

REDUCING THE HEAT STRESS ON MAIZE DURING SPRING SEASON BY USING SOME BIO STIMULANTS

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ABSTRACT

This study was aimed to investigate the effect of bio stimulators (chitosan and Appetizer) in reducing the heat stress on maize during spring season .Field experiment was conducted at the experimental farm, Coll. of Agricultural Engineering Sciences, University of Baghdad, Al-Jadiriya, Iraq, during the seasons of 2020 and 2021. The experiment was laid out in Randomized Complete Blocks Design within split plot arrangement, with three replicates. Where the main plots included two cultivars of maize (Baghdad3 and cultivar 5018), while the sub plots included five levels of chitosan and Appetizer:(soaking seeds with Chitosan at a concentration of 100 and 500 mg L⁻¹, and spraying with Chitosan 100 mg L⁻¹, and spraying Appetizer in two and three stages in addition to the two control treatments, which are soaking seeds with distilled water and dry seeds. The results showed that the cultivar 5018 was superioered in grain yield 11.82 and 11.08 Mg ha⁻¹ in both seasons. The treatment of spraying Chitosan gave the highest grain yield 13.26 and 12.94 Mg ha⁻¹, spraying the Appetizer in two stages gave 13.32 and 12.54 Mg ha⁻¹, and spraying Appetizer in three stages gave 13.70 and 12.97 Mg ha⁻¹ for both seasons. The interaction was significant in most of the studied traits. So we recommend the use of Chitosan spray at a concentration of 100 mg L⁻¹ and Appetizer spray at a concentration of 1.25 ml L⁻¹ in two or three stages to reduce the effect of heat stress on maize planted in the spring season to increase grain yield.

Key words: maize, Chitosan, Appetizer, grain yield, cultivars, climate change

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تقليل الاجهاد الحراري على الذرة الصفراء في العروة الربيعية باستخدام بعض المحفزات الحيوية

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باحث

قسم المحاصيل الحقلية - كلية علوم الهندسة الزراعية /جامعة بغداد

وزارة الزراعة - دائرة التخطيط والمتابعة

المستخلص

هدفت هذه الدراسة إلى معرفة تأثير المحفزات الحيوية (الكايوتوسان والابيتايزر) في تقليل الإجهاد الحراري على الذرة الصفراء خلال العروة الربيعية. أجريت التجربة في حقل تجارب كلية علوم الهندسة الزراعية - جامعة بغداد - الجادرية ، العراق ، خلال الموسمين الربيعيين 2020 و 2021. نفذت التجربة بتصميم القطاعات العشوائية الكاملة وترتيب الالواح المنشقة بثلاثة مكررات. تضمنت الالواح الرئيسية صنفين من الذرة الصفراء (بغداد 3 والصنف 5018)، بينما تضمنت الالواح الثانوية خمسة مستويات من الكايوتوسان والابيتايزر وهي (نقع البذور بالكايوتوسان بتركيز 100 و 500 ملغم لتر⁻¹ والرش السورقي بالكايوتوسان 100 ملغم لتر⁻¹ ورش الابيتايزر بمرحلتين وثلاث مراحل فضلاً عن معالمتي المقارنة وهي نقع البذور بالماء المقطر والبذور الجافة (بدون نقع او رش). أظهرت النتائج تفوق الصنف 5018 في حاصل الحبوب الذي بلغ 11.82 و 11.08 ميكاغرام هكتار⁻¹ في كلا الموسمين. اعطت معاملة رش الكايوتوسان اعلى حاصل حبوب 13.26 و 12.94 ميكاغرام هكتار⁻¹، واعطى رش الابيتايزر بمرحلتين 13.32 و 12.54 ميكاغرام هكتار⁻¹ واعطت معاملة رش الابيتايزر بثلاث مراحل 13.70 و 12.97 ميكاغرام هكتار⁻¹ لكلا الموسمين . كان التداخل معنوياً في معظم الصفات المدروسة. نوصي بأستعمال رش الكايوتوسان بتركيز 100 ملغم لتر⁻¹ ورش الابيتايزر بتركيز 1.25 مل لتر⁻¹ بمرحلتين او ثلاث مراحل لتقليل اثر الاجهاد الحراري الذي تتعرض له الذرة الصفراء المزروعة في العروة الربيعية لزيادة حاصل الحبوب.

الكلمات المفتاحية: ذرة صفراء ، الكايوتوسان، الابيتايزر، حاصل حبوب، الاصناف، تغير مناخي

