salt stress; the uptake of these ions is almost identical to the control plants.

In our study, the lowest Na ion accumulation in the leaves was obtained from Mg4+NaCl and Mg5+NaCl salt applications. As the Mg dose increases, it is seen that the Na ion accumulation of the plants decreases. It is possible to say the same in terms of Cl ion accumulation. It has been determined that increasing doses of Mg in salt stressed plants reduces the accumulation of these two ions. With increasing doses of Mg, the accumulation of Mg ions in the leaves of the plants increased until the last dose of Mg, Mg 5+, but at this dose, the amount of Mg decreased in the leaves because the plant did not take the excess magnesium.

In some studies, Ca, Mg and K compounds added to the nutrient solution together with NaCl applications had generally positive effects on the amount of macro elements in the plant, reducing and / or eliminating the harmful effect of NaCl [15,16].

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## Effect of Inoculation with Plant-Growth Promoting Rhizobacteria on Development Root Systems of Lemon Basil (*Ocimum x citriodorum* Vis.)

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## Abstract

Basil is a popular culinary herb and ornamental plant, its leaves, flowers, and seeds are used in the medicinal industry, cuisine and for aromatic and culinary purposes, and it is grown as a garden ornamental. On the other hand, *Ocimum* x *citriodorum* Vis., popularly known as lemon basil, is an aromatic medicinal plant and strongly aromatic and possesses a lemon flavour and fragrance, belonging to the Lamiaceae family. The objective of this study was to evaluate possible effects of mineral fertilizer (NPK), two commercial bio-fertilizers and ten indole acetic acid (IAA) producing, N<sub>2</sub>-fixing and P-solubilizing bacteria based formulations in single, dual and triple strains combinations (Pseudomonas fluorescens RC512; Bacillus licheniformis RC601; Bacillus subtilis RC210; Bacillus megaterium RC16; Pseudomonas fluorescens RC512+Bacillus licheniformis RC601; Pseudomonas fluorescens RC512+ Bacillus megaterium RC16; Pseudomonas fluorescens RC512+ Bacillus licheniformis RC601; Pseudomonas fluorescens RC512+Bacillus licheniformis RC601 + Bacillus subtilis RC210; Pseudomonas fluorescens RC512+Bacillus licheniformis RC601+ Bacillus megaterium RC16 and *Pseudomonas fluorescens* RC512+ *Bacillus subtilis* RC210 + *Bacillus megaterium* RC16) on shoot and root growth of lemon basil (Ocimum x citriodorum Vis). The results suggest that multi-traits bacteria (especially IAA-production) play a major role in the development of the plant root during the early stages of growth. Plant growth-promoting rhizobacteria (PGPR) caused high root weight, total root numbers and encouraged adventitious root formation but it's were strongly dependent on bacterial formulations. PGPR may change root growth, increase root weight, proliferation of lateral and adventitious roots number and may have an influence on nutrient uptake potentials. The positive effects of PGPR on lemon basil growth were correlated with remarkable changes root growth and morphology, namely increasing the lateral root and root hair number. The elongation or proliferation of root systems has been suggested to be one of the mechanisms by which PGPR stimulate medicinal and aromatic lemon basil growth. The experiment revealed that the PGPR inoculation was effective treatment to improve the root growth and change in root morphology; bacteria tested during our study have great potential being formulated and used as biofertilizer.

Keywords: Lemon basil, plant growth-promoting rhizobacteria, root growth, yield

## Introduction

The basil plants (*Ocimum* sp.; Lamiaceae) are among the most important crops which essential oils stand out for the quality, quantity and chemical diversity. Basil is generally used as a flavoring agent in food industry. Basil (*Ocimum basilicum* L.) is one of the most important herb plants; its leaves, flowers, and seeds are used in the medicinal industry, perfumery and for

culinary purposes, food, pharmaceuticals, fragrances, beverages, antimicrobial, and insecticides, and it is grown as a garden ornamental [1-4]. Basil is an annual plant and a popular culinary herb and ornamental plant, which is also used in herbal medicine, aromatic, cuisine, ornamental, sacred, vegetable and different sectors of economy including food industry. Fresh sweet basil leaves is used as an ingredient in various dishes and food preparations, especially in the Mediterranean cuisine [5]. Besides the essential oil compounds, different varieties of basil plants are characterized with a significant content of polyphenols, particularly total phenolics, phenolic acids, and flavonoids with significant antioxidant activity. The essential oil of *Ocimum* spp. were rich in camphor, citral, eugenol, geraniol, linalool, linalyl acetate and methyl chavicol [6].

*Ocimum x citriodorum* Vis., popularly known as lemon basil, is an aromatic medicinal plant, belonging to the Lamiaceae family. *Ocimum citriodorum* Vis or lemon basil is a cross-pollination cultivar from two chemotypes of sweet basil; *Ocimum basilicum* and *Ocimum americanum* [7, 8]. Lemon basil is a culinary annual plant that is strongly aromatic and possesses a lemon flavour and fragrance due to the presence of citral in the form of its geometrical isomers neral and geranial [9]. Due to its lemony scent and flavour, it could find use as a source of citrus flavour and an alternative to lemon grass and other lemon scented botanicals for its culinary and flavour attributes. It presents a strong lemon flavour because citral constitutes the majority of its essential oil [10]. Also, lemon basil has an ability to produce high content of phenolic compounds, particularly rosmarinic and caffeic acids with significant antioxidant activity [11]. The main components of lemon basil essential oil were citral, geranial, neral, and linalool [12]. Citral (neral+geranial) was the major component of the essential oil lemon basil and was abundant in the flower (64.5%) but was also present in the branch (65%) and the leaf (42%) in significant quantities [9].

Lemon basil has also been used as a raw material by the chemical, pharmaceutical, and food industries. In addition, essential oils have been widely used in condiments for meats, salads, nonalcoholic beverages, ice cream, the perfume industry, and oral hygiene products [12]. In addition, the soaked basil seed can be combined with juice or milk to drink, and fiber production from basil seed [13]. Essential oils have been widely used in condiments for meats, salads, nonalcoholic beverages, ice cream, the perfume industry, and oral hygiene products [14]. The essential oil is of high economic value as it has been reported to be rich in antimicrobial and anti-inflammatory properties, thus, making it an important component of herbal medicines [15].

The aim of this work was to evaluate effects of mineral NPK fertilizer, two commercial biofertilizers, and single and co-inoculation with auxin (IAA)–producing, N<sub>2</sub>-fixing, and Psolubilizing bacteria on growth promotion and yield in lemon basil seedlings under natural soil conditions by conducting pot experiments at the Çanakkale Onsekiz Mart University, Faculty of Agriculture, Department of Field Crops (Table 1).

## Material and methods

The objective of this study was to evaluate possible effects of mineral NPK fertilizer (300 mg N + 250 mg P + 200 mg K per seedlings), two commercial bio-fertilizers and ten indole acetic acid (IAA)-producing, N<sub>2</sub>-fixing and P-solubilizing bacteria-based formulations in single, dual, triple and and quadruple strains combinations (BF1: *Pseudomonas fluorescens* RC512, BF2: *Bacillus licheniformis* RC601, BF3: *Bacillus subtilis* RC210, BF4: *Bacillus megaterium* RC16, BF5: *Pseudomonas fluorescens* RC512+*Bacillus licheniformis* RC601, BF6: *Pseudomonas fluorescens* RC512+*Bacillus licheniformis* RC601, BF6: *Pseudomonas fluorescens* RC512+

Bacillus subtilis RC210, BF8: Pseudomonas fluorescens RC512+ Bacillus subtilis RC210 +Bacillus licheniformis RC601, BF9: Pseudomonas fluorescens RC512+Bacillus licheniformis RC601+ Bacillus megaterium RC16, BF10: Pseudomonas fluorescens RC512 + Bacillus subtilis RC210 + Bacillus megaterium RC16, BF11: Pseudomonas fluorescens RC512+ Bacillus subtilis RC210 + Bacillus licheniformis RC601+ Bacillus megaterium RC16) on shoot and root growth, yield, chlorophyll (SPAD) and anthocyanin (ACI) content of lemon basil (Ocimum x citriodorum Vis). The pots experiments were arranged as a completely randomized design fifteen treatments and four replicates (each having three seedling).

For this experiment, pure cultures were grown in 50% strength tryptic soy broth on a rotary shaker (120 rpm; 25 °C) for 3 days. Bacteria were then harvested by centrifugation (ca. 3000 x g for 10 in), washed and re-suspended in 10 mM sterile phosphate buffer, pH 7 to a density of 10<sup>9</sup> cfu ml<sup>-1</sup> for the bacterial strains. For dual, triple and quadruple inoculation, equal volume  $(10^9 \text{ cfu ml}^{-1} \text{ of each inoculant})$  of each culture were mixed and then used for lemon basil. Seedlings were surface-sterilized prior to inoculation by soaking in 25% commercial-grade bleach for 5 min, followed by thorough washing under running tap water and air-drying aseptically overnight at room temperature. For the two microorganisms based commercial biofertilizers and four triple and quadruple bacterial formulations, frozen bacterial culture seeded in petri dish Nutrient Agar (NA) containing medium, incubated for 24 hours at 27 °C. Pure colonies were taken from fresh culture and transferred to Nutrient Broth (NB) culture media. Horizontal shaker incubator developed a 24-hour culture, inoculated in NB containing the liquid culture media, previously prepared by fermenters and sterilized by autoclaving at 121 °C for 20 min [16]. Bacteria were developed 24 h optimum pH, oxygen, and temperature values. Then, all of the organic substances and steam-sterilized liquid carrier mixing ratio of 1:10 inoculations. Bacteria inoculated organic liquid carrier the optimum growth conditions were incubated in the bioreactor. Counts of viable bacteria per millilitre (cfu) made in bacterial concentration was  $1 \times 10^8$  cells / ml at the end of 48 hours, during which time exceeds, packaging made completely sterile conditions, the product has been kept in a cold room at 5 °C [16]. Uniform height young seedlings were inoculated with each of the bacteria-based bio-fertilizers formulations. The bacterial inoculation involved dipping the root system of the seedling into a suspension of each bio-formulations for 60 min, prior to planting [17,18]. The remaining part of the suspension was injected into the root area. Control plants received 5 ml of diluted SPB with no bacteria. The inoculated seedlings were planted without waiting and the water was applied after planting. Fresh and dry shoot and root weight, shoot length, shoot and trunk diameter and plant height were collected for all lemon basil.

Plants were grown under open air conditions of natural light during the experiment. The greenness or total chlorophyll contents of the top fourth and fifth leaves were measured using a chlorophyll meter SPAD-502 (Minolta, Japan), which is used to measure leaf greenness of the plants. The first experiment set was harvested 100 days after planting. An analysis of variance (ANOVA) and Duncan's multiple range test (at p<0.05) were performed to analyse statistical differences and to discriminate between means (Stat Soft Inc, 1998).

## **Result and Discussion**

Inoculation of single and mixed bioformulations of  $N_2$ -fixing and P-solubilizing bacteria promoted growth and yield of lemon basil under natural conditions by conducting pot

experiment. The best effects were obtained in plant growth in soil with commercial bio fertilizer of BMusaVita and BMusa Green, and mixed combination of the three (BF8: Pseudomonas fluorescens RC512+ Bacillus subtilis RC210+ Bacillus licheniformis RC601). Fresh and dry herbage yield and chlorophyll contents and branch number greatest with BMusa Green, whereas maximal root fresh and dry weight and lateral root number were with BMusa Vita (Table 1). Except for RC210, RC16, Rc512+RC601, and RC512+RC210+RC16, other bacterial inoculation and NPK fertilizer application significantly increased dry herbage yield in lemon basil. Also, except for RC16 and RC512+RC210+RC16 formulations, other treatments significantly increased root fresh weight compared with control. Of the bacterial inoculations and mineral fertilizers, the maximum dry herbage yield in lemon basil was measured in the B Musa Green commercial bio-fertilizer and triple inoculation with RC512+RC210+RC601 followed by B Musa Vita, NPK fertilizer, and RC512+RC16 treatments. The maximum dry root weight in lemon basil seedlings was measured after BMusa Vita and BMusa Green commercial bio-fertilizers, followed inoculation of triple RC512+RC210+RC601 and single RC512, and mineral fertilizer application of NPK. The highest lateral root number was observed after single inoculation with RC512 and commercial bio-fertilizer BMusa Vita, followed by triple RC512+RC210+RC601 and single RC601 bacterial inoculation (Table 1).

Treatments*	Fresh herbage yield	Dry herbage yield	Root fresh weight	Root dry weight	Lateral
	(g/plant)**	(g/plant)	(g plant)	(g plant)	number
Control	18.1±2.0 d	4.20± 0.62 f	4.18±0.46 b	0.39±0.04 f	9.3±1.3 g
NPK	25.6±2.8 ab	6.28± 0.69 ab	5.4±0.59 a	0.51±0.06 a-d	13.8±0.5 c-e
BMusa Vita	26.4±1.6 a	6.43±0.71 ab	5.9±0.80 a	0.58±0.06 a	16.8±1.0 a
BMusa Green	26.4±2.9 a	6.64± 0.73 a	5.63±0.62 a	0.55±0.06 ab	16±1.4 a-c
RC512	22.9±2.5 а-с	5.39± 0.59 c-e	5.46±0.6 a	0.51±0.06 a-d	17±1.4 a
RC601	21.3±2.3 cd	5.13± 0.56 c-e	5.41±0.59 a	0.47±0.05 b-e	15.8±1.7 a-c
RC210	19.9±2.2 cd	$4.63 \pm 0.71$ ef	5.3±0.58 a	0.45±0.05 c-f	14.3±1.7 b-d
RC16	19.7±2.2 cd	4.74±0.41 d-f	5.06±0.56 ab	0.38±0.04 f	10.7±1.3 fg
RC512+RC601	19.9±2.2 cd	4.83±0.53 d-f	5.35±0.59 a	0.43±0.05 ef	12.0±1.4 d-f
RC512+RC16	21.8±2.4 b-d	5.86±0.65 a-c	5.27±0.58 a	0.42±0.05 ef	12.0±0.8 d-f
RC512+RC210	22.0±2.4 b-d	5.25±0.58 с-е	5.43±0.6 a	0.5±0.05 а-е	12.5±1.3 d-f
RC512+RC210+RC601	23.0±2.5 а-с	6.56±0.72 a	5.6±0.62 a	0.53±0.06 a-c	16.50±2.1 ab
RC512+RC210+RC16	22.5±2.5 bc	5.08±0.29 c-f	5.04±0.55 ab	0.41±0.05 e-f	12.5±0.6 d-f
RC512+R601+RC16	21.1±2.3 cd	5.64±0.35 b-d	5.41±0.6 a	0.47±0.05 b-f	13.3±3.1 d-f
RC512+RC210+	20.9±2.3 cd	5.14±0.57 с-е	5.27±0.58 a	0.45±0.05 c-f	11512100
RC601+RC16	20.9±2.5 CU	J.14±0.J/ C-C	5.27±0.38 å	0.45±0.05 C-I	11.5±2.1 e-g

Table 1. Effect of fertilizer application and bacterial inoculations of lemon basil with plant growth promoting rhizobacteria on herbage yield, root weight and lateral root number

\*Control: Without bacteria and fertilizer, NPK fertilizer (300 mg N + 250 mg P + 200 mg K per seedlings); BF1: *Pseudomonas fluorescens* RC512, BF2: *Bacillus licheniformis* RC601, BF3: *Bacillus subtilis* RC210, BF4: *Bacillus megaterium* RC16, BF5: *Pseudomonas fluorescens* RC512+*Bacillus licheniformis* RC601, BF6: *Pseudomonas fluorescens* RC512+ *Bacillus megaterium* RC16, BF7: *Pseudomonas fluorescens* RC512+ *Bacillus subtilis* RC210, BF8: *Pseudomonas fluorescens* RC512+ *Bacillus subtilis* RC210, BF8: *Pseudomonas fluorescens* RC512+ *Bacillus subtilis* RC210, BF8: *Pseudomonas fluorescens* RC512+ *Bacillus licheniformis* RC601, BF9: *Pseudomonas fluorescens* RC512 + *Bacillus licheniformis* RC601+ *Bacillus megaterium* RC16, BF10: *Pseudomonas fluorescens* RC512 + *Bacillus subtilis* RC210 + *Bacillus megaterium* RC16, BF11: *Pseudomonas fluorescens* RC512+ *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC210 + *Bacillus subtilis* RC2010 + *Bacillus subtilis* RC2010 + *Bacillus subtilis* RC2010 + *Bacillus subtilis* RC2010 + *Bacillus subtilis* RC2010 + *Bacillus subtilis* RC2010 + *Bacillus licheniformis* RC601+ *Bacillus megaterium* RC16); \*\*Values followed by different letters in a column were significantly different (*p* < 0.05) using Duncan's multiple range test

Statically significant difference in chlorophyll content were observed for BMusa Green, BMusa Vita and RC512+RC210+RC601, as compared to control, NPK fertilizer and other bacterial formulations (Table 2). The maximum plant height was measured after triple RC512+RC210+RC601 inoculation, followed by BMusa Green and RC512+RC16 inoculations. The highest lateral root number was observed after single inoculation with RC512 and commercial biofertilizer BMusa Vita, followed by triple RC512+RC210+RC601 and single RC601 bacterial inoculation. The maximum Branch number was measured after BMusa Green biofertilizers, followed by RC512+RC16 BMusa Vita and NPK treatments.

Treatments*	Chlorophyll contents (SPAD) **	Anthocyanin contents (ACI)	Plant Height (cm)	Trunk diameter (mm)	Branch number
Control	32.9±1.1 e	3.67±0.2 gh	37±1.41 cd	4.71±0.09 de	10.0±1.4 c
NPK	36.6±1.7 cd	3.80±0.17 gh	41.5±2.08 a-d	5.46±0.23 ab	13.3±0,9 ab
B Musa Vita	41.0±1.1 a	4.81±0.12 bc	44±3.34 a-d	5.53±0.31 a	13.5±2.1 ab
B Musa Green	41.2±1.9 a	4.87±0.37 с-е	45.18±4.99 ab	5.33±0.39 a-c	14.7±1.0 a
RC512	34.8±1.4 c-e	4.24±0.14 ef	36.25±2.87 d	4.95±0.39 b-e	11.8±0.96 bc
RC601	36.3±1.7 cd	4.26±0.11 ef	39±7.12 a-d	4.51±0.31e	11.7±1.0 bc
RC210	34.4±1.0 de	4.71±0.30 cd	38.38±6.34 b-d	4.74±0.51 de	13.3±2.2 ab
RC16	36.6±1.1 cd	4.35±0.25 d-f	41.5±3.51 a-d	4.60±0.59 e	11.8±2.6 bc
RC512+RC601	33.1±1.4 e	4.54±0.17 c-e	39.88±3.33 a-d	4.63±0.37 e	13.0±1.4 ab
RC512+RC16	35.0±0.7 с-е	3.55±0.21 h	44.25±6.95 a-c	5.26±0.43 a-d	14.3±1.7 ab
RC512+RC210	37.1±1.2 bc	3.51±0.23 h	39.88±5.63 a-d	5.26±0.23 a-d	12.8±1.7 a-c
RC512+RC210+RC601	40.0±1.2 ab	3.64±0.29 gh	46.5±3.39 a	5.53±0.12 a	14.0±1.8 ab

Table 2. Effect of fertilizer applications and bacterial inoculations of lemon basil with plant growth promoting rhizobacteria on chlorophyll and anthocyanin contents, plant height, trunk diameter and branch number

*Proceedings of the* 2<sup>nd</sup> **International Conference on Food, Agriculture and Animal Sciences** (**ICOFAAS 2019**) *Antalya, Turkey, 8-11 November 2019* 

RC512+RC210+RC16	37.9±1.3 bc	5.18±0.55 ab	42±5.26 a-d	4.91±0.19 b-e	12.0±2.2 a-c
RC512+R601+RC16	36.8±2.8 cd	4.03±0.21 fg	39.75±3.5 a-d	4.85±0.15 с-е	12.0±1.8 a-c
RC512+RC210+		5 2(+0.1 -	40.75 4.56 - 1	5.02+0.45 a.c.	12511200
RC601+RC16	37.2±4.7 b-d	3.∠0±0.1 a	40./J±4.36 a-d	5,02±0.45 a-e	12.5±1.5 a-c

\*Bacterial strains, formulations and treatments are explained in Table 1; \*\*Values followed by different letters in a column were significantly different (p < 0.05) using Duncan's multiple range test.

IAA-producing bacterial inoculations provided better root growth for plant and improvements in plant growth and chlorophyll contents were the main mechanisms involved Inoculations, especially with the phytohormone-producing bacteria produced the highest root weights and total root numbers and encouraged adventitious-root formation. IAA stimulates excessive production of capillary roots hairs and lateral roots in the plant and promotes the release of saccharides from plant cell walls during the elongation process [19]. Auxins play important roles in plant growth and root development, hairy root growth and lateral root initiation. The IAA is the most common endogenous auxin which plays a role in root development, elongation both lateral and adventitious root formation, and it is a common product of L-tryptophan metabolism by several microorganisms including plant growth-promoting bacteria [20-24]. The positive effects of PGPR on plant growth were correlated with remarkable changes root growth and morphology, namely increasing the lateral root and root hair number and length. The elongation or proliferation of root systems has been suggested to be one of the mechanisms by which PGPR stimulate plant growth. PGPR promote root growth by increasing root surface area, which, in turn, promotes nutrient uptake, thereby indirectly stimulating plant growth positively [25]. Positive effects of these selected strains on fresh and dry herbage and root weight, chlorophyll contents, lateral root and branch number and trunk diameter of lemon basil plants showed the beneficial role of these PGPR, which might be attributed to IAA production, N<sub>2</sub>-fixation, P-solubilisation, or even other non-evaluated PGPR traits that stimulated the plant growth. Bacterial formulations can change the lemon basil seedlings root growth and can also affect the yield and growth parameters, but it was strongly dependent on the inoculant strain formulations and parameters evaluated.

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## Effects of Organic and Conventional Agriculture on Food Quality and Environmental Sustainability

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### Abstract

The key principles and practices of organic food production aim to encourage and enhance biological cycles within the farming system to maintain and increase long-term fertility of soils, to maintain genetic diversity of the production system, to consider the wider social and ecological impact of the food production and processing system, to produce food of high quality in sufficient quantity using biological and ecological processes, and to promote of a social, economic and ecologically sustainable production system. Organic farming systems rely on crop rotation, ecosystem management, integrated cropping and livestock systems, intercropping, crop residues, organic and bio-fertilizers and green manures, mechanical cultivation, non-chemical and biological control methods, high quality standards, and holistic production systems. Organic food production eliminates soil and water contamination; helps preserve local wildlife, conserve biodiversity and fight against global warming; encourages a natural balance within the ecosystem, reduces erosion, consumes less energy, and focus on the protection of all environmental components against agricultural-induced pressure. While conventional crop production methods have been linked to the negative effects on the environment and human health and safety, organic farming has potential for reducing some of the negative impacts of conventional agriculture on the environment and natural resources. Organic farming has potential for reducing some of the negative impacts of conventional agriculture on the environment. The majority studies of organic plant raw materials and their products from organic production systems indicate that they contain less nitrates, nitrites and pesticide residues and heavy metals, but more secondary substances, some vitamins and minerals, aromatic and phenolic compounds, anthocyanins, total sugars, organic and amino acids, vitamin C, polyphenols, flavonoids and carotenoids, essential amino acids and protein quality. This review is concerned with the summary and evaluation of the results of studies comparing the effects of organic and conventional production systems on the environment and the quality of foods produced with these systems.

**Keywords:** Organic farming, environment, sustainability, food quality, nutritional value, bioactive substances, energy use

## Introduction

Recent production and market trends show the importance that organics has gained in the recent years. Organic agriculture as a new agricultural development model which has less impact on

the environment and sustainable development had rapidly developed and promoted in the word [1]. Organic agriculture responds to a specific consumer demand for sustainable food products, promotes more sustainable farming practices and contributes to environmental protection and animal welfare improvement. As a matter of fact, organic production and demand for these products increased rapidly, the EU organic area increased by 70% in the last decade and organic retail sales reached EUR 34 billion in 2017 [2]. Organic agriculture is the most well-known form of sustainable agriculture due to food safety scares and health concerns associated with chemical fertilizers [3].

Organic products of plant origin are grown without the chemical-synthetic pesticides and largely without the use of readily soluble mineral fertilizers. It is a fact that there is a relationship between the different fertilization and plant protection methods of these two plant production systems and the nutritional composition of the plants. Organic food production is avoided by all non-organic farming methods such as pesticides, artificial fertilizers, antibiotics, growth hormones and so on uses all natural farming methods such as crop rotation, ecosystem management, integrated cropping, composting, organic and green manures, companion planting, intercropping, mechanical cultivation, stimulating biodiversity, and holistic production systems, etc [4,5]. In addition, organic food products are not allowed to contain any genetically modified ingredients nor artificial additives. Consequently, organic products do not pose any risk of pesticide residues and other potentially harmful chemicals.

Organic regulations, on the other hand, prohibit or severely restrict the use of food additives, processing aids and fortifying agents commonly used in nonorganic foods, including preservatives, artificial sweeteners, colorings and flavorings, and monosodium glutamate [6]. The main difference between organic and traditional food products is the chemicals involved in production and processing. Organic farming does not only strictly avoid the use of chemicals during plant growth, but also prohibits the use of any preservatives or chemicals to extend shelf life. The conventional food production practices involve the use of a number of chemicals, which have a destructive effect on the environment, while the residues of these chemicals in food products have suspicious effects on human health.

In short, organic agriculture is an agricultural system that aims to provide consumers with fresh, delicious and authentic foods while respecting natural life-cycle systems. Organic farming is a sustainable production system focused on producing food without environmental degradation, and consumer confidence in organic food in general is based both in reducing environmental impacts and health disease. Although conventional practices result in reliable high yield crops, there are concerns about adverse biological and environmental consequences and long-term sustainability associated with these practices. Sustainable agriculture means managing and conserving natural resource base continuously, meeting the needs of future generations

## **Environmental Benefits of Organic Food Production**

The organic production methods are focused on the protection of all environmental components against the pressure of the agricultural aspects. In general, organic food production eliminates soil and water contamination and risk of chemical residues, helps preserve local wildlife, conserve natural resources and biodiversity and fight against global warming. Organic production avoids the use of all synthetic chemicals, uses mixed plants as a natural pest control measure, promotes a natural balance within the ecosystem, and reduces carbon dioxide emissions and erosion. Strict avoidance of all chemicals in all stages of production makes

organic food much more environmentally friendly. It has been scientifically proven that the effects of pesticides and fertilizers cause serious harm to both the environment and human health, and there is no doubt that organic food production is environmentally friendly compared to conventional. Organic farming has been demonstrated to have less environmental impact and less exposure to pesticides residues, than conventional approaches [7]. These pesticides leads to contamination of the soil over the long term.Organic food production provides countless benefits to health, food, agriculture as well as the environment. Organic farming is the best way to protect the ecosystem and has some benefits, such as eliminates soil and water contamination, reduces erosion, fight against global warming, helps to conserve biodiversity and preserve ecosystem. In addition, Organic farming processes are widely thought to reduce agricultural greenhouse gas emissions with global warming potential. The manure used in the organic system has less nitrous oxide emissions than the conventional system [8,9], and in the conventional system post-harvest substances remaining in the field contain extra emissions [10].Organic food production provides countless benefits to health, food, agriculture as well as the environment. Organic farming is the best way to protect the ecosystem and has some benefits, such as eliminates soil and water contamination, facilitates healthy soil formation. consumes less energy, reduces the use of fossil fuels overall, reduces nitrogen run-off induced pollution, reduces erosion, increases soil organic matter, fight against global warming, helps to conserve biodiversity and preserve eco-system and native life. The most important environmental benefits of organic agriculture are: Maintains the quality of the soil, fosters biodiversity, promotes soil conservation, protects nature, reduces pollution from nitrogen runoff, avoids the heavy pesticide and herbicide use typical of conventional farming, uses less energy for a given yield than conventional farming, and stores more carbon in the soil, thus offsetting carbon dioxide emissions. The reduction of chemicals in the organic farming system encourages a natural balance and helps forestall domination of specific species over the others, all natural farming strategies have been shown to assist conserve diversity. The expansion of intensive modern conventional agriculture, with its monoculture crops and intense use of pesticides and herbicides, threatens endangered species.Organic farms, in contrast, use no herbicides, fewer pesticides, have more organic matter in the soil, and tolerate uncultivated areas. The organic system, however, is a haven for endangered plant species, insects, birds and animals, and it is known that the abundance and richness of plants, soil germs, worms, spiders, butterflies, insects, birds and mammal species tend to be higher higher in organic farm. Sustainable agriculture is closer to organic agriculture in terms of principles such as satisfy human food, enhance environmental quality and the natural resource base, sustain the economic viability of farm, make the most efficient use of nonrenewable resources and on-farm resources and integrate, natural biological cycles and controls, and also enhance the quality of life [11].

Conventional agriculture relies predominantly based on synthetic fertilizers, especially nitrogen, and worldwide, the use of nitrogen as fertilizer has increased tenfold in the last fifty years, with more than half entering rivers and other ecosystems affecting freshwater and marine environments. Increase in fertilizer use leads to changes in the global nitrogen cycle. With the intake of contaminated drinking water, human is exposed to nitrogenous fertilizers; nitrate, which is the final breakdown product of nitrogen fertilizers, accumulates in underground waters under agricultural land and may also be high in streams and rivers due to excessive flow of nitrogenous fertilizer from agricultural areas [12].

### The difference between organic and conventional foods

Since the use of synthetic fertilizers, pesticides, herbicides, irradiation, sewage droppings, hormones, antibiotics and genetic engineering is strictly prohibited, the popularity of organic foods is increasing day by day. While farmers using conventional methods might spray synthetic chemical fertilizers to promote plant growth, organic farmers would, instead apply biological and natural fertilizers such as fertilizers or compost to feed the soil and plants. On the other hand, the conventional farmer would use insecticides for pest control and herbicides for weed control, the organic farmer would make use of beneficial insects, birds or traps, and rotate crops, till the dirt and hand-weed or mulch to manage the weeds. Conventional agricultural practices utilize high-yield crop cultivars, chemical fertilizers and pesticides, irrigation, and mechanization. Considering the differences between organic and conventional foods, health risks associated with exposure to pesticides are one of the most important issues. Organic foods differ greatly from conventional ones, since the absence of pesticides, fertilizers and heavy metals residues because of regulated production rules applied. The main conclusion was that organic products had a higher dry matter and lower nitrate content and contained less pesticide residues [13].

Another important factor among the differences between organic and traditional foods is nutritional value, and many studies show that organic foods are richer in nutrients, especially organic acids and polyphenolic compounds, which have been shown to have human health benefits as antioxidants. Differences between the content of phenolic metabolites in organic and conventionally produced fruit, vegetables and other foods allows for the possibility that organically grown products may benefit human health better than corresponding conventionally ones [14,15]. In general, organic food does not contain artificial colors, flavors, sweeteners, preservatives or other additives.

Secondary plant metabolites such as phenolic acids and flavonoids play a critical role in human health and proper functioning of the human body depends on the appropriate compound content. According to Brandt et al. [16], organic ones contain higher levels of favorable secondary metabolites than corresponding conventional fruits and vegetables. A number of studies have actually shown that the content of phenols and polyphenols [17-24], vitamin C, and Fe, Mg, Zn, Cu, and P nutrient [14, 25,26], dry matter and total sugars [20, 27,28] are higher in organic products.

Overall, organic crops and foods have a higher antioxidant activity and contain higher concentrations of a wide range of nutritionally desirable antioxidants/polyphenolics, equal or higher nutrient minerals, but much lower levels of hazardous heavy metals. Research has confirmed low nitrate and nitrite content, especially in leafy vegetables, and higher antioxidant compounds in organically-grown fruits in comparison to conventional ones [29]. It is a fact that there is a clear trend towards low pesticide residues, nitrate and some heavy metal contamination in vegetables, fruits and nuts and their products from organic production [14].

Dangour et al. [30] concluded than conventionally produced crops had a significantly higher content of nitrogen. This explains the high content of nitrate in conventionally cultivated crops with high levels of mineral fertilizer, particularly in nitrophilic vegetables, green leafy, rooted and tuber vegetables. Fertilization increases the concentration of nitrogen compounds and protein-carbohydrate ratio, and decreases firmness and content of glucose in crop plants [15]. Research clearly shows higher nitrate and nitrite content in conventionally produced crops

compared to organic ones [13]. It is emphasized that this is the result of treatment with synthetic, easily soluble nitrogen fertilizers, which are absorbed in large quantities through the root system and leads to accumulation of nitrates in the leaves and other plant organs [31].

## Nutritional Value, Food Quality and Safety

Food quality is determined by its nutritional quality, which is describes the inherent biological or health value of a product including the ratio of beneficial to harmful substances, benefits, nutritive value, and taste, fragrance, freshness, and shelf-life as important quality characteristics influencing and governing consumer preference [32]. It is well known that different types and levels of fertilisation affect plant nutrient uptake and plant quality [33]. Foods obtained from organic and konvantraditional growing systems are mostly compared in terms of nutritional value, sensory quality and food safety. Organic food is not only healthy because it does not contain any synthetic chemical residue, but it is also more nutritious. The main advantage of organic food is without a doubt absence of all non-natural ingredients. The advantages of organic production are not limited to consumption and nutritional benefits, but the improved taste and higher nutritional value of organic products are the most common reasons. Many organic farming methods are more beneficial for human health than their conventional counterparts. A report by Yu et al. [26] showed that organic products tasted better; the percentage of leanness was higher, and the products tasted much tender, and dry matter content, vitamin C, anthocyanins, isoflavones, carotenoids, other phenolic compounds, more elements such as P, Fe, Mg, Zn, and Cu higher. In addition, it has been reported that the microbiological quality and safety of fresh vegetables are not affected by organic or conventional farming practices [34,35,36]. Also, organic foods must meet the food safety requirements unlike its conventional counterpart [37]. Winter and Davis [38] reported that organic food is better than a conventional one, as regards safety and nutritional quality.

It is a fact that the main problem with organic agriculture, in terms of food security, is that it produces lower yields than conventional systems. This makes it difficult to produce enough food to feed an entire population affordably with organic crops and livestock. On the positive side, organic does have benefits for promoting food security in the developing world. Organic farming requires fewer and less expensive inputs leading to a greater level of self-sufficiency for low-income rural farmers. If crop diversity is high, organic agriculture can also reduce the risk that farmers will lose an entire crop due to pests or disease. Organic farms in rural areas can create jobs for inhabitants of rural communities and deter mass migration to urban areas.

## Conclusions

Organic food is generally considered as having healthier properties as consequence of ecofriendly management. The present review based on available scientific literature highlights that organic plant raw materials and their products from organic production indicate that they contain fewer nitrates, nitrites and pesticide residues, antibiotics, food additives, industrial pollutants, and heavy metals, but more secondary substances, some vitamins and minerals, nutrient content, aromatic and phenolic compounds, carotenoids, anthocyanins, carotenoids, total sugars and dry matter, organic and amino acids, certain antioxidants, and essential amino acids. The flavor of organic fruits and vegetables may be more intense compared to conventional materials. From the consumer's point of view, raw materials obtained from organic production in terms of pesticides and nitrates are certainly safer than conventional ones.

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## The Ameliorative Effects of Exogenously Applied Chitosan on Growth and Physiological Parameters of Tomato (*Solanum lycopersicum* L.) Seedlings under Drought Stress

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### Abstract

In recent years, drought has become an important factor affecting the renewal of vegetation and the sustainability of agriculture. Severe water restriction due to global warming and population growth in the future is expected to be threaten world plant production. In this study, the possibility of mitigating drought stress tolerance of tomato during seedling period by application of exogenously chitosan (CS) was investigated. Tomato seedlings were subjected to drought stress for 36 days in three irrigation schedules (100%, 80%, %50 of field capacity) after spraying CS at three different concentrations (0, 0.5 or 1.0 mg/l). Drought stress has caused a significant reduction in tissue relative water content (TRWC), plant fresh weight, plant dry weight, root fresh weight, root dry weight, and chlorophyll a, chlorophyll b and total chlorophyll content, while increase in membrane permeability (EC). However, the application of exogenous CS counteracted the drought stress-induced adverse effects by increasing TRWC, plant fresh weight, plant dry weight, plant dry weight, root fresh weight, root dry weight, root dry weight, chlorophyll a, chlorophyll b and total chlorophyll b and total chlorophyll content, while reducing EC. Therefore, in can be concluded that CS applications as a leaf spray could be used as an ameliorative for tomato seedlings against the harmful effects of water stress.

Keywords: Solanum lycopersicum L., Water stress, Chitosan, Physiological response

## Introduction

Tomato (*Solanum lycopersicum* L.) is the most grown vegetable in the world with 182 million tons of production in 5 million hectares of production area (FAOSTAD 2017). The Tomato is not only one of the most widely grown important crops, but it is also a model crop for plant research (Zhou et al. 2019). Although differences between tomato varieties have been reported (Rus-Alvarez and Guerrier, 1994; Cano et al., 1996; Alian et al., 2000; Sánchez-Rodríguez et al., 2010), tomato generally is considered susceptible to abiotic stress conditions (Ghanem et al., 2016). Agricultural drought is a major environmental stress to hinder the productivity of cultivated crops worldwide (Noaman et al., 2004). Studies show that climate change caused by global warming will cause more severe, permanent and widespread droughts in the future (Dai 2013; Huang et al., 2016; Mukherjee et al., 2018). Chitosan, (1-4) -2-amino-2-deoxy- $\beta$ -d-glucan, is a deacylated derivative of chitin after cellulose, the most abundant polysaccharide on earth. Chitosan has been reported to have very positive impact on plants growth, improving

roots and shoots under normal and stress conditions (Chmielewski et al., 2007; Katiyar et al., 2015). Zeng and Luo (2012) noted that exogenous chitosan treatment improved leaf water content, chlorophyll concentration, photosynthesis performance, and organic matter accumulation of wheat plants under drought stress. Furthermore, the same researchers reported that chitosan could effectively stimulate the development of the root system by reducing the inhibition of root growth and strengthen its ability to absorb water, thus increasing the plant's resistance to drought. In the literature survey, different plants such as canola (Li et al., 2008), wheat (Zeng and Luo, 2012), potatoes (Jiao et al., 2012), pepper (Bittelli et al., 2001) were tested for the use of chitosan to alleviate the effects of drought stress. However, the effects of chitosan on tomato seedlings under drought stress have not been yet examined in detailed. The objective of this study was to determine the impact of different concentrations of CS on improving drought tolerance of tomato seedlings, as measured by selected growth and physiological properties.

## Material and Methods

This study was conducted as a pot experiment under controlled greenhouse conditions at Ataturk University, Erzurum, between May and August, 2019. Tomato seedlings (*Solanum lycopersicum* L. cv. Falcon) were used as plant material in the experiment. Tomato seeds were sown into multi-celled trays. Three true leaf seedlings were transplanted to 14 L plastic pots (720x195x155 mm in size) filled with peat, perlite and soil (v:v:v; 2:1:2). The pots were placed randomly on the benches in the greenhouse. The study was done using a factorial experiment in a randomized complete block design with three replications and 3 plants per repeat. CS treatments (0, 0.5, 1.0 mg/L) prepared in ultrapure water with Tween 20 (Sigma Chemicals) were sprayed onto upper and under of leaves a day after transplanting, and then every week regularly until harvest.

Volumetric water measurements were done with Wet-2 sensor (Delta-T Devices, UK) which was accurately suitable for growing media. The planned irrigation program was started on June 19 and continued at intervals of 3 or 4 days. Drought treatment in pots was continued by controlling ambient humidity using Wet-2 sensor. The volumetric moisture content of the pots was monitored daily. Water issues were generated by providing 100% (D0; control), 80% (D1) and 50% (D2) of the water required to reach the field capacity.

**Membrane permeability (MP):** 0.5 g weighed leaf samples were put in 50 mL glass vials for the measurement of membrane permeability. Vials were then filled with 30 mL of distilled water and allowed to stand in the dark for 24 h at room temperature. The EC (EC1) of the bathing solution was obtained at the end of the incubation period. Then these vials were autoclaved at 121 °C with 15 psi for 10 min. to release all the electrolytes from the cells. Vials were allowed to cool at room temperature and EC (EC2) was re-measured. EC % was calculated by following formula (Kaya et al. 2003).

## MP = (EC1 / EC2)x100

**Leaf relative water content (LRWC)**: Two young fully opened leaves of the plant was cut. The fresh weight (FW) of the sample leaves was recorded and the leaves were immersed in distilled water in a Petri dish. After 24 h, the leaves were removed, the surface water was blotted-off and the turgid weight (TW) recorded. Samples were then dried in an oven at 70°C until reach constant weight and dry weights of leaves have been recorded. Leaf relative water content was calculated using the following formula (Turner, 1981):

 $LRWC = [(FW - DW)/(TW - DW)] \times 100.$ 

**Chlorophyll content:** The leaf tissue which weighed 0.3 g (W) first in a dry form for 5 minutes, then homogenized with TissueLyser LT (Qiagen) homogenizer in 1.6 ml acetone for 20 minutes. The resulting solution was taken into a 2 ml ependorph tube with acetone, completed to 2 ml (V) and shaken thoroughly. Samples were readed at wavelengths of 663 nm and 645 nm by placing in the supernatant spectral tub after when centrifugation at 4 °C, at 6000 rpm,15 minutes for chlorophyll determination. The values obtained were calculated according to the following formula and chlorophyll a, chlorophyll b and total chlorophyll values were recorded as mg /L. (Arnon,1949; Strain and Svec, 1966; Lichtenthaler 1983; Lichtenthaler 1987; Demiralay et al., 2019).

*Chlorophyll a* (*Cl a*) = [12.7x(A663) - 2.69x(A645)]x V/W x1000

*Chlorophyll b* (*Cl b*) = (22.91x (A645) – 4.68x(A663)]x V/W x1000

**Total Chlorophyll** = Cl a + Cl b

*Growth parameters:* Shoots and roots were separated and their fresh weights were directly determined. For dry weight determination, the shoots and roots were dried at 70 °C for 48 h.

**Statistical calculations:** Analysis of variance was done by IBM SPSS (version 25.0) software package and the comparison of means was carried out using Duncan's multiple range test.

#### **3. Results and Discussion**

The results showed that drought stress negatively affected all parameters examined (Figure 1:2:2:4). In the study, it was observed that D2 drought stress decreased plant fresh weight, root fresh weight, plant dry weight and root dry weight decreased by 52.7%, 46.8%, 45,4%, 48,1% respectively. However, exogenous CS treatments improved these parameters under drought stress compared to the control. At the highest drought level (D2: 50% of field capacity), 0.5 mg/L doses of CS mitigated significantly to negative effect of drought stress on the fresh and dry weights of plant and root samples (Figure 2 and 3). Bittelli et al. (2001) stated that foliar application of chitosan, by reducing plant transpiration, reduced in water use of pepper plants by 26–43%, while maintaining biomass production and yield. On the other hand, positive effects of CS (0,5-1.0 mg/L) on MP, LRWC, chlorophyll a, b and total chlorophyll content were observed in tomato seedlings under drought stress. MB increasing as the level of drought increases, decreased 23.6% in D1 and 14.1% in D2 with 1.0 mg/L CS leaf treatment, compared to the control group. However, the LRWC decreasing as the drought level increases alleviated with the best 1.0 mg / L CS application at D1 drought stress while at D2 drought stress with the best 0.5 mg / L CS application, respectively 20.9%, 19.4% (Figure 1). Ors et al. (2016) determined that the water deficit conditions during seedling period caused an increase in MP but a decrease in LRWC in squash. Earlier researchers reported that chitosan treatment could reduce the inhibition of roots and stem growth due to drought stress, thus strengthening the capability of water absorption (Zeng ve Luo, 2012; Katiyar et al., 2015). CS treatment counteracted the adverse effect of drought stress on chlorophyll content (Figure 4). Particularly foliar application of CS 0.5 mg / L, chlorophyll a, b and total chlorophyll content increased by 11.8%, 6.3% and 12.5%, respectively, according to control group. In another study, content of

chlorophylls in the leaves of coffee seedlings grown under drought conditions enhanced strongly up to 46.38–73.51% compared to the control with chitosan oligomer treatments, thus were mitigated the effects of drought (Dzung et al., 2011).

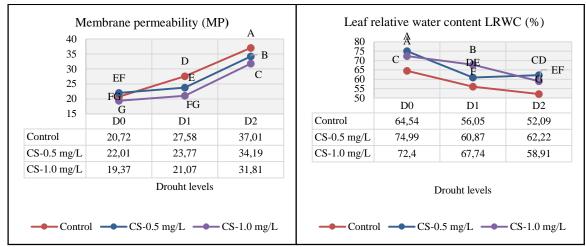
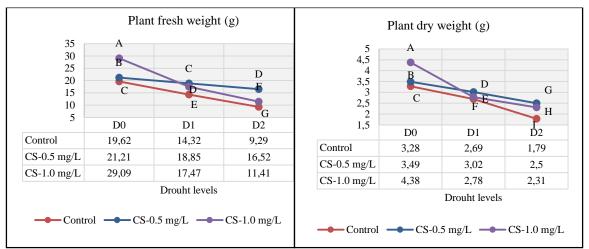
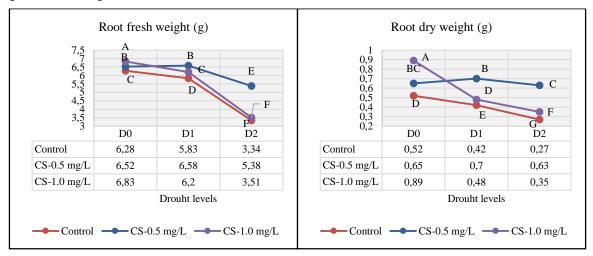
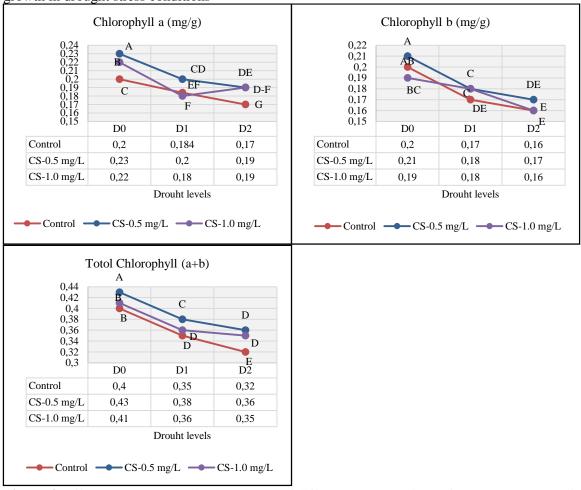


Figure 1. Effects on MP and LRWC of different concentrations of CS on tomato seedling growth in drought stress conditions



**Figure 2.** Effects on plant fresh and dry weight of different concentrations of CS on tomato seedling growth in drought stress conditions





**Figure 3.** Effects on root fresh and dry weight of different concentrations of CS on tomato seedling growth in drought stress conditions

**Figure 4.** Effects on Chlorophyll a, b and total of different concentrations of CS on tomato seedling growth in drought stress conditions

## Conclusion

In conclusion, CS can be used an alternative treatment to ameliorate the negative effects of drought stress in tomato seedlings.

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## Production of beans (Phaseolus vulgaris L.) in Turkey

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#### Abstract

Beans, which are consumed for dry and immature seeds with fresh and dry pods, hold a special place in developing countries. The aim of the study is to give information about bean production, World Trade and Exchange. The research data was obtained from Global Food and Agriculture Statistics of FAO (FAOSTAT), Turkish Statistical Institute (TurkStat) and International Trade Centre (ITC) records by scanning, filtering and grouping and summarized in charts and graphs. According to the findings, fresh and dry beans are produced in a total area of 136 947 ha in Turkey. With 630 thousand tons of fresh bean production, our country meets 2% of the world's fresh bean production and 239 thousand tons of dried bean production 1%. According to data obtained from the ITC, beans of Turkey's imports was 41.2 million U.S. dollars, it was reported that while our exports reached 23.5 million U.S. dollars. In conclusion, it can be expressed that Turkey has an important place in the World in regard to bean production, consumption and trade.

Keywords: Phaseolus vulgaris L., Turkey, Production, Imports, Export

#### Introduction

Turkey is located between 36° and 42° N and from 26° to 45°E, and is characterized by mountains in the central area, and flat coastal plains in other locations. Turkey is very rich with respect to plant diversity and is lucky with respect to sustainability of agriculture because of the advantages provided by its geographic diversity [1; 2; 3]. It also is one of the richest countries of the world in terms of plant diversity due to its geographical structure and climate conditions and one of the rare countries located on 2 gene centers (Near East and Mediterranean) in the world [4]. The Turkish flora contains over 10,000 plant species and many of them are endemic [5; 6]. Turkey grows a lot of agricultural products which are important in world commerce; including many leguminous plants [7]. Among edible legumes crops, bean (Phaseolus vulgaris L., 2n=2x=22) is a self-pollinated crop species [8]. Archeo-morphological, biochemical and molecular evidence suggest that bean was independently domesticated in Andean and Mesoamerican centres [9; 10; 11]. Beans are believed to have been brought together with maize to Mediterranean coast by Portuguese and Spanish traders in the 16th and 17th century [12; 13] and then introduced to Anatolia in the 17th century [1]. Asian traders were responsible for the introduction of beans into Turkey from Europe. Since then beans have been grown as populations in different climates and geographies of Turkey and hundreds of natural hybrids occurred in this growing system and it hosts to local landraces of bean in different geographical provinces [14; 15]. One result of this early dispersal of bean worldwide, however it may have happened, is the development of potential secondary centers of diversity in Asia and Europe [16; 17]. The bean are the first among the edible grain legumes in terms of harvested area and production in the world. It is also widely consumed as fresh vegetables besides dry grains. The bean is an agricultural crop cultivated for fresh pods and for dry seed all over Turkey, and is regarded as a national food. It is a traditional crop often grown in small gardens for its pod and

in large fields as legume for its dry seeds. The bean is cultivated primarily for its green pods, green shelled seeds, and dry seeds. The cultivars for green-pod harvests are also called French, garden, green, snap, or stringless bean [18]. The bean has now secured a unique place in Turkish agricultural and culinary systems. The annual bean production of Turkey highlights its importance in Turkish economy and diet [14]. Bean is also a crop of considerable global importance as a grain legume plant with the largest cultivation area in the World [1]. It is an important grain legume throughout the world providing a source of protein, dietary fibre, starch and minerals such as potassium, thiamine, vitamin B6 and folic acid in diets affordable by the poor [19; 20]. In many parts of the world, it is an underutilized staple food crop with remarkable yet unappreciated potential to reduce the risk for chronic diseases such as obesity, diabetes-type II, cardiovascular disease and cancer [21]. Bean are very important for the agriculture of the country in terms of human food, animal nutrition and the additional nutrients it provides for subsequent crop production. Especially in countries with high meat prices, it has an important place in human nutrition in terms of being a source of cheap protein. Beans are globally important crop and appeal both to farmer and consumers

## **Current Situation of Bean Production in the World and Turkey**

According to data from the 2017 FAOSTAT, world produced 24.2 million tons of fresh beans in 1.6 million ha areas, while dried beans produced 31.4 million tons in 3.6 million ha areas [22] (Table 1; Figure 1;2). Turkey produced 630 thousand tons of green beans in 48.9 ha area and 239 thousand tons of dry beans in 89.7 ha area during the same period [22] (Figure 1;2). Considering the bean cultivation continents, the largest cultivation area and production is located in the Asian continent. However, the highest grain yield is obtained in the America continent. According to the 2017 data of the World Food and Agriculture Organization, countries with a bean yield of more than 3 tons/ha; Ireland, Montenegro, Barbados, Tajikistan, Iraq, Belgium, Sudan, Latvia, Netherlands, Lithuania, Libya, Syrian Arab Republic and South Sudan. Turkey's dry bean yield was recorded as 2.67 tons/ha. The dry bean yields of Angola, Panama, Zimbabwe, India, Djibouti, Lesotho, Somalia and Eritrea have been reported to be lower than 0.5 tons/ha. Although the yield of the crop is very low per hectare, the largest dry bean producer country is India (20.4% of world production). Myanmar (17.4%), Brazil (9.7%), United States of America (5.2%), China mainland (4.2%), Mexico (3.8%), United Republic of Tanzania (3.6%), Uganda (3.3%) are the other major dry bean producer countries. Turkey producing 0.8% of the world's dry bean production, it was 22th ranked. According to FAO 2017 green bean production data, China provided to 79.6% of world production. As well as Indonesia (3.8%), India (2.8%), Turkey (2.6%), Thailand (1.3%), Egypt (1.2%) are the other major producers of fresh beans. Green bean production of Turkey (630 thousand tons) took place 4th ranking according to this statistical data (Table 1). Luxembourg achieves a high yield from other countries with 40 tons/ha of fresh beans. Poland, Morocco, Ukraine and Kuwait are the countries with yield higher from 20.0 tons/ha, while China (mainland) is the second highest country with 28.32 tons/ha fresh beans yield. In addition, Turkey green bean yield was determined as 13.34 tons/ha. On the other hand, the lowest yield has been observed in the countries of Montenegro, Thailand, Puerto Rico, Finland, Serbia, Lithuania, Ecuador, Timor-Leste, Slovakia, Costa Rica, North Macedonia, with yields of less than 2 tons per hectare.

Area	Dry bean (tons)	Area	Green bear (tons)
World	31405912	World	24221252
India	6390000	China, mainland	19281055
Myanmar	5466166	Indonesia	915591
Brazil	3033017	India	675188
United States of America	1625900	Turkey	630347
China, mainland	1322214	Thailand	313596
Mexico	1183868	Egypt	283520
United Republic of Tanzania	1140444	Morocco	168887
Uganda	1024742	Spain	163649
Kenya	846000	Italy	154851
Ethiopia	543984	Bangladesh	137495
Kazakhstan	456044	Belgium	123113
Rwanda	455822	Mexico	98572
Argentina	413605	Algeria	98003
Cameroon	413072	Sri Lanka	75787
Burundi	379861	Iran (Islamic Republic of)	61676
Angola	375494	Greece	60200
Belarus	351725	Romania	55621
Democratic People's Republic of Korea	318376	Canada	53950
Canada	256835	France	53492
Guatemala	251735	Germany	51469
Indonesia	244000	Chile	42948
Turkey	239000	Australia	41373
Other Countries	4674008	Other Countries	680869

 Table 1. Bean production data of the world and some important countries [22]

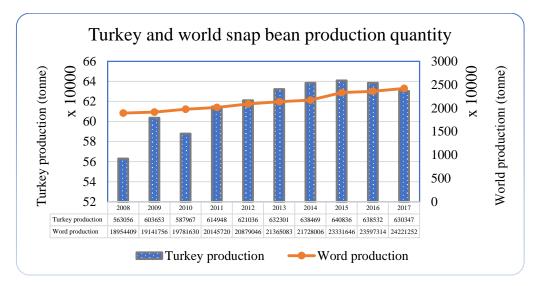


Figure 1. Turkey and world snap bean production quantity (Last ten years, 2008-2017) [22]

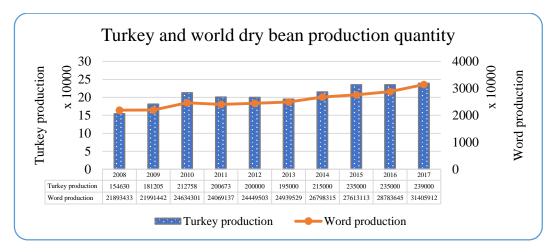


Figure 2. Turkey and world dry bean production quantity (Last ten years, 2008-2017) [22]

	Dry bean in Turkey				Green bean in Turkey		
Year	Production Area (da)	Production Quantity (tons)	Yield (tons/da)	Production Area (da)	Production Quantity (tons)	Yield (tons/da)	
2009	947509	181205	0.19	535172	603653	1.13	
2010	1032549	212758	0.21	531340	587967	1.11	
2011	944980	200673	0.21	528931	614948	1.16	
2012	930904	200000	0.21	528506	621036	1.18	
2013	846912	195000	0.23	506619	632301	1.25	
2014	904496	215000	0.24	501767	638469	1.27	
2015	935433	235000	0.25	501218	640836	1.28	
2016	885476	235000	0.27	495639	638532	1.29	

Table 2. Turkey bean production area, production quantity and yield [23]

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2017	896778	239000	0.27	489392	630347	1.29
2018	847863	220000	0.26	455263	580949	1.28
2009-2018 (%)	-10.52	21.41	35.68	-14.93	-3.76	13.13
2017-2018 (%)	-5.45	-7.95	-2.64	-6.97	-7.84	-0.93

Turkey producted 220 thousand tons of dry bean in the area 848 thousand decare in 2018. According to the regions, dry bean cultivation area is most common in the Central Anatolia region, followed by the Black Sea region and the Aegean region. According to TurkStat (2018) data, since 2009 the production area of dry beans has been reported to have decreased by 10.52%, while the production amount has increased by 21.41% [23]. Compared to the previous year (2017), production area and quantity decreased by 5.45% and 7.95%, respectively. While the bean yield increased by 35.68% compared to 2009, it decreased by 2.64% compared to 2017. According to TurkStat data in 2009, Turkey's largest dry bean producer provinces was Konya (51 477 tons), Karaman (17712 tons), Erzincan (12009 tons), Nigde (8919 tons), Istanbul (7581 tons) and Kütahya (5739 tons). But, dry bean production of Konya (53439 tons), Niğde (36398 tons), Karaman (30193 tons), Bitlis (19685 tons), Nevşehir (18232 tons) provinces were found to be more in 2018. In this product year, a significant increase was observed in Bitlis (1849%), Nevsehir (1544%), Niğde (308%) and Karaman (71%) provinces. These four provinces, which have more than half (72%) of the dry bean production in Turkey, show that production is concentrated in certain regions. In addition, in the same year, the production amount decreased by 82% in Kırsehir, 73% in Erzincan, 65% in Samsun, 63% in Tokat, 44% in Aksaray, 39% in Kütahya, 38% in Kahramanmaraş and 33% in Balıkesir [23] (Figure 3).

Turkey produced 581 thousand tons of green bean in the area 455 thousand decare in 2018 [23]. The production area of green beans has decreased by 14.93% and the decrease in production amount is 3.76% according to 2017 production years. However, according to 2017 production years, production increased 4.4% since 2009. According to TurkStat (2018) data, since 2009 the yield has increased by 13.13%, however, it has decreased by 0.93% compared to the previous year. In terms of 2018 green bean production area, Izmir (37270 da), Bursa (36102 da), Mersin (34480 da), Antalya (30357 da), Tokat (26513 da), Samsun (24642 da), Hatay (16642 da), Konya (14360 da), Muğla (13438 da), Burdur (12889 da) and Balıkesir (11546 da) cover 57% of the total production area. The production area of the remaining provinces is below 10 thousand decares. Compared to the previous year, the production of green bean decreased significantly in Samsun (69%), Karaman (32%), Afyonkarahisar (17%), Van (17%) and Ankara (15%) provinces with significant levels of production (Figure 4). On the other hand, production in Antalya (14%), Izmir (14%), Muğla (5%), Burdur (6%) and Mersin (8%) has increased significantly [23] (Figure 4).

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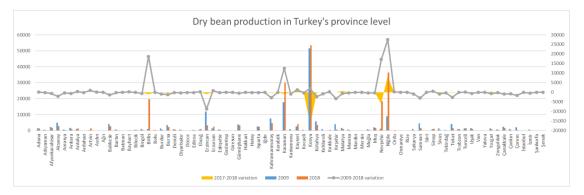


Figure 3. Dry bean production in Turkey's province level [23]

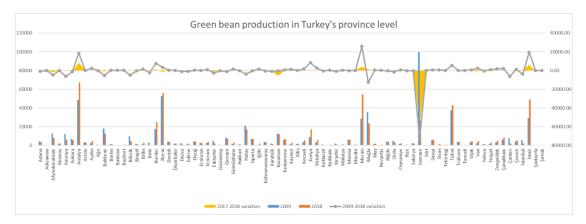


Figure 4. Green bean production in Turkey's province level [23]

#### Turkey's bean exports and imports

Turkey imports approximately in worth 41.2 million U.S. dollars of bean [24]. The vast majority of beans imported from Turkey are exported from Canada (25.2%), Kyrgyzstan (22.6%), Argentina (20.2%), the United States (USA) (12.8%) and China (10.7%) (Figure 5;7; Table 3). Turkey imports 37.4 thousand tons of beans and pays 1.103 USD per unit. Between 2014-2018, the amount of imports decreased by 4%, while the value of imports decreased by 12% [24]. Egypt, USA and Kyrgyzstan have seen a decrease in the value of bean imports, whereas in France, Argentina, China and Spain have seen an increase in the value of imports in the last four years (Figure 8; Table 3). According to export data for 2018, Turkey has exports of beans of 23.5 million U.S. dollars. While the export value of beans is 1223 USD, the amount of exports in the last year is around 19.3 thousand tons. Iraq (25.7%), Iran (20.8%), Libya (16.4%) and Germany (7.1%) are the largest exporting countries importing beans from Turkey (Figure 6;8; Table 4). Since 2014, the export value and quantity of beans have increased by 14% and 20% respectively [24]. In the last 4 years, Turkey's Bean importing countries, especially Libya and Iran, have significantly increased the amount and value of imports. But, import indicators of Spain, Italy, Algeria and the Syrian Arab Republic have decreased during the same period.



Figure 5. Countries imported beans by Turkey [24]



Figure 6. Countries exported beans by Turkey [24]

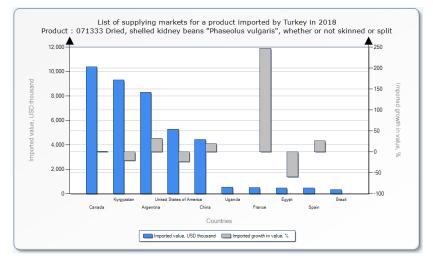


Figure 7. Bean imported growth (%) between 2014-2018 and 2018 imported value in Turkey [24]

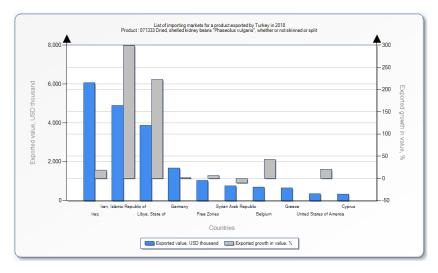


Figure 8. Bean exported growth (%) between 2014-2018 and 2018 exported value in Turkey [24]

 Table 3. Turkey's important bean exporters and some trade indicators [24]

Exporters	Value imported in 2018 (USD thousand)	Share in Turkey's imports (%)	Quantity imported in 2018 (Tons)	Unit value (USD/unit)	Growth in imported value between 2014-2018 (%)	Growth in imported quantity between 2014-2018 (%)
World	41238	100	37385	1103	-12	-4
Canada	10395	25.2	9959	1044	1	9
Kyrgyzstan	9304	22.6	9721	957	-20	-9
Argentina	8317	20.2	7577	1098	32	47

United States of America	5296	12.8	4407	1202	-23	-17
China	4431	10.7	2951	1502	20	27
Uganda	545	1.3	519	1050		
France	510	1.2	111	4595	247	225
Egypt	471	1.1	493	955	-60	-54
Spain	468	1.1	321	1458	27	98
Brazil	324	0.8	283	1145		
Ukraine	269	0.7	273	985	46	61
Ethiopia	207	0.5	288	719	-19	-11
South Africa	167	0.4	120	1392		
Peru	137	0.3	76	1803	-72	-48
Poland	98	0.2	88	1114		
Uzbekistan	85	0.2	91	934		
Taipei, Chinese	69	0.2	15	4600	7	3
Madagascar	38	0.1	44	864	-37	-52
Tanzania, United Republic of	35	0.1	11	3182	74	8
Italy	31	0.1	9	3444	18	32

 Table 4. Turkey's important bean importers and some trade indicators [24]

Importers	Value exported in 2018 (USD thousand)	Share in Turkey's exports (%)	Quantity exported in 2018 (Tons)	Unit value (USD/unit)	Growth in exported value between 2014-2018 (%)	Growth in exported quantity between 2014-2018 (%)
World	23589	100	19291	1223	14	20
Iraq	6057	25.7	5372	1128	19	29
Iran	4898	20.8	3020	1622	299	225
Libya, State of	3868	16.4	3241	1193	223	382
Germany	1681	7.1	1155	1455	2	15
Free Zones	1030	4.4	1589	648	6	29
Syrian Arab Republic	761	3.2	803	948	-10	1
Belgium	683	2.9	465	1469	43	54

Greece	646	2.7	585	1104	-	-
United States of America	354	1.5	220	1609	20	37
Cyprus	330	1.4	262	1260	-	-
United Kingdom	288	1.2	202	1426	10	21
Italy	280	1.2	221	1267	-31	15
Lebanon	243	1	209	1163	26	29
Sweden	231	1	180	1283	12	28
Serbia	219	0.9	184	1190	13	-15
Algeria	211	0.9	140	1507	-23	-21
Tunisia	180	0.8	203	887	-	-
Spain	175	0.7	84	2083	-43	21
Qatar	121	0.5	104	1163	64	53
Jordan	119	0.5	112	1063	56	118

## Conclusion

Bean production has shown a growing chart in the world and in Turkey. While the area of production has decreased, the main reason for the increase in the amount of production were improved cultivars, irrigation, nitrogen fertilizers, and control of diseases, insects or weeds contributing to increased yields of annual crops during the 20th and 21th century. Although bean was introduced to Turkey quite late, it had adapted quite well thanks to the diversity in its geographical and climatic structure. It can be produced in almost all provinces of Turkey in a comfortable way. The high value of our unit exports is also an indication that Turkey produces quality beans. The total import value is considerably higher than Turkey's total exports. Turkey may be in a much better state in its current state of bean production. For this, appropriate strategies on bean production can be developed. In conclusion, it can be expressed that Turkey has an important place in the World in regard to bean production, consumption and trade.

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## Stability of polyphenoloxidase and peroxidase activities of Deglet Nour and Ghars dates at different pH and temperatures

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## Abstract

Effect of pH and temperature on polyphenoloxidase and peroxidase activities was studied to highlight the enzymatic browning which limits the conservation of Deglet Nour and Ghars dates by leading to the loss of their nutritional quality. Stability rates at wide ranges of pH (2.4 - 8.6) and temperatures (10 - 90 °C) were assessed by assaying the residual enzymatic activities of PPO and POD from both varieties. PPO's and POD's activities of Deglet Nour and Ghars date fruits are highly inhibited at extreme pH and temperatures but remain significant over a wide range. PPO presented a maximal stability at pH 7.2 for the two varieties, while POD at pH 7.2 for Deglet Nour and 6.4 for Ghars. 35 °C was the temperature of maximal stability for both enzymes of the two varieties.

Key words: Phoenix dactylifera, polyphenoloxidase, peroxidase, pH, temperature

## Introduction

Date fruit quality and nutritional value have been reported in many research studies. They are rich in simple sugars like glucose and fructose; they are a good source of fibers, some essential vitamins and minerals. They also contain phenolic compounds with number of therapeutic virtues. Indeed, these molecules have recently attracted increased attention in the field of nutrition and medicine because of their potential properties and health benefits. However, phenolics are subject to enzymatic oxidation leading to browning which causes fruit color changes. The conservation of Deglet Nour (DN) and Ghars (Gh) dates is limited by enzymatic browning inducing the loss of nutritional quality of fruits as a result of the increased lack of post-harvest processing and preservation technologies. Enzymatic browning is a physiological phenomenon essentially involving activity of polyphenoloxidase (PPO) and peroxidase (POD) enzymes; it deteriorates the sensory and organoleptic properties of date fruit. Although browning is desirable during fruit development, its continued process after maturation and harvesting leads to damage and wastage (Daas Amiour and Hambaba, 2016).

This motivated us to study the effect of some physical parameters (pH and temperature) on PPO and POD activities of Deglet Nour (DN) and Ghars (GH) date varieties in order to better understand this phenomenon.

## Materials and methods

## Plant material

The plant material used in this study consists of two varieties of Algerian dates, which are widespread in taste quality, market availability and wide consumption nationally and internationally. These varieties are Deglet Nour date which holds the monopoly of the world markets and Ghars date, the famous star of the Algerian pastry. The fruits were harvested at full maturity, having reached the final stage of development and ripening of date the 'Tamr' stage. All the fruits used had homogeneity of maturity defined by the aspect and the color of the pericarp and thus homogeneity of consistency.

## Methods

Effect of pH on activities of the two enzymes polyphenoloxidase (PPO) and peroxidase (POD) was studied at 30 ° C in the presence of pH buffers ranging from 2.4 to 8.6. Stability at different pH was determined by assaying the residual enzymatic activities after maintaining the enzyme extracts for 12 hours at 10 ° C and at different pH values. The influence of temperature on the activities of the two enzymes (PPO) and (POD) was also studied at pH 7 by incubating the reaction mixture at several temperatures ranging from 10 to 90 °C. The thermal stability was evaluated after incubation of the enzyme solution at different temperatures (10-80 °C) for 60 minutes before the assay.

For each variety of dates, three replicates were made for each test, starting with samples taken from different fruits, chosen randomly.

# Extraction of polyphenoloxidase (PPO) and peroxidase (POD) enzymes and determination of their residual activities

For each replicate, 2 g of the date paste of each variety was homogenized in 8 ml of 5 cmol L-1 of phosphate buffer (pH 7.0) and 0.2 g of polyvinylpyrrolidone. After filtering the homogenate through a cotton cloth, the filtrate is centrifuged for 20 min at 12,000 x g and 4  $^{\circ}$  C; this extraction was carried out as described by Duan et al. (2007). The supernatant was used as a crude extract for the determination of the activities of both PPO and POD enzymes.

PPO activity(A) was assayed by measuring the oxidation of methylcatechol as substrate at 410 nm, according to the procedure described by Jiang et al. (2005); 1.0 ml of 0.1 M phosphate buffer (pH 6.8), 0.5 ml of 0.1 M 4-methylcatechol, and 0.5 ml of enzyme solution. The increase in absorbance at 410 nm at 30  $^{\circ}$  C was automatically recorded for 5 minutes. One unit of enzymatic activity is defined as the amount of enzyme that caused a change in absorbance of 0.01 per minute.

POD activity(A) was determined by the method of Zhang et al. (2005), using guaiacol as a substrate, in a 3 ml reaction mixture, containing 25  $\mu$ l of enzyme extract, 2.78 ml of 5 cmol L-1 phosphate buffer (pH 7.0), 0.1 ml of 1% hydrogen peroxide and 0.1 ml of 4% guaiacol. The reaction was carried out at 30 °C.

The increase in absorbance at 470 nm, due to the oxidation of guaiacol, was recorded for the first 5 minutes of the reaction. One unit of enzymatic activity is defined as the amount of enzyme that caused a change of 0.01 per minute of absorbance. The results are expressed in enzymatic unit per gram of the fresh date pulp (EU  $g^{-1}$  PF).

The residual enzyme activity of PPO and POD after treatment at different pHs and temperatures was calculated by the same relationship given by Bayindirli (2010) for the calculation of residual enzyme activity in heat-treated foods; it is expressed as a fraction of the initial activity (A<sub>0</sub>): Residual activity = A / A<sub>0</sub>

#### Statistical analysis

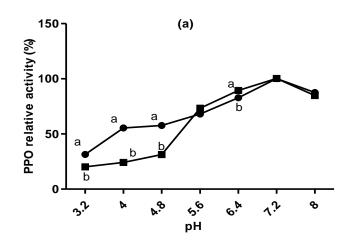
Statistical analysis was performed using Graph Pad PRISM version 5.01. All analytical determinations were made in triplicate. Data were expressed as mean  $\pm$  standard error. Means were compared by t-test at a significance level of 0.05.

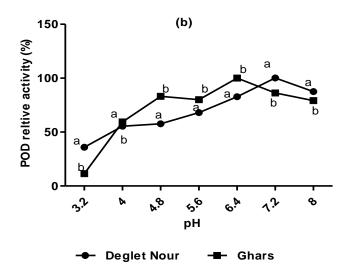
#### **Results and discussion**

#### pH effect

The enzymatic activity measurement revealed, in the case of the PPO of the two varieties, a high residual activity between pH 5.6 and 8. At pH values below 3 and 5.6 this activity decreases significantly, respectively, for Ghars and Deglet Nour. However, it remains considerable at pH 8 for both varieties, so that the enzyme retains 80% of its activity. The stability of the PPO at different pH reaches a maximum at pH 7 (Figure 1 a).

The determination of the residual activity of Ghars date's POD, which has been treated in the same way as the PPO, has shown that it remains high ( $\geq 60\%$ ) between pH 4 and 8 and reaches its maximum at a pH of 6.4 (Figure 1 b). However, POD of Deglet Nour date was found to be less stable than Ghars one in the pH range of 4 to 6.4. On the other hand, it seems the most stable at pH 3.2 and in the pH range 7.2 - 8. Deglet Nour POD gave a maximum of residual activity at pH 7. The residual activity of POD of both varieties remains above 50% from pH 4 to more than pH 8. The loss of activity at pH extremes is probably due to the denaturation of enzymes by ionization.



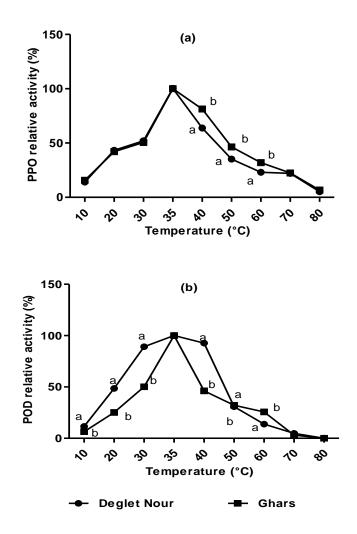


**Figure 1**. Effect of pH on stability of PPO (a) and POD (b) of Deglet Nour and Ghars dates (expressed as percentage of control activity) after incubation of enzymes preparations for 12 h at 10 °C with buffer solutions of various pH (pH = 2.4-8.6).

#### **Temperature effect**

The maximum residual activity of PPO and POD of both date varieties was obtained at 35 °C (Figure 2, a and b). Incubation of PPO and POD enzyme preparations at temperatures other than 35 °C resulted in a gradual deactivation of both enzymes, ranging from this temperature to extreme ones, so that the PPO retained only about 50% of its activity at 30 °C and 50 °C and retains only less than 20% of this activity at 70 °C and 10 °C. This is in agreement with the results of Dorantes-Alvarez and Chiralt (2000) and Tomas-Barberan and Espin (2001) who reported that a temperature above 80 °C is needed to ensure PPO inactivation in processed horticultural products, such as juices, canned fruits, vegetables, etc. On the other hand, the POD is completely deactivated at 80 °C; its activity is almost zero at 70 °C and 10 °C. The thermal stability of PODs varies according to the different sources. Indeed, it has been shown that some PODs are less thermostable than other enzymes within the same plant (Terefe et al., 2014).

This proves that the PPO of the two fruit has more thermostability than the PODs. This was also confirmed by Chisari et al. (2007) who finds that peroxidase from both cultivars of strawberry was much more thermolabile as compared with PPO, losing over 60% of relative activity already after 60 min of incubation at 40°C.



**Figure 2**. Effect of temperature on stability of PPO (a) and POD (b) of Deglet Nour and Ghars dates (expressed as percentage of control activity) after incubation of enzymes preparations for 1h at pH 7 and T = 10-80 °C.

#### pH and temperatures of maximal stability of PPO and POD

From this study pH and temperatures of maximal stability of PPO and POD of both date fruit varieties were detected and they are mentioned in table 1. Results of pH giving maximum of stability to both enzymes, PPO and POD, are different from those of Ciou et al. (2011) who found pH 5.0 for both PPO and POD of water caltrop pericarps, based on their reactions with catechol and guaiacol, respectively. Otherwise, it was found that the optimum pH value was around 7.0 for all PPO and POD tested for different substrates (Alici and Arabaci, 2016). In addition, the stability of the PPO and POD from water caltrop pericarp has been proven between 40 and 50  $^{\circ}$ C (Ciou et al., 2011).

Date variety	Enzyme	pH of maximal stability	temperature (°C) of maximal stability
Deglet Nour	РРО	7.2	35
	POD	7.2	35
Ghars	РРО	7.2	35
	POD	6.4	35

Table 1. pH and temperature of maximal stability of PPO and POD from Deglet Nour and Ghars

## Conclusion

From the results obtained in this study, it's clear that stability rates of PPO and POD from Deglet Nour and Ghars dates against different pH and temperatures remained comparable in both cases for each parameter and each variety. These activities were highly inhibited at extreme pH and temperatures, but remained significant over a wide range.

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## The Macrophyte Problem in Yedigöller (Erzurum) and The Solution Proposals

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#### Abstract

Today, our natural water resources are being polluted and even become unusable due to industrialization and rapid population increase. Erzurum is a province that has a high freshwater potential since it is located within the Aras, Çoruh, and Fırat basins. However, certain agricultural and husbandry activities in the region started to affect the quality of the water negatively. This study was conducted in order to reveal the nutritious level of Yedigöller Lake which is located in Erzurum province due to the excessive increase of macrophytes (aquatic plants) and to research the reason for the increase in the macrophyte. Certain water quality parameters were examined in the water samples that were taken from 5 stations determined in the lake. Water temperature, dissolved oxygen, pH, and electrical conductivity values were measured on-site. Total phosphorus (TP), ammonia nitrogen (NH<sub>3</sub>-N), nitrite nitrogen (NO<sub>2</sub>-N), and nitrate-nitrogen (NO<sub>3</sub>-N) values were examined.

Keywords: Macrophyte, Water quality, Yedigöller, Erzurum

#### Introduction

Erzurum and its surroundings are one of the main regions of Turkey in terms of water reserve considering the province as a whole within the Eastern Anatolia Region. It is a province with high freshwater potential since it is located in Aras, Çoruh, and Fırat basins and has a high altitude. The surface water potential of the province which consists of rivers, natural lakes, ponds, and reservoirs is 11.685 hm<sup>3</sup>/year, underground water potential is 433 hm<sup>3</sup>/year, and the total water potential is 12.118 hm<sup>3</sup>/year (Anonim 2018) (Table 1).

Table 1. Water Potential of Erzurum province	Table 1.	Water	Potential	of Erzurum	province
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River name	River's Length within the Provincial Borders (km)	River's Tributary
Çoruh River	104	-
Tortum River	70	Çoruh River
Karasu River	152	Fırat River
Aras River	212	-
Pond name	Pond volume (m <sup>3</sup> )	Irrigation area (ha)
Palandöken pond	1.558.000	340

Şenkaya pond	2.450.000	350
Ürünlü pond	5.620.000	1273
Köyceğiz pond	6.085.000	1350
Porsuk pond	76.500	170
Kapıkaya pond	1.040.000	230
Resource		Underground water volume
Erzurum Plain (Erzurum-Kümbet- Cinis Plains)		87.81
Hınıs-Karaçoban		69.00
Pasinler		82.50
Horasan		46.90
Tekman		113.02

The origin and the morphology of a lake is the most researched subject. Lake geomorphology is a fundamental issue that is examined in three stages as physical, chemical, and biological and it is interpreted along with the climate of the basin (Wetzel, 2001).

It is required to determine the quality of the resource in studies about the use of natural resources. Several physicochemical (such as N, P, Si, Ca, Mg) and biological parameters (such as phytoplankton, zooplankton, macrophyte) are used in the determination of the quality of water resources. Since the use of bioindicator demonstrates the effects of environmental changes on the group of biological assets, it is crucial in evaluating the trophic level and it is suggested to use both methods and to examine the results together (Cox, 1991).

This study examines the increase and control of macrophytes in Yedigöller.

## MATERIAL AND METHOD

The samples that were taken from 5 locations in Yedigöller Lake which are located within the border of Erzurum province, water temperature, dissolved oxygen, pH, and conductivity (with YSI Multiparameter) were measured on-site. The map and the stations of Yedigöller were given in Figure 1.



#### Figure 1. Yedigöller Lake and Stations

#### Total Phosphorus (TP), Orthophosphate phosphorus (PO<sub>4</sub>-P)

In the total phosphorus analysis, the persulfate digestion method was used in the first stage (digestion process), and then the released orthophosphate was determined with the ascorbic acid method according to Anonymous (1995).

#### Ammonia Nitrogen (NH<sub>3</sub>-N), Nitrite Nitrogen (NO<sub>2</sub>-N), and Nitrate Nitrogen (NO<sub>3</sub>-N)

The ammonia nitrogen was determined according to the Nesslerization. In this method, the color intensity based on the concentration of yellow color given by the Nessler reactive was measured as 410 nm wavelength in the spectrophotometer. Nitrite nitrogen was determined by measuring the color that emerges with the diazotization of sulphanilic acid and N-1- naphthyl ethylenediamine dihydrochloride in the spectrophotometer in 523 nm wavelength. The nitrate-nitrogen was calculated with measuring the yellow color that emerges as a result of the reaction between the nitrate ions and brucine sulfate in the water sample in 410 nm wavelength (Anonymous 1995).

#### FINDINGS

The following findings were obtained as a result of the examination which was conducted due to the excessive increase of macrophytes (aquatic plants) in Yedigöller Lake located within the borders of Uzundere country of Erzurum province (Table 2).

Parameters	Results	Limit Values (Water Quality)
Water Temperature (°C)	14	Grade I
pH	8.4	Grade I-II
Dissolved Oxygen (mg/L)	7.6	Grade II
Electrical conductivity (mS/cm)	0.343	Grade I
Total Hardness (mg/L CaCO <sub>3</sub> )	193	Hard waters
Ammonia Nitrogen	1.3	
Nitrite Nitrogen (NO <sub>2</sub> -N mg/L)	0.2	Grade IV
Nitrate Nitrogen (NO <sub>3</sub> -N mg/L)	0.003	Grade II
Orthophosphate Phosphorus (PO <sub>4</sub> -P mg/L)	0.002	
Total Phosphorus (TP mg/L)	1.52	Grade IV

Table 2. Results of Certain Water Quality Parameters of Yedigöler (June 19, 2019)

It was determined that the TP and NO<sub>2</sub>-N values in the lake water ranked as **bad** water according to the intra-continental water quality standards. The lake is classified as hard water. The lake includes *Spirogyra, Phragmites, Klebsormidiales* algae. Underwater and surface algae growth was observed intensely (Figure 2).

The results that were obtained from the samples taken from Yedigöller on 09/07/2019 were given in Table 3. According to the analysis results, although the ammonia nitrogen values were high in the resources of the lake, the form may change before it reaches to  $3^{rd}$ ,  $4^{th}$ , and  $5^{th}$  stations. However, it is important to eliminate the nitrogen resources in the  $1^{st}$  and  $2^{nd}$  stations since this stations are directly given to the stations and the high value of nitrite nitrogen in the lake. Additionally, total phosphorus value was determined quite high in these stations. This is considered to happen due to the access of domestic wastes to the lake from the same station.

Table 3. Results of Certain Water Quality Parameters of Yedigöller (July 09, 2019).

Parameters	1 <sup>st</sup>	2 <sup>nd</sup> Station	3 <sup>rd</sup>	4 <sup>th</sup>	5 <sup>th</sup>	Limit Values for
	Station		Station	Station	Station	Natural Lakes
Total Hardness	145	142	170	159	170	75-150 (medium
(mg/L CaCO <sub>3</sub> )						hard)
						150-300 (hard)
Ammonia nitrogen	0.91	0.91	0.90	0.91	0.98	0.02
(NH <sub>3</sub> -N mg/L)						

Nitrite Nitrogen	0.08	0.08	0	0	0	0.002-0.01
(NO <sub>2</sub> -N mg/L)						
Total Phosphorus	0.57	0.55	0.36	0.36	0.39	0.3>TP>1
(TP mg/L)						(eutrophic)



Figure 2. Overview of The Lake and Sample Station Points

## DISCUSSION AND CONCLUSION

It was determined in both studies conducted in the lake that the main reason for the intense growth of macrophyte (aquatic plants) in the lake is the high nitrogen and phosphorus values. For this reason, it is suggested to;

1. Domestic wastes should not be directly mixed with the lake,

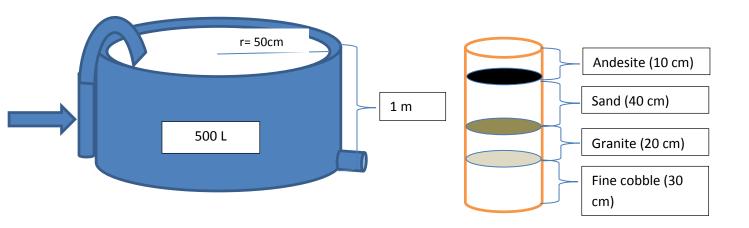
2. Garbage containers located around the water resources should be at least 50 m away from the resource and the garbage should be collected carefully,

3. The water from the fish production farm should not be directly mixed with the lake,

4. Required precautions should be taken on preventing solid wastes to be thrown to the lake such as plastics in order to eliminate the resources that come to the lake.

#### Suggestions:

1. The nitrite nitrogen and total phosphorus values of the lake water were determined high. This originates from the mixing of intense organic pollution to the lake. It is of importance to filter the wastewater that comes from fish production facilities and houses with sand filter system before mixing the wastewater with the lake (Figure 3).



#### Figure 3. Sand Filter System

**2.** The ammonia nitrogen value was also found high in the lake. For this reason, establishing ventilation for the lake is important in terms of increasing the dissolved oxygen value in the water (Figure 4). Because while water plants produce  $O_2$  (oxygen) in the daytime, they start to produce  $CO_2$  (carbon dioxide) at nights and this situation negatively affects the living creatures in the water.



Figure 4. Lake Water Ventilation Systems

**3.** It was observed that there was an intense growth of underwater plants. Thus, it is required to harvest these plants. There are companies that rent and sell **Wetland Amphibious Excavator Vehicles** (Figure 5). These vehicles are used by many municipalities in our country. Another procedure that should be performed is the removal of sediment. It is suggested to remove the detritus that accumulates in the lake.



Figure 5. Wetland Amphibious Excavator Vehicle and Bucket

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## A potential pest of rhubarb (*Rheum ribes* L.) (Polygonaceae) in Turkey: *Capnodis marquardti* Reitter, 1913 (Coleoptera: Buprestidae)

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#### Abstract

The rhubarb (*Rheum ribes* L.) (Polygonaceae) is element of Iran-Turanian phytogeographic region. It is distributed Turkey, Azerbaijan, Armenia, Syria, Israel, Jordan, Lebanon, Northern Iraq, Iran, Afghanistan and Pakistan. In Turkey, it is quite abundant in the Eastern and the Southern Anatolia. This plant occures on the altitude between 1800 and 2800 m on rocky and gravel slopes, and can grow up to 40 cm of height. The rhubarb is a perennial herbaceous plant blooming with yellowish-white flowers during May and June. In addition to medical use its shoots and petioles are consumed fresh and cooked for different recipes in Turkey. Buprestid beetle Capnodis marquardti Reitter, 1913 (Coleoptera: Buprestidae: Dicercini) known from northeastern Turkey exclusively, was found to be associated with this plant. There is very limited information about the interrelations between this species and rhubarb. Large population of C. marquardti was found in Cirisli Pass between Erzurum and Bingol, 13th km N of Karliova. The adults were observed on the soil close to the stems of rhubarb plants; while a single larva was found in the roots of this plant. The adult feeding and oviposition were observed on caged plants in the field. Further observations of the biology of C. marguardti and its impact on the plant are necessary to study. In any case, known this exclusively rare and presumably endemic species for northeastern Turkey by people working with this plant has great importance.

Keywords: Capnodis marquardti, Rheum ribes, biological observations, Turkey

## Introduction

Rhubarb (*Rheum ribes* L.) is a perennial, herbaceous, and wild plant from Polygonaceae family and it is the only species that grow from *Rheum* species in our country. This species which is known as işgın or işgın weed in Turkish and it is called different names such as ışkın, ıçgın, ıçkın, uçkun, uçgun, uşgun, uşkun, aşgın, aşkın, eşkin, eşgin in public language (Baytop, 1997). Rhubarb is a element of phytogeographical region in Iran-Turan (Anonymous, 2019a). *R. ribes* distributed in Turkey, Syria, Israel, Palestine, Jordan, Lebanon (Anonymous, 2019b; 2019c), Armenia, Northern Iraq, Iran, Afghanistan, Pakistan (Anonymous 2019d) and Azerbaijan (Seyidahmedov et al., 2008). The most densely grow region of the rhubarb are reported as Kars, Erzincan, Bingöl, Van, Ağrı, Hakkari, Elazığ, Erzurum (Aziziye), between Erzurum and Bayburt (Kop Mountain Pass), between Erzurum and Bingöl (Karlıova, Cirisli Pass), Muş, Malatya (Anonymous, 2019e), Bitlis, Tunceli, Iğdır (Anonymous, 2006a) (Fig. 1).



#### Fig. 1. Distribution of *Rheum ribes* in Turkey

It was observed that *R. ribes* prefer medium or heavy textured soil and also, it can be grown in clay, acidic or alkaline soil. It has resistance against extremely cold weather and can survive up to minus 20 °C (Öztürk et al., 2007). The plant which can be seen in heights of 1800-2800 m grows in rocky and pebble slopes and can grow up to 40-150 cm (Andıç et al., 2009).

It is a perennial, hard, taproot, and herbaceous plant which blooms yellowish-white in May and June. The stem is foliated. The large leaves that are not completely lobed resemble a kidney or elephant ear with reddish, ramentaceous, grooved features. The surface of the leaves and petiole is rough and hard (Tanker, 2004).

It is stated that the sucker of the plant is foliated on the base and aphyllous on the top and these suckers and petioles are consumed freshly and different meals are prepared by cooking (Munzuroğlu et al., 2000). The fresh suckers of the plant are rich in Vitamin C and poor in Vitamin A and E, furthermore, it was reported that the level of selenium is sufficient in terms of nutrition (Munzuroğlu et al., 2000). In the study of Andiç et al. (2009) in which the content of chemical compounds was examined for *R. ribes* plant that was collected from Ağrı, Bitlis, Erzurum, Hakkari, Muş, and Van provinces, it was determined that the average total solids were 5.589%, ash was 0.636%, nitrogen was 0.206%, crude protein was 1.292%, crude fiber was 1.798%, soluble solid content was 3.897%, total sugar was 1.760%, inverted sugar was 1.643%, saccharose was 0.112%, acidity was 0.967%, pH was 3.566, vitamin C was 5.196 mg/100g, Fe was 3.752 µg/g, Zn was 1.132 µg/g, P was 411.2, Pb was 0.016 µg/g, Cu was 0.501 µg/g, and Mn was 0.423 µg/g.

It is reported that *R. ribes* are a raw material source for the most important drugs in the Middle-East (Kashiwada et al., 1988). It is also reported that the leaves and stem of the plant are sour, strengthen the stomach, and prevent vomiting (Munzuroğlu et al., 2000). It is stated that the roots of *Rheum* are used as a laxative and antipsoriatic drug (Shokravi and Agha Nasiri, 1997). It is also recorded that beige (Öztürk and Özçelik, 1991), blue, mustard colour (Özgökçe and Yılmaz, 2003), brick red, dark green, maroon and canary yellow colors (Gönen, 2008) were obtained from the roots of rhubarb. Furthermore, *R. ribes* plant which has commercial value is

picked by the locals where the plant is grown in Turkey, sold freshly and a significant income is obtained (Fig. 2). In addition to all of this information, warnings are given by various researchers about not using the plant randomly, not destroying its population insensibly (rooting out in particular) and the fact that the plant might be harmful to human health (Anonymous, 2014; 2017).



Fig. 2. Trade of fresh *Rheum ribes* (in Erzurum, Original)

There are several insect species that harm the plant by feeding on it which has commercial and nutritious value for human health. The aim of this study is to reveal the existence of *Capnodis marquardti* Reitter (Coleoptera: Buprestidae) damages to *R. ribes* and endemic for Turkey, its eggs and its larvae for the first time in Turkey.

## **Material and Method**

The material of the study consists *Capnodis marquardti* adults that were caught around the roots and leaves of *Rheum ribes* plant in the rocky and pebble slopes between the heights of 1940 and 2200 m and between Erzurum-Bingöl provinces (13 km North of Karliova) (Fig. 3), its larvae obtained from the root of the plant, and its eggs laid by the female insects as a result of the mating of adults that were taken to culture.

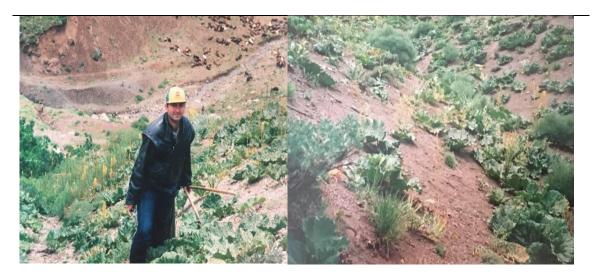


Fig. 3. A view from the area (Cirisli Pass between Erzurum and Bingol) where the study was conducted

## **Results and Discussion**

The length of the *Capnodis* species is between 10 to 40 mm. Their body is solid and hard. The color is black with bronze shining or cupreous. The insect has big eyes and slightly approximate in the vertex. The sense pores at the tip of the antenna spread through the slight holes of which the borders are uncertain. The pronotum expands and has smooth black areas which are aligned straight and characteristic (Fig. 4 b). There is a space shaped like reverse (V) letter in the middle of the pronotum posterior. The pits on the elytra create deep lines in certain species (Tozlu, 1997). It is reported that 22 species and sub-species of this insect in the world (Löbl and Smetana, 2006) and 10 of this species exist in Turkey (Lodos and Tezcan, 1995).

The larvae of this genus are called as "fidan dipkurtları" in Turkish. Most of the species damage fruit (such as almond, peach, apricot, plum, cherry, sour cherry, pistachio, and hackberry) and forest trees (such as poplar and willow) and the roots of bushy plants.

Capnodis marquardti Reitter, 1913 türünün sistematikteki yeri:

Order: Coleoptera Superfamily: Buprestoidea Leach, 1815 Family: Buprestidae Leach, 1815 Subfamily: Chrysochroinae, Laporte de Castelnau, 1835 Tribe: Dicercini, Gistel, 1848 Genus: *Capnodis* Eschscholtz, 1829

*C. marquardti* samples (16 units) were found for the first time on June 16, 2001, in Cirisli Pass (between Erzurum and Bingöl, 13 km north of Karliova) in 1940 meters (Fig. 4a,c). On the same date, adult insects were found between the heights of 2000 to 2200 meters. On July 8, 2005, adult insects (22 units) were determined in 2100 meters. The larvae was obtained from

inside the plant roots uprooted (in 10 October 2005) (Fig. 4i,j,k,l,m,n,o). These adults and a plant that was rooted out were brought to the Department of Plant Protection, Faculty of Agriculture at Atatürk University for cultivation. This plant was transferred to a plastic box with soil within the landscaping of the Department of Plant Protection and the adult insects were released on the plant (Fig. 4d,f). Although the plant was not preserved for a long time, it was provided for adult females to lay eggs (Fig. 4g,h,1). Another remarkable condition in the area is that the adult insects were found on the soil near the root parts of plants (Fig. 4e) and on the leaves of *R. ribes* plant between 12:00-14:00 which is the hottest hours of the day and brightest hours of the sun. Moreover, *C. marquardti* was not determined prior to this time of the day although the area was studied. This situation verifies that the Buprestidae species are active in the hottest hours of the day and the period when the sun is brightest. The determinations about the egg and larvae of the species and the images shown here are presented for the first time in this study in the world and Turkey.

*C. marquardti* was first identified in Van province in the world and it is endemic to Turkey. Koçak and Kemal (2009) recorded about this species that "Van Prov., Erekdağı 2150 m,.19.05.2009; inhabits at stony slopes, where *Rheum ribes* grows" and they also stated that this species was found in Van and Bingöl (Koçak and Kemal, 2010). Koratyaev et al. (2016) was stated that "*Rheum ribes* forms dense population in the location (Bingöl–Erzurum provinces border, 13 km N of Karlıova, Cirisli Pass, 2000-2300 m) on the deforested and eroded mountain. A jewel beetle *C. marquardti* is specialized coleopterous herbivores associated with *R. ribes* in this location, all of them common on the plants" in their study entitled "Bioindicator beetles and plants in desertified and eroded lands in Turkey". They also reported that they encountered this species on 07.03.2005 and the plant was a new record as a host. Although it was determined by us in 2001 and were made certain biological observations in 2005, the study can be published this year. Due to the terrorist threat in the region, detailed data cannot be obtained. Even though it is late, this study was put forward since it was considered to present this information.







Fig. 4. The views of this study (field and laboratory) on Capnodis marquardti

As is seen, there are only a few studies about the determination of this insect. This study is significant in terms of providing information about the species for those who are studying or will study the insect. We need to know the existence of *C. maquardti* which is endemic and damaging to *R. ribes* an important plant for many different areas such as medical and human nutritient. It is necessary to consider this for the plans that will be made about this plant in future studies.

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# Role of Exogenous Melatonin on Seedling Growth and Some Physiological Properties of Bean under Salt Stress

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## Abstract

In this study, the effects of melatonin application on bean seedlings were investigated under salt stress. Three salt (0, 75 and 150 mM NaCl) and four doses of melatonin (0, 25, 50 and 100  $\mu$ M) were applied to plant in the study. The seeds kept in melatonin solutions were sown into pots, and after the seedling emergence, irrigation was done with different doses of saline water. Melatonin treatments were also sprayed to the leaves three times at intervals of one week during plant development. In this study, stem diameter, plant fresh and dry weight, root fresh and dry weight, chlorophyll reading value (SPAD), electrical conductivity (EC), and leaf relative water content (LRWC) were measured. As a result of the research, stem diameter, plant fresh and dry weight, root fresh and dry weight, SPAD and LWRC decreased but EC increased in bean seedlings with increasing salinity. There were different effects between melatonin doses. The effects of melatonin application on bean seedlings under salt stress were statistically significant. It was determined that the damage caused by the salt stress in the plant was decreased with melatonin applications.

Keywords: Melatonin, salinity, plant growth

## Introduction

Melatonin (N-acetyl-5-methoxy-tryptamine) is a molecule commonly found in animals and plants and has many physiological functions in their metabolisms. Melatonin plays a role in important events such as seed germination, plant growth and reproduction, fruit formation and protects plants against many abiotic and biotic stress factors [1, 2, 3, 4]. It was determined that melatonin has similar biosynthesis with auxin in plants and acts as hormone [3]. Melatonin cleans reactive oxygen species and regulates gene expression by increasing antioxidant enzyme activity, photosynthetic compounds and metabolic compounds to provide tolerance to stress in plants [3, 4].

Melatonin could be tolerated against abiotic stresses such as chilling stress in cucumber [5, 6], drought stress in tomato [7] and maize [8], cold stress in carrot [9] and tomato [10], high-temperature stress in radish [11] and maize [12], metal stress in red cabbage [13]. Also, the mitigating effect of melatonin to salt stress in the plant was determined by the researchers. The researchers determined that salt stress damage in various plants could be alleviated by exogenous melatonin applications [2, 14, 15, 16, 17].

Especially in arid and semi-arid regions, salinity is one of the major problems causing agricultural problems. Salinity causes significant yield and production losses in agricultural products. Increased salinity causes ion toxicity and osmotic stress [18, 19]. Also, salt stress cause to reactive oxygen species (ROS), peroxidation of membrane lipids or proteins and

destroy the structure of cell membranes, leading to oxidative damage and cell death to the plant [18, 20].

Bean is one of the most widely cultivated agricultural crops in the world. Bean is sensitive to salt and it is stated that yield losses can occur even at soil salinity lower than 2 dSm<sup>-1</sup> [21, 22]. In the bean growing areas, irrigation is insufficient and partly salty soils cause salinity problems in bean cultivation. There may be differences between bean genotypes in terms of salinity tolerance [23]. However, exogenous applications may be made to alleviate the damage to salt stress in the salt-sensitive genotypes. Although the effects of melatonin applications against various stress factors have been investigated in many plants, there are not enough studies on the effect of melatonin on bean plants. Therefore, in this study, the mitigating effect of melatonin application on the negative effect of salt stress on bean seedling was investigated.

# Material and Method

This study was conducted in controlled greenhouse condition and bean seed (*Phaseolus vulgaris* L.) was used as plant materials. In the study where 3 different salt doses (0, 75 and 150 mM NaCl) were used as salt stress, 4 different doses of melatonin (0, 25, 50 and 100  $\mu$ M) were applied to the plants. Seeds were sown into mixture of soil: sand: peat (2:1:1) after keeping in melatonin solutions. Irrigation was done with different doses of saline water after the seedling emergence. Melatonin treatments were sprayed to the leaves three times at intervals of one week during plant development. The salt stress applications were made as irrigation water. The salt application was initially started with 25 mM dose and gradually increased and fixed at the actual dose. The pot experiment was made according to randomized block designs with three replications and four plants in the each replicate.

In this study, stem diameter, plant fresh and dry weight, root fresh and dry weight, chlorophyll reading value (SPAD), electrical conductivity (EC) and leaf relative water content (LRWC) were measured 50 days after seed sowing.

For statistically analysis of data SPSS program was used [24]. Data were subjected to analysis of variance to compare the effects of salt levels, melatonin doses and their interactions. The differences among the means were compared using the Duncan multiple range tests (DMRT).

## **Results and Discussion**

In this study, effects of melatonin application on bean seedlings grown under salt stress were determined. Statistically significant effects of melatonin application and salinity were determined separately. Significant results were obtained on seedling growth and some physiological properties of bean with melatonin applications under saline condition (Table 1 and 2, Figure 1-8).

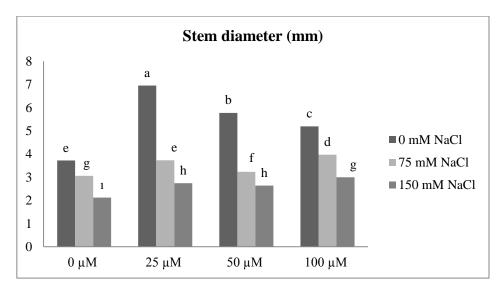
Stem diameter, SPAD and LWRC increased with melatonin applications, but increased salinity significantly reduced this parameters. But EC not changes significant with melatonin application, while EC increased with salinity (Table 1).

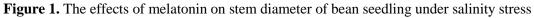
Application	Dose	Stem diameter (mm)	Chlorophyll reading value (SPAD)	EC (%)	LWRC (%)
	0	2,97 d	31,22 b	57,25 ns	65,26 c
	25 μΜ	4,74 a	30,67 b	55,61	70,06 b
Melatonin	50 µM	3,88 c	34,11 a	56,32	71,00 ab
	100 µM	4,05 b	31,44 b	56,63	72,04 a
	0	5,41 a	42,75 a	31,29 c	76,24 a
Salt	75 mM	3,50 b	28,17 b	64,86 b	69,17 b
	150 mM	2,63 c	24,67 c	73,21 a	63,37 c

**Table 1.** The effects of melatonin and salt stress on stem diameter, SPAD, EC and LWRC of bean seedling

\*: Data followed by a different letter in column were significantly different according to the DMRT (p<0,001)

In salt-free conditions, the highest stem diameter was obtained by application of 25  $\mu$ M (6,95 mm) melatonin, while the lowest stem diameter was obtained by the control plant (3,72 mm). 50  $\mu$ M and 100  $\mu$ M applications also increased the stem diameter under salt-free conditions. The stem diameter showed a significant increase with the application of 100  $\mu$ M melatonin under 75 mM (3,97 mm) and 150 mM (3,00 mm) salt stress conditions. Furthermore, the applications of 25  $\mu$ M and 50  $\mu$ M melatonin, respectively, were greater in stem diameter than control (Figure 1).





\*: Data followed by a different letter on bars were significantly different according to the DMRT (p<0,001)

In the absence of salt stress, the SPAD value did not change significantly with melatonin applications, whereas the decrease in salinity with SPAD was higher in 50  $\mu$ M melatonin

application with both saline (29,67 in 75 mM NaCl and 28,67 in 150 mM NaCl) levels compared to control and other applications (Figure 2).

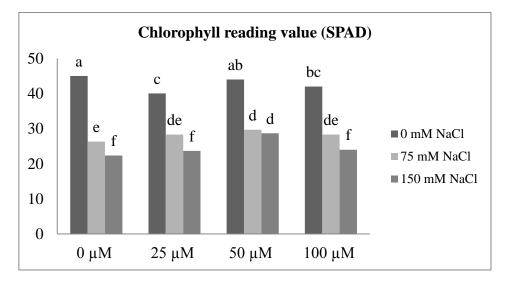


Figure 2. The effects of melatonin on chlorophyll reading value of bean seedling under salinity stress

\*: Data followed by a different letter on bars were significantly different according to the DMRT (p<0,001)

The EC ratio increased with increasing salinity, EC was the highest in 100  $\mu$ M melatonin (74,26%) application at the highest (150 mM NaCl) salt level. However, 25  $\mu$ M melatonin (71,27%) reduced the EC content compared to other applications at the 150 mM salt level (Figure 3).

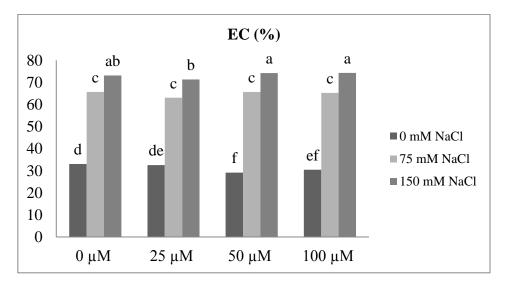


Figure 3. The effects of melatonin on EC of bean seedling under salinity stress

\*: Data followed by a different letter on bars were significantly different according to the DMRT (p<0,001)

LWRC decreased with salt stress and LWRC decreased from 73,78% in control to 57,45% in 150 mM salt stress. The highest LWRC was obtained with 50  $\mu$ M melatonin application (79,84%) in conditions without salt stress, whereas the 100  $\mu$ M melatonin application was

higher than the LWRC at 75 (72,68%) and 150 mM (67,41%) salt level compared to the control and other applications (Figure 4).

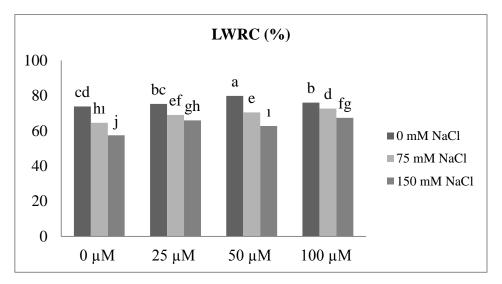


Figure 4. The effects of melatonin on LWRC of bean seedling under salinity stress

\*: Data followed by a different letter on bars were significantly different according to the DMRT (p<0,001)

In parallel with increasing salt level, plant fresh and dry weight and root fresh and dry weight decreased significantly. According to the averages, while the plant fresh and dry weight, root fresh and dry weight were 32,00 g, 4,93 g, 24,25 g and 1,25 g in the control (0 mM NaCl), respectively, they were 18,04 g, 2,87 g, 13,98 g and 0,61 g (respectively) in the 150 mM NaCl. Plant fresh (27,73 g) and dry weight (4,32 g), root fresh (24,80 g) and dry weight (1,08) in 100  $\mu$ M melatonin application was higher than the other applications (Table 2).

**Table 2.** The effects of melatonin and salt stress on plant fresh and dry weight, root fresh and dry weight of bean seedling

Application	Dose	Plant fresh weight (g)	Plant dry weight (g)	Root fresh weight (g)	Root dry weight (g)
	0	24,13 b	3,81 b	16,67 c	0,82 b
	25 μΜ	23,94 b	3,71 b	17,74 b	0,79 b
Melatonin	50 µM	24,53 b	3,76 b	16,30 c	0,81 b
	100 μΜ	27,73 a	4,32 a	24,80 a	1,08 a
	0	32,00 a	4,93 a	24,25 a	1,25 a
Salt	75 mM	25,22 b	3,91 b	18,41 b	0,77 b
	150 mM	18,04 c	2,87 c	13,98 c	0,61 c

\*: Data followed by a different letter in column were significantly different according to the DMRT (p<0,001)

The plant fresh weight decreased with salt stress, however, with melatonin applications, especially 100  $\mu$ M application gave the highest plant fresh weight compared to control and

other applications under both stress-free (0 mM) (34,90 g) conditions and 75 mM (28,12 g) and 150 mM (20,18 g) salt stress conditions (Figure 5).

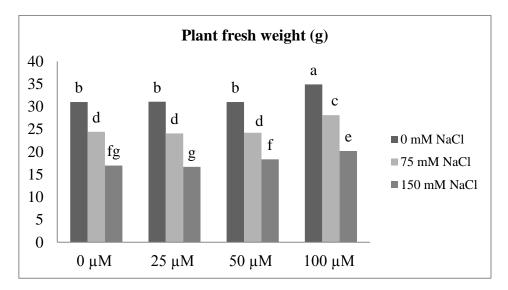


Figure 5. The effects of melatonin on plant fresh weight of bean seedling under salinity stress

\*: Data followed by a different letter on bars were significantly different according to the DMRT (p<0,001)

There was no statistically significant difference between applications in terms of plant dry weight in conditions without salt stress (0 mM). However, the dry weight of the plant was higher with 100  $\mu$ M melatonin application than other applications at 75 (4,53 g) and 150 mM (3,45 g) salt conditions (Figure 6).

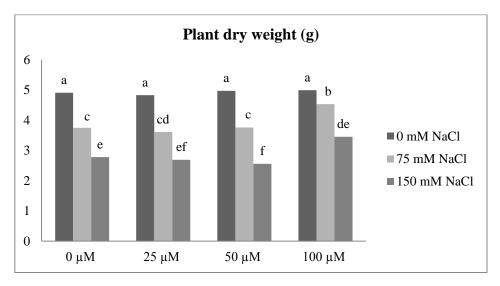


Figure 6. The effects of melatonin on plant dry weight of bean seedling under salinity stress

\*: Data followed by a different letter on bars were significantly different according to the DMRT (p<0,001)

The root fresh weight decreased with salt stress, however, with melatonin applications, especially  $100 \ \mu$ M application gave the highest root fresh weight compared to control and other

applications under 0 mM (30,53 g), 75 mM (23,43 g) and 150 mM (20,44 g) salt stress conditions (Figure 7).

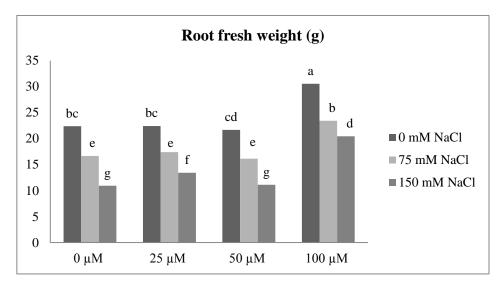
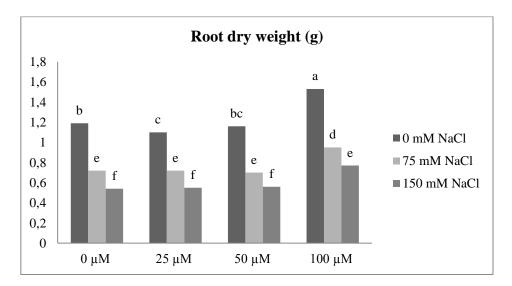
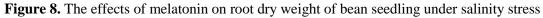


Figure 7. The effects of melatonin on root fresh weight of bean seedling under salinity stress

\*: Data followed by a different letter on bars were significantly different according to the DMRT (p<0,001)

In this study, the root dry weight decreased with salt stress, however, with melatonin applications, especially 100  $\mu$ M application gave the highest root dry weight compared to control and other applications under both 0 mM (1,53 g) conditions and 75 mM (0,95 g) and 150 mM (0,77 g) salt stress conditions (Figure 8).





\*: Data followed by a different letter on bars were significantly different according to the DMRT (p<0,001)

In the study, it was determined that salt stress negatively affected the development of bean seedling. Similarly, Dantas et al. [25] reported that salinity caused to decrease on seed germination and seedling growth of bean. Furthermore, chlorophyll content, stomatal

conductance and photosynthetic activity of bean were decreased by salt stress [26]. However, it was determined that the damage caused by salt stress in the plant was less with melatonin application in our study. Similarly, in a study conducted on tomato, exogenous melatonin application by affecting the expression of some antioxidant related genes and related enzymes reduced the ROS accumulation, and plants under the stress conditions had better growth [4].

In the present study, especially 100  $\mu$ M doses of melatonin had a positive effect on salt stress. Similarly, Xie et al. [17] found that in salt stress, an increase in chlorophyll content, carotenoid content, photosynthesis rate, stomatal conductance and transpiration rate occurred in parallel with increasing doses of melatonin applied exogenous in lettuce seedlings, however, 100  $\mu$ M melatonin application provided the best effect on salt stress tolerance in the plant. It was determined that 50-150  $\mu$ M melatonin applications increased photosynthetic activity and antioxidant enzyme activity, oxidative damage decreased and increased plant growth against salt stress in cucumber [2]. It can be said that the reason for increasing tolerance to salt stress in melatonin treated bean seedlings is to reduce reactive oxygen species by increase antioxidant enzyme activity, regulating photosynthetic properties, metabolite compounds and transcription factors [27]. As a matter of fact, SOD, POD, CAT and APX enzyme activities were increased with melatonin in melon seedlings grown in saline conditions, electrolyte leakage, MDA and oxidative damage decreased [28]. So, in this study was determined that the harmful effect of salinity on growth of melatonin treated bean seedlings of different researchers.

## Conclusion

In the study, salt stress negatively affected the growth of bean. However, the negative effect of salt stress on beans could be alleviated by the application of exogenous melatonin. Melatonin applications in different doses showed positive effects in both normal and saline conditions. In particular, the negative effect of salt stress on bean seedlings with 100  $\mu$ M melatonin application was found to be less.

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# Functions of Phytohormones on Plants under Drought and Salt Stress

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## Abstract

Phytohormones are molecules in low concentrations and have important functions in plant metabolism. Hormones such as auxin, cytokinin, gibberallin and abscisic acid are known to have important functions in plants. They are involved in morphological and physiological metabolism such as seed germination, seedling development, leaf and fruit formation, ripening and senescence in plants. In addition, phytohormones can also play a role in the resistance of plants to stress conditions. Plants can face many stress factors during their life. Abiotic stress factors caused by drought and salinity cause significant yield and quality losses in agricultural production. Plants react differently to the damage caused by abiotic stress factors in plant metabolism. One of these is the change in endogenous hormones. Recent studies have investigated the change of phytohormones and their effects on plants under stress conditions. In this review, the changes in phytohormones and their effects in plants under drought and salt stress conditions have been given with the studies of different researchers in recent years.

Keywords: hormone, salt, drought, stress, plant

## Introduction

Phytohormones are molecules that are produced in very low concentrations and regulate different cellular processes in plants [1]. Phytohormones have an effect that coordinates various signal transduction pathways as a response to abiotic stress factors [1, 2]. Some of the phytohormones are known as stress hormones. One of them is abscisic acid (ABA). ABA is an inhibitory hormone that is effective in maintaining seed dormancy, preventing germination, stoma closure, fruit casting etc. in plants. Researchers also stated that it is effective in abiotic stress [1, 3]. Phytohormones are considered to be important endogenous substances in regulating physiological and molecular responses in plants [1, 4]. Phytohormones, which have important effects on plants are auxin, cytokinin, gibberellin, salicylic acid, abscisic acid, ethylene, brassinosteroid and jasmonate [1, 5].

ABA is known as an important messenger in response to abiotic stress in plants, and its role in stress tolerance is considered important. An ABA level increase in plant metabolism in response to abiotic stress and is effective in activating specific signaling pathways and regulating gene expression level [1, 6, 7, 8]. Endogenous ABA level and biosynthetic gene expression is reported to increase in plants exposed to stress factors such as drought and salinity [9]. It is stated that ABA is effective in physiological processes such as stomatal opening and protein storage and it is an isoprenoid phytohormone that provides plant adaptation and tolerance against many stress factors [8, 10, 11, 12, 13, 14].

Auxin as plant growth hormone provides important effects in many stages of plant growth and development (such as seed germination, fruit ripening, tropisms, apical dominance) [15]. High levels of auxin in plants are associated with plant growth. The mechanism of adaptation to stress

in plants has an effect of auxin metabolic regulation [6, 16]. Indoleacetic acid (IAA), which is an auxin, is one of the most functional phytohormones, as it is effective in plant growth and development under normal and stress conditions [1, 17]. It was stated that the auxin level in plant decreased under stress conditions and it provided stress tolerance by increasing the plant growth with exogenous applications [6, 16, 18]. Du et al. [18] reported that the IAA content decreased under drought stress conditions but increased under cold and heat stress conditions. The researchers stated that many genes are involved in the IAA biosynthesis under stress conditions and show changes.

Cytokinins (CKs) have important functions in plant growth and development. CKs have an effect on functions such as cell division and maintenance of stem cells of apical meristems in shoots and roots, inhibiting of lateral root onset and leaf senescence, chloroplast biogenesis, anthocyanin production, nutrient mobilization etc. in plants [19, 20]. Endogenous CK levels change in response to abiotic stress in plants. CKs have antagonistic effects against ABA in plants. It is stated that there is a decrease in endogen cytokine level in plant under stress conditions and response to adverse effects of stress. However, it provides stress tolerance with exogenous applications to the plant [1, 20, 21].

Ethylene is another hormone that is effective on the growth of plant growth such as fruit ripening, flower and leaf formation and senescence of plant. In addition, it has a regulating effect against stress in plants [22, 23].

Gibberellic acids (GA) are effective in the formation of many vital functions such as seed germination, leaf enlargement, stem growth, flower and fruit formation [14, 24]. As one of the growth-promoting hormones, GA also has important effects on the adaptation and response of plants to stress conditions [25].

Brassinosteroids (BRs) are naturally occurring plant steroidal compounds and cause increase crop yields by altering plant metabolism, regulate plant development and physiology and protect plants to abiotic stresses [26, 27]. It is known that BRs is a phytohormone and have increased resistance on plant to abiotic stress such as drought and salt [26, 28].

Salicylic acid (SA) is a phenol compound found in many plant species. SA is effect on growth, flowering, thermogenesis, ion uptake, ethylene biosynthesis, stoma movement, chlorophyll and carotenoid pigments formation, photosynthesis etc. in plant [29, 30]. SA also acts as an important signaling molecule in the reaction of plants to stress [31]. It is stated that SA is involved in many abiotic stress reactions in plants, it has the potential to increase stress tolerance in agricultural products, however application dose, plant species and application time are important for stress tolerance of plants [32].

Jasmonic acid (JA) is a regulatory hormone that is effective in many vital activities of the plant such as seed germination, root growth, flowering, fertilization and senescence [18]. In a study where plant endogenous JA content increased with drought and cold stress and decreased with heat stress, it was determined that many genes involved in JA biosynthesis and signaling were stimulated by drought and cold stress but suppressed by heat stress [18]. It is known that JA increases in abiotic stress conditions such as drought and salinity, but its role in stress conditions is not fully established [18, 33, 34, 35].

## **Drought stress**

Drought stress is one of the important abiotic stress factors that negatively affect yield and quality in agricultural production. Drought stress, which changes the biochemical, molecular and physiological functions of the plant, causes a decrease in respiration and photosynthetic activity, causes the disruption of stomatal conductance, transpiration, the cell membrane structure and the disruption of the plant-water relationship, causing the degradation of plant morphological structure and even plant deaths [36, 37, 38, 39].

It has been reported by many researchers that ABA is the most effective hormone in the plant under drought stress. ABA can show an increase in the plant metabolism under stress conditions and also it has important functions in providing tolerance to drought stress in plants by exogenous ABA applications. Researchers suggest that the concentration of cellular ABA increases with water restriction and causes activation of many stress-responsible genes [39]. It was reported that ABA accumulate during drought stress and ABA levels control many reaction that assign plant growth, development, and survival during drought stress [40].

Phytohormones can reduce oxidative damage and alleviate stress damage in plants under drought stress especially due to their effects on enzyme activity. In a study conducted in *Coffea canephora*, it was suggested that the concentration of ABA in the roots and stems of drought tolerant clones increased, that ABA delayed the onset of severe water deficit and reduced oxidative damage [41]. In another study, it was reported that exogenous ABA application increased tolerance in wheat seedlings under drought conditions made with polyethylene glycol (PEG), increased shoot length, shoot and root dry weight, decreased hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) and malondialdehyde (MDA) content [42]. Chen et al., [43] explained that ABA application to *Tall fescue* plant decreased relative conductivity and MDA, leaf relative water content, antioxidant enzyme activity and proline content increased. In addition, ABA application reduced stomatal conductance, transpiration and photosynthesis.

The effect of ABA on enzymes was also determined by other phytohormones. Thus, in the study of phytohormone application to marigold in drought stress, it was determined that drought condition increased superoxide dismutase (SOD) and catalase (CAT) enzyme activities of plant, GA and BAP (benzyl amine purine) sprayed on leaves alleviated the effect of drought and the applications of ABA, JA, SA and BR stimulated activation of enzymes [44].

Hormones can create tolerance to stress with their effects on plant photosynthesis metabolism and transpiration mechanism in drought conditions. ABA is regulator of water use and regulates stomatal opening and transpiration. Stoma movement in plants reacts to environmental signals and affects hormonal content. ABA is a hormone that is effective in closing stomatal, and CK, JA, BR and ethylene also play a role in the reaction to stress. In drought, ABA was reported to interact with JA and nitric oxide to induce closure of stomata [45].

Mega et al. [46] have found that ABA receptor expression increases ABA sensitivity, which significantly reduces water consumption throughout plant life. Researchers have suggested that this water-saving property maintains efficiency during water constraint by reducing transpiration and increasing photosynthetic activity.

Nir et al. [47] suggested that in drought conditions, transpiration would be reduced with reduced GA activity in the plant, thus leaf drying would be less, and plant could maintain higher capacity.

Drought tolerance may vary according to plant species and genotypes. The tolerance mechanism of the hormones in plants under drought stress can first be seen on the plant growth parameters. Example, in drought tolerant and drought sensitive wheat genotypes, ABA and ethylene concentration increased and ABA/ethylene ratio decreased in mild drought. It was found that this situation decreased shoot dry weight in sensitive group and increased in tolerant group. In addition, in the same study, it was determined that foliar ABA application at low concentration increased the shoot dry weight in the drought sensitive group [48].

It was determined that auxin both in plant endogenous metabolism and with exogenous application affected drought tolerance positively in the plant, auxin responded to drought stress by affecting Reactive oxygen species (ROS) metabolism, regulated stress-related genes, hemeostasis and root structure [16].

Nazar et al. [49] stated that foliar SA application to mustard plant increased photosynthesis and plant growth, and so improved drought tolerance. It has also been reported that SA increases proline production and thus alleviates the damage of drought stress. The researchers found that

exogenous application of SA limits the formation of ethylene by inhibiting 1aminocyclopropane carboxylic acid synthase (ACS) activity. Smilarly, exogenous SA applications reduced the negative effects of drought on maize [50]. SA regulates the reaction of wheat seedlings to drought, and improves plant growth in water stress. It has been found that this effect is achieved by increasing moisture content, dry weight, SOD enzyme activity and chlorophyll content of wheat seedling [31].

In a study conducted in wheat, IAA and GA content decreased in drought conditions and ABA and proline content increased in leaves. Researchers stated that ABA and BA (benzyladenine) applications were involved in osmoregulation with proline production and ABA was more effective after grain filling and BA was more effective at an earlier stage in drought conditions [51].

GA3 and CK applications mitigated the negative effect of drought stress, especially at vegetative stage in corn plant [52].

The researchers state that BRs can mitigate the negative effects of various abiotic stress factors. One of them is the effect of plants against drought stress. It was reported that BRs application mitigates drought stress damage in tomato by regulating endogenous ABA concentration and antioxidant enzyme activity [53]. Similarly, it was found that BRs application improve the leaf water economy and  $CO_2$  assimilation, and ensure rice to resist drought [54]. Exogenous application of BRs increased SOD activity, yield, and plant growth parameters and improved drought tolerance of tomato [55].

The effect of JA against drought stress has been determined in many studies. Exogenous JA application to the *Allium cepa* mitigated the negative effect of drought on photosynthetic pigments and relative water content and resulted increase in plant growth under drought stress [56]. It has been determined that application of JA can be effective for acceptable corn yield in areas with limited irrigation [57]. Similarly, Miranshahi and Sayyari [58] reported that MeJA (derivatives of JA) application on savory plant improved growth by regulating proline content, antioxidant activity, relative water content, and essential oil content and yield under drought stress.

## Salt stress

Salt stress is another abiotic stress factor that negatively affects plant growth and yield in agricultural production and thus causes economic losses. Salinity effects negatively plant development by respond biochemical and physiological (such as mineral nutrients homeostasis, antioxidant metabolism, photosynthesis, osmolytes accumulation and hormonal signaling) [59]. Hormones are molecules that have effects on plant growth and are effective in regulating tolerance to stress factors such as salinity. Plant hormones, Ca ions and phospholipids, etc. act as an osmotic or homeostasis regulator to regulate plant growth and development under salt stress [8].

It is known that phytohormones have significant effects on salt stress tolerance in plants. It was determined that ethylene and GA reduce the negative effect of salt stress on plant by increasing plant growth or defense mechanism. It is emphasized that these two hormones interact with each other [60]. Changes in GA content of seeds in salt stress affect germination. It is known that the negative effect of salt stress decreases with exogenous GA applications improved germination properties of many plants such as okra [61], cucumber [62, 63], maize and pea [64]. GA3 seed application improved the seed germination and plant growth of wheat under saline conditions, and it were found to reduce the negative effect of salt stress by increasing leaf area, chlorophyll content, stomatal conductivity and photosynthesis [63]. Similarly, it was reported that, GA have positively effect the hypocotyl length, fresh and dry weight, and the water content of the okra seedlings under salinity [61].

ABA is a hormone that acts as an initiator to regulate plant water status in the event of stress, regulating growth by inducing related genes and with guard cells [9]. With increased internal ABA concentration, tolerance to salt may be high in the plant. As a matter of fact, ABA concentration was high in some saline resistant corn and tomato varieties [65, 66]. In salt stress, the level of endogenous ABA may vary in different organs of the plant, and it can create tolerance to stress by exogenous applications. It was determined that the ABA accumulation in the roots of corn plant in salt stress was higher than that of the leaves and the responses of leaves and roots to salinity differed according to ABA accumulation [67].

Khadri et al. [68] determined that the addition of 1 µM ABA to the nutrient solution before the salt stress was initiated reduced the negative effect of salt stress on beans. Auxins, one of the hormones important for plant growth and development, have important effects on plant tolerance under salt stress conditions. Zörb et al., [69] determined that IBA concentration was increased in the leaves and IAA concentration was maintained in the roots of corn resistant to salinity. These hormones have been reported to protect plant growth under saline conditions. Furthermore, the concentration of ABA in salt-resistant maize leaves increased and provided the acidifying of the apoplast, which is important for growth. In a study investigating the effects of IAA application on germination of wheat seeds in saline conditions, it was found that IAA increased the adaptation of salinity by regulating the synthesis of osmosis and osmotic substances in the root and shoot [70]. Salt stress decreased seed germination, seedling and plant growth and the damage of salt stress was mitigated with auxin applications [71, 72, 73]. The exogenous application of IAA and CK increased grain yield, 1000 grain weight and grain rate of rice under salinity. Also, the researchers reported that the contents of starch, glucose, fructose and sucrose in grain increased with IAA and CK application [72]. Similarly, exogenous IAA application and inorganic nutrient on maize plant caused increase photosynthetic pigment concentration and leaf Na+/K+ ratio, decreased membrane permeability by change the antioxidant enzyme activity, thus improving plant growth under salt condition [73].

BRs, known as steroidal phytohormone, which can be found in the natural structure of the plant, could tolerate the plant on salt stress. It was determined that the damaging effect of salt stress on wheat plant growth was reduced by application of 24-epibrassinolide, it increased grain yield under stress conditions and increased plant growth by changing photosynthetic rate [74]. Similarly, in another study, exogenous application of BRs as a foliar spray has been reported to improve the growth of wheat plants under salted and non-salted conditions [75]. Also, BRs have positive effect on tomato seed germination and plant growth, yield and physiological properties under salinity conditions [76]. Anuradha and Rao [77] reported that exogenous BRs application mitigated the negative effects of salinity on seed germination, root elongation and growth of rice by arranging pigments and increasing activity of nitrate reductase.

JA can activate the plant defense responses to different abiotic stress factors. It was determined that JA treatment reduces the negative effect of salt stress by regulating antioxidant enzyme activity in sorghum seedlings [78]. With the exogenous application of MeJA (Methyl jasmonate), it was determined that prevented decrease of endogenous CK concentration which is effective in plant growth and providing tolerance to salt stress [79].

It has been found that SA is more effective in regulating responses to biotic stresses in plants, however, SA responds to abiotic stresses such as salt stress. As a matter of fact, it was determined that foliar application of SA mitigated salt damage by increasing plant growth parameters and increasing photosynthetic activity in maize [80]. It was explained that exogenous SA improved abiotic stress tolerance depending on the nutrient status of the plant. SA application in cucumber improved the ability of plants to regulate endogenous SA levels after salt stress at adequate nutrient status [81]. SA has been proposed to develop an antioxidant system against salt stress in pea plants and to increase plant growth and yield by affecting many biochemical activities [82].

The CKs are can alleviate the negative effects of salt stress on plant growth. It was determined that salt tolerance was achieved by applying CK to wheat seeds [83]. It was reported that when the plant was exposed to stress, the concentration of CK increased temporarily, decreased under reasonable stress conditions, but reached concentrations higher than the initial level under severe stress [20]. Wang et al. [84] reported that broken in the homeostasis of CKs in cells could modulate response to salt stress via a ROS-mediated arrangement in plant. With the exogenous CK application alleviated the aging of the leaves, decreased the salt accumulation with the increase in antioxidant enzymes, and decreased the salt stress harm by increasing the K + / Na + ratio [85]. Under drought and salt stress conditions, CK enables plant growth to continue by activating gene expression that is effective in plant growth and preventing expression of genes related to premature senescence [86].

## Conclusion

Phytohormones, which are effective in many stages of plant growth and development, also have important functions in plant under stress conditions. Drought and salinity, which are the abiotic stresses, cause plant endogenous phytohormones decrease (IAA, GA, etc.) and increases (ABA, etylene, etc.) and reactions occur in the plant. In addition, it has been determined that tolerance of the plant to stress can be increased by applying exogenously these hormones to the plant in recent years. Further studies should be done on the effects of phytohormones to provide tolerance under adverse stress conditions in plants and thus, the advantages of hormones in plant cultivation in problematic areas can be utilized.

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# Natural Sweeteners as Food Additives

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#### Abstract

Sweeteners are functional food additives that give a sweet taste to food. They can be divided into two categories as natural and synthetic sweeteners. Synthetic sweeteners cannot be metabolized in the human body and provide no calories; hence, it is also called as non-nutritive sweeteners. Due to their low calorie, non-nutritive sweeteners has advantageous in terms of the control of obesity, hypertension, diabetes and dental caries. The increase in demand for low-calorie foods in recent years has led to an increase in the use of non-nutritive sweeteners. As a consequence, non-nutritive sweeteners have become available in many foods. On the other hand, the great interest on natural sweeteners have significantly increased over in recent years due to concerns about the long-term consumption of syntetic sweeteners as sugar substitutes. In this review, benefits, disadvantages, and resources of natural food additives are revisited.

Keywords: Natural sweteners, food additive, low-calorie, health

## 1. Introduction

Chronic, non-communicable diseases are affect negatively the lives of millions of people each year. In the prevention of these types of diseases has an important place the commercial development of non-caloric sweeteners to replace sucrose. Commercially available non-caloric sweeteners can be classified as natural and syntetic. Among the synthetics are best knowns saccharin, cyclamate and aspartame, which have been widely used in food manufacturing because of their high sweetening power and similar taste to sucrose. On the other hand, the demand for non-caloric sweeteners of natural origin, such as steviol glucosides, monellin, thaumatin, has significantly increased [1, 2].

## 2. Natural sweeteners

Sugars and polyols (sugar alcohols) are significant part of a human diet. Sucrose, glucose, fructose, lactose and maltose are sugars in the diet originating both from natural sources such as fruits and honey.

**2.1.** Sucrose, a disaccharide, is the most used table sugar. It is commonly available as either granulated sugar or table sugar and is the most used sweetening agent in the world. Sucrose is present in all green plants. Especially in sugarcane and sugarbeet are found high concentrations of sugars which is sufficient for commercial extraction [3]. The most important disadvantage of sucrose is relation between teeth decay and the consumption of this sugar. Namely, bacteria such as *Streptococcus mutans* and *S. sanguis* use this disaccharide and convert to pyruvic, acetic and lactic acid. Thereby, sucrose supports bacterial colonization dissolving the tooth enamel. Moreover, the very fast absorbance of sucrose can increase glycemic values. In addition to these, other diseases and disorders such as cardiovascular diseases, type II diabetes, metabolic

syndrome, cognitive decline, insulin resistance, cancer (breast, colon), obesity, hypertension and kidney disease are related to sugar consumption [2, 3]. Due to these negative properties of sucrose on health, the attention has directed to calorie-free sweeteners.

**2.2. Steviol glycosides:** *Stevia rebaudiana* (Stevia) extract, a natural non-nutritive sweeteners, is known with properties such as hypotensive, hypoglycaemic, antidiabetic, anticariogenic, antioxidant, antihypertensive, antimicrobial, antiinflammatory and antitumour activities. Also, it has a noncariogenic sweetener property. Steviol glycosides degrade at temperature higher than100°C [4]. The use of purified stevia extracts as sweeteners is at present permitted in numerous countries. Stevia, with a sweetening power 300 times greater than sugar, is a product used as additive for manufacturing of various food products such as ice creams, yogurts, cakes, sauces, drinks, bread, pastry, flavored milk, and spices (Carocho et al., 2017). Urban et al. [5] reported that there have been no reports of stevia-related allergy in the literature since 2008.

**2.3.** Polyols (Sugar alcohols), organic alcoholic compounds containing several hydroxyl groups, are usually applied in food science. Common polyols used in food industry consist of mannitol, sorbitol (hydrogenated monosaccharides), lactitol, maltitol, isomaltitol (hydrogenated disaccharides) glycerol, erythritol, xylitol, and so on [6]. Sugar alcohols might be added to foods as alternative sweeteners. They are nutritive, low calorie, digestible carbohydrates. The foods with these sweeteners are called as sugar free. Even though are not specified ADI values, they are known with laxative effect and gastrointestinal symptoms when eaten in excessive [3, 7].

**2.3.1. Sorbitol,** the sugar alcohol, is produced through the hydrogenation or electrolytic reduction of glucose. Besides, it has a wide range of area of usage in the food, medicine, and cosmetic industry. Sorbitol is recognized as one of the top twelve building blocks produced from renewable feedstocks [8]. Sorbitol is 50-60% sweeter than mannitol and it rather used as sweetener. This polyol is used in baked goods, sweets, sausages, and drinks. Although there is no evidence about sorbitol toxicity, results of a study conducted in 2016 reported that sorbitol can be genotoxic. Yet another study determined that patients with irritable bowel syndrome have negative gastrointestinal reactions against polyols. Also, it seems to be safe for healthy individuals, although there are reports of laxative effects when consumed in high doses. It has been reported that this effect is related to the fructose:glucose:sorbitol ratio, and not to sorbitol itself [2].

**2.3.2. Xylitol** is a representative sugar alcohol obtained by hydrogenation of xylose and widely used as a low calorie sweetener. It is known to increase salivation. Thus xylitol has reducing effect teeth decay by helping teeth cleansing and reducing the bacterial load in the mouth. It causes very little increase in glucose level and insulin in blood. In biological production of xylitol is used either xylose fermenting or non-xylose fermenting

microorganisms. Among those microorganisms, *Saccharomyces cerevisiae*, a GRAS microorganism and used for various foods manufacturing, is found. Xylitol is also found in many fruits and vegetables. Xylitol is popularly used in the formulation of various food such as chewing gum, cold refreshments beverages, confectionery products [2, 9,10].

**2.3.3. Mannitol** is a polyol that exists in most fruits and vegetables such as pumpkin, celery, onion, olive and also in the mycelium of diverse species of *Aspergillus* and *Penicillium* fungi genus. In addition to these both homofermentative and heterofermentative lactic acid bacteria (LAB) also produce mannitol. Mannitol is also found in various wines due to the metabolism of LAB or yeasts. Due to its beneficial physiological effects, mannitol is used as a functional sweetener in the food industry. It is employed in foods as an anticaking agent. In addition, mannitol has also various applications in pharmaceutical, chemical and medical industries [2, 11].

**2.3.4. Maltitol (E965)** is a crystalline polyhydric alcohol formed by hydrolyzation, reduction and hydrogenation of starch. It is about 75–90% of the sweetness of sucrose and has been used usually in the manufacturing of sugar-free chocolate, sweet coatings, lactic products, baked goods, muffins, jams, jelly, and chewing gum. Of all the sugar alcohols, it is the most resembling the flavor of sugar. It is not cariogenic and is safe for diabetics. Maltitol is an favorable sugar substitution for the people with diabetes. It has a very slow digestion rate and may be partially fermented in the colon [2, 12, 13].

**2.3.5.** Lactitol is not found in nature and it is industrially produced through catalytic hydrogenation (a set of chemical reactions in which hydrogen is added to a reactive functional group)of lactose. Although sugar alcohols are not essential nutrients, its consumption has been clinically linked to a number of health claims. In general, sugar alcohols are a low source of energy for oral bacteria contrary of other sugars (sucrose and starches). It is not cariogenic and is one of the most frequently recommended laxative agents to treat chronic constipation. lactitol can also use as a cryoprotectant, a surfactant and stabilize agent. It has prebiotic properties. Additionally, it is also used manufacturing of bakery products, chocolate and confectionary products, dessert, chewing gum, ans ice-cream [14, 15].

**2.3.6. Isomaltitol (E953)** is a polyol obtained through enzymatic transformation of sucrose and is stable at high temperatures and has a very low hygroscopic value. This molecule is not absorbed by the small intestine, and is easily fermented in the colon by colonic bacteria. Isomalt is used in chewing gums, gelatins, chocolate, coatings, baked products and yogurts. Isomalt has low glycemic index and relatively small caloric value. Isomalt has also prebiotic properties, promoting the growth of *Bifidobacteriae* [2,16].

# 2.4.Nitrogenated derivatives

Monellin, brazzein and thaumatin are among the three sweetest known proteins. Although numerous proteins and peptides can show beneficial effects in living organisms, such as antimicrobial, antioxidant, antihypertensive, and antiinflammatory effects, the bioactivities of this sweeter proteins are unknown. But they have an important place in the food industry [17].

**2.4.1. Thaumatin** is a natural proteinaceous sweetener extracted from the fruits of a tropical plant, known as *Thaumatococcus daniellii* Benth growting in Western Africa [18,19]. It has approximately 100,000 times more sweetening power from sucrose. Thaumatin is quite proper for industrial applications because of being an low-calorie sweetener [20,21]. Thaumatin which has taste like carbohydrate sweeteners has high water solubility and is stable to heat and acidic pH [19]. The major foods where it is used of thaumatin are soups, sauces, processed vegetables and egg derived products Recently is reported that several studies have focused on the production of recombinant thaumatin through microorganisms and transgenic plants [2].

**2.4.2.** Monellin is isolated from African berries and is a members of the family of sweet proteins. It is one of the most sweet proteins used instead of carbohydrates in the diabetic diets and pharmaceuticals. However, it loses its sweetening power when the protein is heated above 50°C. This situation is the critical problem related to its usage in the food or pharmaceutical industry [20,22,23]. Monellin has a very low caloric power, therefore, it is used as excipients for food and pharmaceutical preparations [22].

**2.4.3. Brazzein** is a sweet-tasting protein isolated from Pentadiplandra brazzeana Baillon being an edible fruit growing in West African. It is an attractive sweetener because of its high sweetness, sugar-like taste, and good stability at high temperature and wide pH ranges. It is approximately 500–2000-fold sweeter than sucrose. Brazzein exhibits high water solubility, high pH durability and thermostability. It is rich in terms of lysine but contains no methionine, threonine or tryptophan [2,24]

## 2.5.Flavonoid derivatives

**2.5.1.Neohesperidin dihydrochalcone** (NHDC) (E959) is another sweetener that comes from the skin of the immature fruits of *Citrus aurantium* L. This sweetener is at a certain level hygroscopic and is stable at high temperatures like pasteurization. NHDC has wide application in various foods (ice creams, chewing gums, pastry, water-based flavored drinks, milk and derivatives, snacks, confectionary foodstuffs, beer, soups.) as a low caloric sweetener [25, 26]

## **3.** Conclusions

In recent years, a worldwide increase in the prevalence of overweight and obesity has occurred .The overweight and obesity are important risk factors a lot of diseases like diabetes mellitus, hypertension, cardiovascular disease, several cancers and premature death. In this context, weight control is extreme important. Therefore sugar is began to replace with low-and non-nutritive sweeteners in drinks and foods and become widespread intake of alternative sweeteners. However, usage of alternative sweeteners (especially synthetic sweeteners) is created concern for their impact on metabolic health.

Consequently, natural sweeteners has begun to interest of consumers. In this context, new studies about natural sweeteners have been center of interest

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# Some Pathogenic Bacteria in Raw Milk and Products

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## Abstract

More than 200 known diseases are transmitted through food by various agents including bacteria, fungi, viruses and parasites. As a result, prevention of diseases and deaths associated with foodborne pathogens continue to happen a major public health concern. In addition, food safety is a global problem. The increase in the import and export of food products may also lead to the emergence of new diseases in geographical areas where foodborne pathogens are never observed. Milk can contain a variety of microorganisms and can be an important source of foodborne pathogens. The presence of foodborne pathogens in milk can result from direct contact with contaminated sources in the dairy farm and from the udder of an infected animal. The prevalence of foodborne pathogens in milk is influenced by a number of factors, including farm size, number of animals on the farm, hygiene, farm management practices, differences in sampling, and sample types evaluated, the detection methodologies used, geographic location and season.

Keywords: Food safety, raw milk, raw milk product, pathogen

# 1. Introduction

Foodborne diseases are among the major public health problems that are constantly increasing. Today, in both developed and developing countries, a small proportion of foodborne illness cases consult to healthcare organizations and the cause of the disease can only be identified at the species level [1]. Factors that cause foodborne infections and intoxications include chemicals, natural food toxins, metals, pesticides, detergents, plastics, parasites and microorganisms. Among microorganisms, especially bacteria are the main responsible for many foodborne diseases. Bacteria growing in foods that are usually prepared and cooked under unhygienic conditions cause food poisoning [2].

Microorganisms naturally existing in foods and microorganisms that are contaminated to food from the environment constitute the microbial flora of foods. Microorganisms that can be found in foods can be examined under four main groups according to their basic properties [3]:

- 1. Microorganisms called as 'starter' used for to be obtain of fermented foods
- 2.Saprophyte microorganisms causing to spoilage of foodstuffs
- 3.Pathogenic microorganisms causing to intoxication and infections
- 4. Inert microorganisms (neither beneficial nor harmful)

The risk of exposure to potentially pathogenic bacteria that can cause disease in humans through consumption of raw milk is much higher in rural areas. The data clearly show that consumption of raw milk increases the risk of direct contact with foodborne pathogens and / or its toxins and is therefore a risk factor for human-induced diseases. Although pasteurization of milk is seen

as an effective method to eliminate foodborne pathogens, some dairy products are not pasteurized (some special cheeses). In addition, some pathogens may survive and grow in post-pasteurization processing environments. Thus, they lead to re-contamination of dairy products. The increasing number of cases in which foodborne pathogens are detected in milk and ready-to-eat dairy products clearly shows that pasteurization is not the ultimate means of controlling of dairy pathogens. Fecal and foodborne pathogen contamination is likely to occur during milking, collection and storage of raw milk [4].

## 2. Some pathogenic bacteria in raw milk

Pathogens can be contaminated to raw milk from infected breasts, faeces, other secretions, the environment or equipment. Among these microorganisms find pathogenic bacteria such as *Staphylococcus aureus*, *Salmonella* spp., *Echerichia coli*, *Campylobacter* spp., *Streptococcus agalactiae*, *Yersinia enterocolitica*, *Bacillus cereus*, *Clostiridium perfringens*, *Brucella* spp and *Mycobacterium tuberculosis*. However, sometimes milk may be drunk as raw, may be made cheese from raw milk, or milk may contaminated after pasteurization [5]. In past years, pasteurization conditions have been standardized depending eliminate of *Mycobacterium tuberculosis*, one of the most serious pathogenic bacteria present in milk. Today, pasteurization standards are based on the destruction of *Coxiella burnetti*, the most heat-resistant milk-borne zoonotic pathogen known. In the past 30 years, previously unknown foodborne bacterial infections caused by *Campylobacter jejuni*, *Listeria monocytogenes* and *Escherichia coli* O157: H7 bacteria have emerged as major causes of human morbidity and mortality. The pathogens that are predominantly present in raw milk today are *C. jejuni*, *Salmonella* spp., *Staphylococcus aureus*, *L. monocytogenes*, *E. coli* O157: H7 and *Yersinia enterocolitica* [6].

# 2.1. Mycobacterium tuberculosis and Mycobacterium bovis

*Mycobacterium tuberculosis* and *Mycobacterium bovis* are classic agents that cause human and bovine tuberculosis, respectively. *M. bovis* was isolated from bovine tissues and milk as well as *M. tuberculosis*. These two tuberculosis bacilli can be transmitted directly (by aerosol inhalation) between humans and animals, or indirectly from animals to humans (through contaminated raw dairy products and meat consumption) [7].

# 2.2. Coxiella burnetii

Ticks are considered to be the natural primary reservoir of *Coxiella burnetii* and responsible for the spread of infection in wild animals and transmission to domestic animals. Cattle, sheep and goats are the main sources of human infection. Infected animals release highly stable bacteria with urine, feces and milk. In humans, infection is mainly caused by inhalation of contaminated aerosols on amniotic fluid, placenta, or contaminated animal wool. Persons (veterinarians, agricultural workers, butchers) which keeping in contact with laboratory staff working with farm animals and infected animals constitute the most important risk group [8].

# 2.3. Staphylococcus aureus

*Staphylococcus aureus* is an important cause of mastitis in dairy cows in the world. The bovine mammary gland may be an important reservoir of enterotoxogenic strains of *S. aureus*. Although many studies have documented that foodborne pathogens, which are important for public health, are isolated from milk and can cause disease in humans, people continue to consume raw milk. Many farm-owned families consume raw milk. Because it is a traditional

practice and it is cheaper to buy milk from collection tanks than to buy pasteurized retail milk. In addition, some consumers believe that raw milk has a higher nutritional value than pasteurized milk [4]. Staphylococcal food poisoning arising from cheeses is an important problem which is commonly encountered all over the world. The most common causes the emergence of *S. aureus* in cheese are manufacturing cheese from milk containing high amounts of *S. aureus*, insufficient starter culture activity, contamination of milk after pasteurization and unfavorable conditions during processing and storage of the product [9].

## 2.4. Salmonella

Bacterial pathogens such as *Salmonella* often colonize the gastrointestinal tract of cattle. Human diseases caused by these bacteria are generally associated with consumption of beef, raw milk and raw dairy products. *Salmonella* in humans is highly variable, but most are considered to be human pathogens. Virulence of *Salmonella* in humans is highly variable, but all *Salmonella* bacteria are considered to be human pathogens. However, many bacteria that are infectious for humans commensally live in cows and do not cause disease, including *Salmonella enterica*. This infection may or may not affect health, milk production, or reproduction [10].

# 2.5. Escherichia coli O157: H7

*Escherichia coli* O157: H7 is an major pathogen causing foodborne diseases and healthy cattle are a reservoir of *E. coli* O157:H7. One particularly its dangerous species is known as enterohemorrhagic *E. coli* (EHEC). Infection with EHEC strains is often associated with foodborne outbreaks following consumption of contaminated milk, dairy products and, other foods and lead to hemorrhagic colitis (bloody diarrhea) and hemolytic uremic syndrome in humans. The risk of EHEC infection in humans from dairy farms can occur through the consumption of raw milk and dairy products, and through contamination of the dairy environment. Therefore, to reduce the risk of EHEC infection, it is important to improve the measures of control and also to prevent propagation of EHEC strains between animals, the environment and humans [11].

# 2.6. Campylobacter

*Campylobacter*, one of the major causes of diarrhea in the world, is carried by many different animal species and is easily transmitted by contaminated water or food. Bovines can carry the organism, which can persist in mammary glands, and raw milk can be a vehicle for transmission. Among the causes of campylobacteriosis outbreaks caused by raw milk were reported to be of educational field trips to farms by school children [12]. *C. jejuni* infection can occur after consumption or contact of contaminated poultry, meat, milk or water, or after contact with animals and birds [1]. *C. jejuni* is sensitive to heat, drying, air (oxygen), and acidic pH. It is easily inactivated by normal pasteurization norms. It causes mastitis in cows [13].

# 2.7. Streptococcus agalactiae

*Streptococcus agalactiae* causes mastitis in dairy cows. In cattle, intra mammary infectionis are usually chronic and subclinical. High somatic cell numbers and reduced milk quality and production are often observed [14]. Transmission of the pathogen within dairy herds may occur during milking via milkers' hands, liners and use of common milking equipments contaminated with milk from infected cow [15].

# 2.8. Yersinia enterocolitica

*Yersinia enterocolitica* is the causing agent to a great number of diseases, fundamentally gastrointestinal manifestations, in addition to serious complications such as mesenteric lymphadenitis, reactive arthritis and sepsis. some studies reported that *Y. enterocolitica*, *Y. frederiksenii* and *Y. intermedia* were isolated from raw cow milk [16]. In a study that reported an outbreak of *Yersinia enterocolitica* O:8 in relation to the consumption of bottled pasteurized milk in a local dairy company, it was found that no deficiencies were detected in the pasteurization equipment but that milk bottles were rinsed with untreated well water before filling. Although the exact contamination mechanism of milk is not clear, it was stated that contamination after pasteurization is the most likely source. They reported that *Y. enterocolitica* to a variety of sources, including contamination after pasteurization, contamination of milk containers, poor bottle-washing techniques [17].

# 2.9. Bacillus cereus

*Bacillus cereus* a rod-shaped, Gram-positive bacterium that can produce stress-resistant endospor in extreme condition, resisting adverse conditions such as high temperatures, ultraviolet radiation, and damaging chemicals [18]. In milk and dairy products, *Bacillus cereus* may adversely affect product quality because it produces various extracellular enzymes that may be responsible for decrise of organoleptic quality. *B. cereus* can contaminate to milk environment mostly from improperly cleaned and sterilized equipment, from various sources during production, transport and processing. The hydrophobic properties of its endospores and their resistance to heat, drying and disinfectants cause them to bind to processing equipment and survive cleaning procedures. It is stated that contamination can occur in all process steps for spore-forming bacteria are in almost all natural environments [19].

# 2.10. Clostridium perfringens

*Clostridium perfringens* is a Gram-positive, spore-forming anaerobic bacterium that is responsible for a wide range of diseases in animals and humans. This bacteria are common in environments such as soil and water and are usually found in the intestines of animals. *C. perfringens* does not show adherence and invasive properties to healthy intestinal mucosa. The pathogenicity of the organism is associated with the various toxins produced from its [20].

# 2.11. Brucella

The infection of *Brucella* species is commonly occur in our country. *Brucella* is a Gramnegative, small rod-shaped bacteria. It is transmitted to humans from unpasteurized milk and dairy products or from the daily products of infected animals and by direct contact with infected animals. Infection can affect all organs and systems [21].

# 2.12. Listeria monocytogenes

*Listeria monocytogenes* are widely distributed in natüre. It can be isolated from various sources such as soil, fertilizer, leafy vegetables, raw meat and poultry. It is usually the cause of stillbirths or neonatal deaths. Surviving babies often develop meningitis, which can be fatal or result in permanent mental retardation. Bacterial meningitis is very serious and death can occur in as little as a few hours. *Listeria monocytogenes* are heat-sensitive and inactivated by

pasteurization. However, there are some approximations concerning its resist traditional pasteurization in the intracellular phase [13].

## **3.** Conclusions

There are many difficulties in providing a safe and nutritious food supply. Because all aspects of food production (from field to fork) need to be considered. Globally, the incidence of foodborne diseases is increasing and international food trade is disrupted by disputes over food safety and quality requirements. In prevent foodborne diseases is necessary a strong knowledge-based approach to eliminate all the complex troubles of continuing to improve food safety and public health. Research should not only be conducted to solve food safety problems, but also the results of these studies should be communicated effectively to producers and consumers.

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# Prediction of Plant Height in Wheat with Different Salt and Bacteria Applications by Using Fuzzy Logic Method

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## Abstract

Fuzzy logic is a mathematical discipline based on fuzzy set theory. Fuzzy logic, which is based on fuzzy clusters, generally assists in the realization of identical processes to human thought, and helps in modeling uncertain data that often occurs in the real world. Fuzzy logic can be defined as a set of mathematical principles aimed at the presentation of knowledge based on the degree of membership rather than traditional binary logic. In cases where the system is complex and the analysis cannot be performed with classical methods and the qualifications of the information is uncertain, the fuzzy logic method is very suitable. Since fuzzy logic approach does not require mathematical model, time-varying and nonlinear systems whose mathematical model is not well defined are the most successful applications.

The data of the current study consisted of two input factors and one output factor. First factor is salt concentration with 5 levels. Second factor is bacteria application with 6 strains and control level. The coefficient of correlation of the plant height prediction by means of regression analysis was found very small (r=0.2720). Therefore, fuzzy logic approach is applied to the solution of the problem. For fuzzy system modelling MATLAB programme was used. As a result of the fuzzy logic application the coefficient of correlation was found as r=0.8757.

Keywords: Estimation, Fuzzy sets, Mamdani inference system, plant height, salt concentration.

# Introduction

Concept of fuzzy theory and fuzzy set was first presented by Zadeh [1]. Since then fuzzy theory was explored and used to develop model and to solve many partially known system. Being a complex and partially known system, agriculture is a potential candidate to use fuzzy theory [2]. Fuzzy logic and fuzzy inference provide an ideal method to express impreciseness and certainty, thus offering a better level of accuracy and robustness. Many applications of fuzzy logic and fuzzy inference systems in agricultural industry have proved that fuzzy logic system is an important and powerful tool to be used to solve many problems encountered in agro-industry. Fuzzy logic can be applied in many aspects and phases of agriculture, from land and soil suitability, climate prediction, pest and weed management as well as product quality assurance [3, 4]. Ceyhan-99 wheat variety by superior adaptability, efficiency and quality is one of the most cultivated varieties of bread wheat in Turkey and has the potential to also be trained in a similar ecological conditions world [5]. The aim of this study is to estimate the height of the wheat plant with application of different salt doses and bacteria.

## **Material and Method**

# Material

The data used in the paper is Ceyhan-99 bread wheat cultivar [5]. The bacteria inoculated in wheat are *Stenotrophpmonas maltophilia* P TV14B, *Bacillus* sp. P TV119E, *Bacillus atrophaeus* N TV83D, *Cellulomonas turbata* N TV54A, *Kluyvera cryocrescens* NP TV113C, *B. atrophaeus* + *Bacillus* sp. (binary combination) TV83D + TV119E and they were isolated from Van Lake Basin. The strains were identified in Bacteriology Laboratory of Atatürk University Faculty of Agriculture Department of Field Crops and the impacts of PGPR tested under greenhouse and field conditions. In order to create salinity stress in germination medium, 0, 50, 75, 100 and 125 mM NaCl solutions were applied to the pot soil under field conditions in line with [6] to create unsalty, slightly salty, salty, very salty and too salty levels, respectively.

## Method

Fuzzy systems for either modeling or control have similar operations. Figure 1 shows the block diagram of the structure of conventional fuzzy systems for modeling and control. The fuzzy system is composed of four function blocks: fuzzification, rule base, inference engine, and defuzzification. The mechanism of fuzzy systems is as follows: the measurements x of the outside world in the form of crisp data are transformed by fuzzification into linguistic values. Then the linguistic values are processed by the fuzzy rules in the rule base in the form of "if-then" through fuzzy implication. The output expressed in fuzzy sets after fuzzy implication is finally transformed by defuzzification into a nonfuzzy (crisp) output as the output of the system to the outside world [7]. MATLAB Fuzzy Logic Toolbox was used for modeling.

## **Results and Discussion**

For the FL realized via the MATLAB program [8], 2 inputs (Salt and Bacteria) and 1 output (Plant Height) were available. Fuzzy sets for Salt was developed as illustrated in Figure 1.

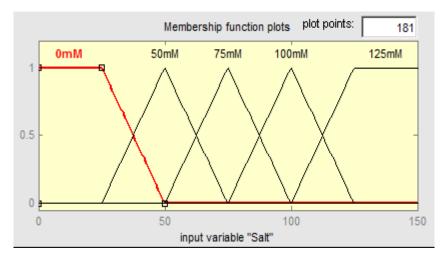


Figure 1. Fuzzy sets for Salt variable

The Bacteria graph formed in MATLAB is illustrated in Figure 2. In the fuzzy logic model (FLM), however, this input is entered as discrete.

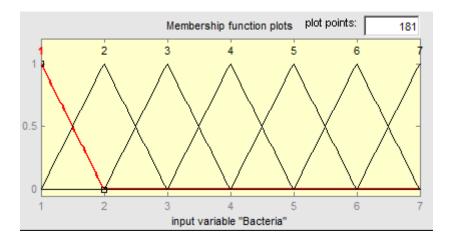


Figure 2. Fuzzy sets for Bacteria variable

Figure 3 shows the output graphic Plant Height (PH). The pH graph, divides the output data into five ranges as very low, low, normal, high and very high, and shows the verbal expression memberships. For the very short and very high fuzzy sets the trapezoidal membership function was selected; for the short, moderate and high fuzzy sets the triangular function was used.

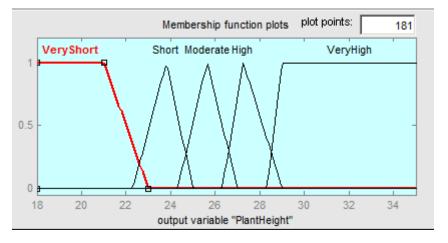


Figure 3. Fuzzy sets for PH variable

Rule base of the FLM was contained of 35 rules. Below are some rules:

Rule 1: If Salt is '0 mM' and Bacteria is '1<sup>st</sup>' then Plant Height is 'Moderate'. Rule 6: If Salt is '0 mM' and Bacteria is '6<sup>th</sup>' then Plant Height is 'Short'. Rule 10: If Salt is '50 mM' and Bacteria is '3<sup>rd</sup>' then Plant Height is 'Moderate'. Rule 16: If Salt is '75 mM' and Bacteria is '2<sup>nd</sup>' then Plant Height is 'Short'. Rule 20: If Salt is '75 mM' and Bacteria is '6<sup>th</sup>' then Plant Height is 'Very short'. Rule 24: If Salt is '100 mM' and Bacteria is '5<sup>th</sup>' then Plant Height is 'Moderate'. Rule 35: If Salt is '125 mM' and Bacteria is '7<sup>th</sup>' then Plant Height is 'High'.

The relationships between PH and Salt and Bacteria were shown in Figure 4 (a,b), respectively.

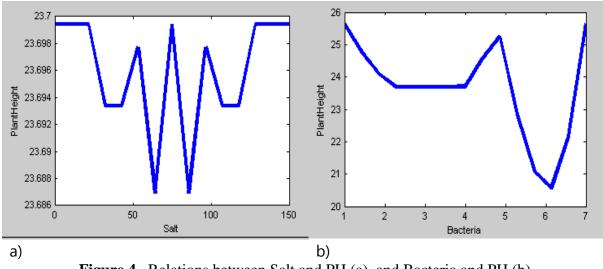


Figure 4. Relations between Salt and PH (a), and Bacteria and PH (b)

Figure 5 shows the 3-dimensional relationships between the inputs and the output.

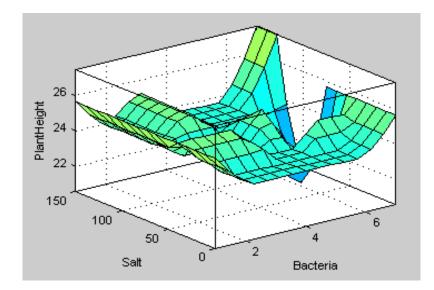


Figure 5. Relation between Salt, Bacteria and PH

For example, with given 75 mM Salt, and 4<sup>th</sup> Bacteria, the observed output value appears next to the statement Plant Height (23.7 cm). Other results can be found by supplying different input data Thereafter, all data were run through the FL model, with the results of PH (Figure 6).

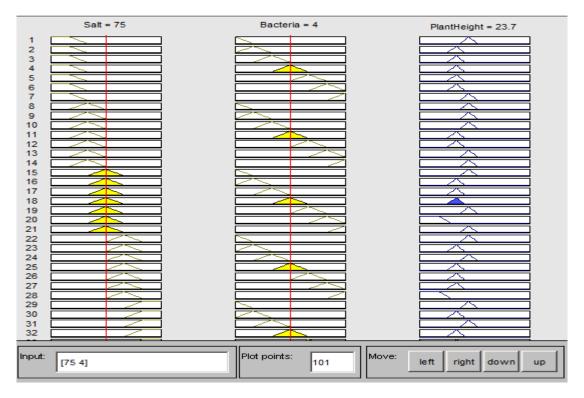


Figure 6. Result window of FLM

The coefficient of correlation between real estimated and predicted by means of FL was r=0.8757 (Figure 7).

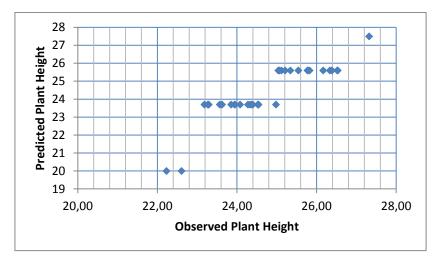


Figure 7. Correlation between predicted and observed PH

Observed and Predicted data distribution was shown in Figure 8.

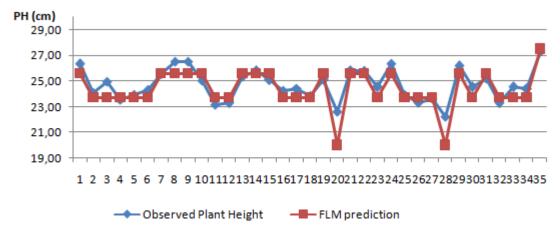


Figure 8. Predicted and observed PH

## Conclusions

This paper was aimed to estimate plant height by means of the FL. Using the input data of Salt doses and Bacteria model was able to predict plant height at a level of success. In the end, the study's FL model revealed coefficient of determination rate of r=0.8757. However, it was found by conventional linear regression model as r=0.2720. It can be said that a fuzzy logic model can be applied in areas where conventional methods is not productive. Considering some different factors, a more punctual and efficient system can be developed in the future.

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# Application of HACCP Principles in the Production of Cooked Sausage with Addition of Hawthorn Fruit

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## Abstract

The article considers the problems of ensuring quality and safety of food products and introduction of quality system based on the principles of HACCP. The paper presents results of studies of critical control points based on the HACCP system, which ensure quality control and safety in the production of meat products (cooked sausages with addition of hawthorn fruit). In technological process, stages of acceptance of raw materials (horse meat, flour from unconventional raw materials (hawthorn), spices, salt and spices), heat treatment, transportation and storage are attributed to the CCP, which should be controlled and monitored to minimize or eliminate potential hazards.

Key words: cooked sausages, meat, vegetable ingredients, HACCP.

## Introduction

Currently, the main requirement for ensuring safety and quality of meat and meat products in food production throughout the technological chain is implementation of the international standard ISO 22000: 2018, which contains principles of HACCP (Hazards analysis and critical control points (CCP)), and monitoring from acceptance of raw materials to the consumer [1,2]. Since 2013, Technical Regulations of the Customs Union (TR CU) have been introduced in action, which establish requirements for food products, production processes, storage, transportation, distribution and disposal. For the first time, requirement has been established in TR CU 021/2011 "Food Safety", which stipulates that the manufacturer, when implementing production processes consistent with established food safety requirements, required to create, introduce and facilitate procedures based on the HACCP principles [3].

Hazard to the safety of food products are biological, chemical or physical, contained in food products, also the state of food products, which can potentially adversely affect human health. Meat and meat products obtained by processing from meat of animal origin occupy a special place in daily diet of the population. They serve as source of obtaining in the human body most essential amino acids and easily digestible iron. Amino acids are evenly involved in protein synthesis. If one of ten amino acids does not get evenly into the human body, then the synthesis will stop and protein starvation will begin [4-6]. As for iron, it is also found in large quantities in plants, but not in the form in meat products, "hemic" iron (included in heme, which taste is part of hemoglobin beef), due to this, meat iron is absorbed much faster and better than vegetable. For majority of the population, sausage is product that distinguished by high palatability and very popular among consumers. Sausage is product of minced meat boiled in casing and cooked, fried and then cooked until ready to eat [4,5,7]. Expertise of cooked sausages

is performed according to GOST 33673-2015 "Cooked sausage products. General specifications".

The purpose of the work is to determine critical control points in production of cooked sausages with addition of hawthorn fruit.

According to the "Codex Alimentarius" Manual, 12 steps have been identified to implement the 7 principles of HACCP. The implementation of these 12 steps involves development of the HACCP plan, which is documented certificate containing details of all critical limit positions for product safety and quality. In this regard, it was advisable to develop HACCP plan in the production of cooked sausages with addition of hawthorn.

## Materials and research methods

The objects of research were raw materials (horse meat, fat, fruits of hawthorn, salt, sugar, spices), samples of finished products (cooked sausage) obtained in laboratory and production conditions.

In the work for analysis of the CCP was used ISO 22000: 2018, taking into account concepts such as prerequisite programs (PRPs) and operational prerequisite programs (OPRPs) and TR CU 021/2011 «Food Safety», TR CU 034/2013 "Safety of meat and meat products» [8,9].

The fruits and flowers of hawthorn species have complex chemical composition and contain number of organic acids, sugar, carotene (provitamin A), pectin and tannins, sorbitol, choline, acetylcholine, quercetin, emigdalin, thiamine, riboflavin (vitamin B2), anthocyanins, trace elements, number of potent alkaloids, proteins, catechins, flavonols and other organic substances. These biologically active substances, combining high physiological effectiveness with small amount of active principle, make it possible to use the flowers and fruits of hawthorn as medicinal raw material. They are used for functional disorders of cardiac activity, hypertension, angioneurosis, angina pectoris, tachycardia, atrial fibrillation, myosthenia, general atherosclerosis, climacteric neurosis and other diseases. The antispasmodic effect of hawthorn preparations is associated with presence of triterpene compounds and flavonoids in the plant. The fruits of many species are edible, have high palatability, sweet or sour-sweet in ripe condition.

## Research results and discussion

For each possible hazard, risk analysis was carried out taking into account probability of occurrence of the factor and importance of its consequences and compiled list of circumstances by which the risk exceeds permissible level.

The production of cooked sausages includes following processes. Input control and acceptance of raw materials, food ingredients, additives and spices. All raw materials, food ingredients, additives, spices must comply with TR CU 021/2011 "Food safety", TR CU 034/2013 "Safety of meat and meat products".

Cutting, deboning, trimming of meat raw materials is carried out in production facilities at an air temperature of not higher than 12 °C, relative humidity not higher than 70%.

Chilled raw materials with temperature in thickness of muscles of  $2 \pm 2$  ° C or defrosted with temperature of  $1 \pm 2$  ° C are sent to the boning. Duration of cutting, deboning and trimming should be no more than 2 hours.

Preparation of minced meat is carried out in cutters designed for grinding frozen meat. The prepared meat and bacon in accordance with recipe were loaded into the cutter in the following order: premium horse meat, fat, spices, salt, sodium nitrite (in solution).

The prepared meat was filled into an artificial protein shell with diameter of 50 mm. After filling the shell, staples were applied to the ends of the loaves.

Heat treatment. The precipitation was carried out at temperature of 0-4<sup>o</sup>C, for 2 hours, then subjected to frying (at temperature of 80-85<sup>o</sup>C, for 100 min), cooking (at temperature of 80-85<sup>o</sup>C, for 50-80 min) and cooled to temperature inside products 8<sup>o</sup>C (within 20 minutes). Hazardous factors established in TR CU 021/2011 "Food safety", TR CU 034/2013 "Safety of meat and meat products" were considered significant regardless of results of assessment. An analysis of serious hazardous circumstances and the application of algorithms for finding CCP, which affect the quality and safety of cooked sausage with addition of hawthorn. The summarized results are presented in table 1.

	Zaiù Anarysis in the	production of bol	led sausage with haw	
Name of	Process	Considering the	Controlled signs	Preventive action
operation	parameters	factors		
Reception of raw material	The meat should be frozen or the meat temperature	Microbiological	The presence of vegetative pathogens	Monitoring of supporting documentation
	should not exceed 80C. Hawthorn flour should be clean, without foreign smell. Spices must comply with regulatory		Cold chain interruption and growth of microorganisms, which present prior to acceptance of raw materials	Work instruction: compliance with temperature regime
	documents	Chemical	The presence of undesirable chemical hazards (toxic elements, antibiotics)	Quality control of raw materials
		Physical	Glass, metal, plastic, small bones, wool	
Cooking minced meat in cutter	High content residual nitrite sodium (not higher than 10 g sodium nitrite per 100 kg of raw meat)	Chemical	High dosage of sodium nitrite	Compliance with the dosage of drug according to the recipe
Precipitation	Non-compliance with technological parameters (at a temperature of $0-4^{0}$ C, within 2 hours)	Microbiological	Growth of pathogenic microflora	Compliance with temperature conditions

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Table I - Hazard	Analysis in the	production of poned	sausage with nawinorn
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Heat treatment	Compliance with temperature and humidity conditions	Microbiological	Growth of pathogenic microflora	Sensor control, laboratory control
Packaging	Contamination outside of the product	Microbiological	Growth of pathogenic microflora	Laboratory control
Transportation and storage	The growth of microorganisms on the surface. Storage temperature $T = 0-8^{\circ}C$	Microbiological	Growth of pathogenic microflora	Compliance with temperature parameters, transportation rules, washing and disinfection of vehicles

In the technological process, stages of accepting raw materials (horse meat, hawthorn flour, spices, salt, sugar), heat treatment of products, transportation and storage were attributed to CCP, which should be controlled and monitored in order to minimize or eliminate potential hazards. CCPs were studied and all data obtained was documented.

These production stages and offered control actions are presented in the Table 2.

Table 2 - CCP risk assessment and control actions in the production of boiled sausage with hawthorn

ССР	Risk	Control actions	Degree of risk
CCP 1	Reception of raw material	Safety indicators in accordance with regulatory documents, antibiotics are not allowed. Storage temperature control	High
CCP 2	Heat treatment	Compliance with temperature and humidity conditions in the chamber	High
CCP 3	Transportation and storage	Temperature sensor control	High

## Conclusions

Thus, HACCP system is one of the effective tools in the field of ensuring safety and quality, which necessary for food products, based on study of serious circumstances, as result of which it can be used to make decisions related to food hygiene, creating programs to ensure quality. The analysis of the most possible hazards in production of cooked sausage with addition of hawthorn was carried out. As control parameters, temperature changes in technological process of production were adopted. A monitoring system for monitoring on the CCP and corrective actions were also proposed. Further, for high-quality operation of the HACCP system necessary

to constantly maintain and improve it. In this case, necessary to establish internal audits according to the plan and in timely manner.

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# DEVELOPMENT OF PROCESS OF VACUUM-ATMOSPHERIC DRYING OF HELIANTHUS TUBEROSUS

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## Abstract

In the given article the results of development of vacuum-atmospheric drying of Helianthus Tuberosus or Jerusalem artichoke tubers are considered. The method of vacuum-atmospheric drying of large-dispersed materials includes vacuum drying of material to intermediate humidity and atmospheric drying to final one. Atmospheric drying of the material is carried out by waste heat of condensation of refrigerating agent. It was found at the same intensity of vacuum and atmospheric drying the nature of drying curves has a similar appearance. Also compliance of vacuum and atmospheric drying the constant rate of above drying methods. Optimal regimes of vacuum-atmospheric drying of dried materials are developed.

**Keywords:** vacuum, drying, Helianthus Tuberosus, Jerusalem artichoke, tubers, method, atmospheric.

## Introduction

Topinambour (Helianthus tuberosus) is a perennial tuberous plant with therapeutic properties. Tubers contain soluble polysaccharide inulin, nitrogenous substances, B group and C vitamins, organic and fatty acids. Mineral substances of topinambour are represented by zinc, silicon, phosphorus, iron, potassium, calcium, magnesium and copper. Many studies confirm the value of Jerusalem artichoke tubers as a functional product [1-4].

Topinambour tubers can be used as a dietary supplement to correct metabolic disorders in the complex treatment of patients with diabetes. This is due to the carbohydrate composition of topinambour tubers, which is represented by polysaccharide inulin. From an industrial point of view, topinambour tubers are a valuable raw material for production of inulin, fructose, molasses and alcohol [5]. Linxi Yang and others consider topinambour tubers as a source of functional food, bioactive compounds, biofuels and chemicals [4].

Due to its good nutritious and taste qualities, tubers of topinambour can be used as a food raw material [6, 7]. The disadvantage of topinambour is deterioration of its quality during storage by methods traditionally used for tuber raw materials: in refrigerated warehouses, piles and trenches [8]. During storage, there is a gradual shrinkage of the material observed. Holding relative humidity at 95% and temperature 0-2  $^{0}$ C allows to store tubers for 4-5 months [9]. Currently, there are a number investigations devoted to drying of topinambour [10-14].

Dnishev T.M., Tanayev N.B., Vitavskaya A.V. and others developed a method [10] of obtaining sweet flour from the inulin-containing plant by drying Jerusalem artichoke tubers for 120 hours in air, crushing into form of cubes and grinding to a particle size of 20-25 microns. The initial moisture content of Jerusalem artichoke tubers is about 75%. The temperature of the dried material with residual moisture of 9.6% is 60  $^{\circ}$ C.

Paker S.K. and Pirogova G.A. [11] the Jerusalem artichoke, previously subjected to operations of washing, sorting and inspection, dried with blowing of coolant at speed  $(5\div15)$  m/s and irradiated with infrared rays with the length of the beam  $(0,7\div2,5)$  micron. In this case, drying is carried out in two stages: the first stage – at a temperature not higher than 78 °C for 20 minutes to 6 h; the second one – at temperature not higher than 62 °C to final humidity  $(2\div12)$  %.

Zelenkov V.N. [12] the dried concentrate of Jerusalem artichoke offers to obtain by crushing tubers and their blowing by coolant with a temperature not above 70  $^{0}$ C.

Kochnev N. K. and others developed a method of production of powder from tubers of Jerusalem artichoke, which consists of preparation of tubers, crushing to puree state, heating the puree till temperature  $(80\div90)$  <sup>0</sup>C, cooling till  $(35\div55)$  <sup>0</sup>C, fermentolysis and atmospheric drying at temperature  $(55\div65)$  <sup>0</sup>C to final moisture content  $(6\div12)$  %. The target product is obtained by re-grinding the dried material [13].

Junko Takeuchi and Toshio Nagashima found that the dried tubers of Jerusalem artichoke have significant water binding capacity [14].

Nowadays there is a wide range of scientific papers devoted to study of methods of dehydration of thermolabile materials in a rarefied medium. Despite the achievements in this area, vacuum and vacuum-sublimation drying methods are not widely used in industry due to increased energy consumption.

## Materials and research methods

The object of research was Helianthus Tuberosus or Jerusalem artichoke tubers. Jerusalem artichoke tubers before drying were cut in the form of a cube with a face size 5 mm. Initial moisture content of Jerusalem artichoke tubers was  $(75\div77)$ %, the final one – 9%. Atmospheric drying was carried out at temperature of condensation of the refrigerant 47 <sup>o</sup>C and variations in its boiling point from -4 to -15 <sup>o</sup>C. The average temperature of air heated by condensation heat of refrigerant was 36, 38 and 40 <sup>o</sup>C. The height of bulk layer of dispersed material ranged from 10 to 40 mm. Air is pumped in the device for final atmospheric drying with speed (0,25÷0,4) m/s.

In order to solve this problem, a method of vacuum-atmospheric drying of large-dispersed materials is proposed, which includes vacuum drying of material to intermediate humidity and atmospheric drying to final one. At the same time atmospheric drying of the material is carried out by waste heat of condensation of refrigerating agent circulating in refrigerating machine which in the scheme of drying plant as a heat pump. Drying the material with air heated from this source saves energy costs for atmospheric drying.

In order to study the nature of processes of vacuum and atmospheric drying of large-dispersed thermolabile materials, experimental studies were conducted on a vacuum-atmospheric dryer. In the vacuum chamber, tests were carried out at medium pressures 4, 6 and 8 kPa and heater temperatures 35, 46 and 60  $^{\circ}$ C.

#### **Research results and discussion**

At studying rates of the process it was found that at the same intensity of vacuum and atmospheric drying the nature of drying curves has a similar appearance (figure 1). Thus, in selection of vacuum and atmospheric drying modes with same intensity of dehydration in both cases, it is possible to achieve such a course of drying process as would occur in vacuum or atmospheric drying. In this case, duration of vacuum-atmospheric drying process should be equal to duration of vacuum or atmospheric drying process.

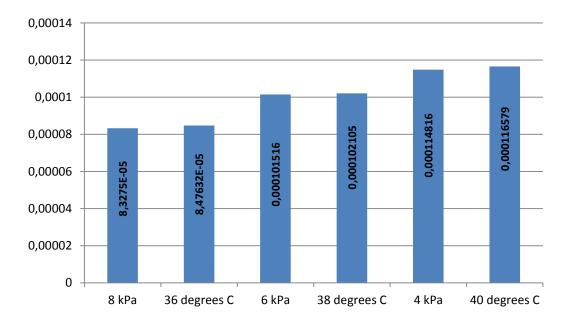


Figure 1. Comparison of intensities of vacuum and atmospheric drying of Jerusalem artichoke: abscissa axis - drying mode (medium pressure (vacuum drying), air temperature (atmospheric drying)); ordinate axis - drying intensity, kg/(m<sup>2</sup>s)

Figure 1 shows the comparison of dehydration intensities of vacuum and atmospheric drying of Jerusalem artichoke tubers. As can be seen from the figure, conditions of vacuum drying of Jerusalem artichoke crushed into the shape of a cube with temperature electric heaters  $60^{\circ}$ C and pressures of environment 8, 6 and 4 kPa most closely correspond to modes of atmospheric drying with air velocity 0.4 m/s and air temperatures 36, 38 and 40  $^{\circ}$ C, respectively.

However, the full compliance of vacuum and atmospheric drying modes must be confirmed by proximity of the material temperature values during the constant rate of above drying methods. In other words, the material dried to critical humidity in vacuum chamber must have a temperature close to temperature of material that is dehydrated to critical humidity in device for atmospheric heat drying. Otherwise, even with same intensity of vacuum and atmospheric drying processes, a lower or higher temperature of material can lead to a slowdown in drying process.

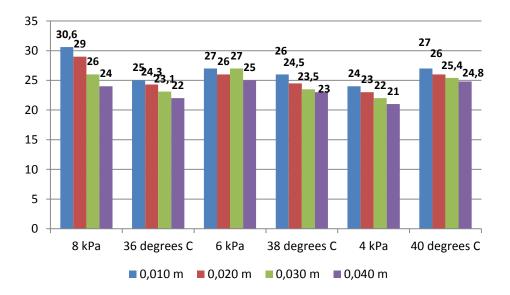


Figure 2. Comparison of material temperatures during period of constant rate of vacuum and atmospheric drying of Jerusalem artichoke with different layer heights: abscissa axis - drying mode (medium pressure (vacuum drying), air temperature (atmospheric drying)); ordinate axis – material temperature, <sup>0</sup>C.

Figure 2 shows the comparison of material temperatures during constant rate period of vacuum and atmospheric drying. Analyzing figure 2, it can be concluded that to requirements of compliance with temperatures of material are not suitable modes of vacuum drying at a pressure of medium 8 kPa and atmospheric drying at air temperature 36  $^{0}$ C.

## Conclusions

It should be noted that according to results of experiments, the dried product, Jerusalem artichoke, obtained by vacuum-atmospheric drying, by its sensory characteristics is not worse to the product dehydrated by vacuum drying.

Thus, using the above described methodology for development of process of vacuumatmospheric drying of large-dispersed materials, it is possible to reduce energy consumption of the process at maintaining intensity of drying.

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