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INTRODUCTION

Maize (*Zea mays* L.) is an important cereal crop for its various nutritional and industrial uses (9, 10). It is grown in Iraq two seasons : during fall season, which was dominant and covered for most of the areas allocated for this crop (11, 12), in addition to the appropriate climatic conditions for it, and spring season, which is still did not prevalent, and is cultivated in very close areas due to its low grain yield compared to the yield of the fall (19). The production of maize in the spring and fall seasons in Iraq at 2021 season was 374,400 tons, spring maize production was estimated about 0.3 tons, or 0.1% of the total production of the spring and fall maize, while the production of the fall maize was estimated at 374,397 tons, or 99.9% from the total production (23) In Iraq varieties were differ in grain yield and its components due to their different response to temperatures at the flowering stages, fertilization stages and accumulation of dry matter during the period of grain filling, as well as the difference in their efficiency in transferring the products of the photosynthesis process from the source to the sink (8, 13). Maintaining highest productivity of crops plants under heat stress is one of the important agricultural aims (16), the reduce of the harmful effects of high temperatures during pollination and fertilization stage of maize in the spring season by using a bio stimulant Chitosan,. Effective to stimulate plant growth and increase production and plant tolerance to oxidative stress, as it works to reduce the activity of reactive oxygen species (ROS) through its role in neutralizing reactive oxygen species, as it has been proven to have protective antioxidant properties. From a large number of available amines and hydroxyl groups that react with free radicals (21). As well as its role in increasing water availability by improving the absorption of water and essential nutrients and modulating cellular osmotic pressure (27). Chitosan also has a great ability to chelate elements such as iron, zinc, manganese and magnesium, which makes it an important element for plants. It is a good natural source of nitrogen and amino acids. Chitosan spraying on maize in three stages had a significant effect on increasing the number of

ears per plant, ear length and width, number of grains per ear, weight of 100 grains, grain yield per plant, and harvest index (21). Bio stimulants of marine algae extracts are agricultural inputs, when applied to plants, improved plant growth and productivity (6), regulate various physiological processes and improve plants' ability to adapt to abiotic stresses (20, 33). The bio stimulant increases the plant's ability to withstand biotic and abiotic stress by regulating the plant's content of plant growth regulators such as auxins and gibberellins (1). Appetizer is a bio stimulant extracted from marine algae *Ascophyllum nodosum*, which is characterized by the basic substance (GA Active 42), which has a key role in the absorption of nutrients by the plant in addition to its ability to withstand stress, in addition to containing microelements such as manganese and zinc. Spraying the bio stimulant of the extract of brown seaweed *Ascophyllum nodosum* on maize with three concentrations of 3, 6 ,9 ml L⁻¹ had a significant effects on grain yield and its components, and the maximum response at the concentration was 6 mg L⁻¹, recording the highest mean ear length, number of grains in the ear ,grains in the ear ⁻¹, and the yield of plant (17). This study was aimed to know the productivity and quality of maize in spring season through the use of biostimulants (Chitosan and Appetizer) to reduce the negative impact of heat stress on pollen vitality, pollination process and fertilization.

MATERIALS AND METHODS

A field experiment was carried out at the experimental farm of the College of Agricultural Engineering Sciences- University of Baghdad / Al-Jadriya,Iraq during the spring seasons of the 2020 and 2021. The experiment was carried in split plots arrangement ,according to the RCBD and with three replicates, spraying Chitosan at a concentration of 100 mg L⁻¹ with the same concentration in three stages, the first stage 6 leaves. at the beginning of the appearance of tasseling and silking, the second stage 12 leaves and the third stage in the tasseling stage, and two levels of Appetizer spray at a concentration of 1.25 ml L⁻¹ (According to the manufacturer's recommendation), which is to spray Appetizer in two stages with the same

concentration: the first stage is the stage of tasseling, the second stage is a week after the appearance of the silks. Appetizer spray with the same concentration in three stages: - the first stage, the 6 leaves stage, the second stage, the stage of the stage of tasseling and the third stage a week after the appearance of the silk, in addition to the two control treatments, which is soaking the seeds with distilled water for the same soaking period (10 hours), dry seeds without soaking and without spraying. The number of experimental units becomes 14 with an area of 5.7 m² (3 m x 2.5 m) containing four rows, the distance between them is 75 cm, and the distance between one plant and another is 20 cm, so that the plant density becomes 66,666 plants ha⁻¹. The seeds were planted at 16/3/2020 and 16/ 3/ 2021. The fertilization process was carried out with DAP fertilizer (46% P₂O₅ and 18% N) at a rate of 240 kg ha⁻¹ and potassium sulfate fertilizer (52% K₂O) at a rate of 200 kg ha⁻¹ added at planting, and urea fertilizer(46% N) at a rate of 360 ha⁻¹ was added by the first two batches a month after the date of planting. The second is at the beginning of the stage of tasseling (27). After preparing the Chitosan at the required concentrations, the seeds were soaked in it at room temperature 25C. The Chitosan and Appetizer spraying process was carried out according to the mentioned stages and the concentrations used in the morning using a 16-liter dorsal sprayer and a moving physical barrier was used when spraying.

Studied traits: When the plants reached the stage of full maturity, five plants were harvested randomly from the two middle lines for each experimental unit, the mean of which was calculated for all the characteristics: fertility percentage (%) It was calculated by dividing the number of ear grains by the number of total ovaries in the ear (seed and seedless sites) multiplied by 100 (14), number of ears in plant(ear plant⁻¹), number of rows in the main ear (row ear⁻¹), number of grains in a row for the main ear (grain row⁻¹), weight 500 grain (g) ,grain yield (Mg ha⁻¹), based on

15.5% .Using analysis of variance, and the means compared by (LSD) 5% level (32).

RESULTS AND DISCUSSION

Fertility percentage (%): The results in Table 1 show significant effects of cultivars, Chitosan and Appetizer treatments, and interaction on the fertility percentage in both seasons. Cultivar Baghdad 3, gave the highest mean fertility percentage , reached 93.26% and 92.39% for both seasons, respectively. This is a reflection of the different response of varieties to temperatures during flowering, stages fertilization and accumulation of dry matter during the period of grain filling. This is in agreement with what was found by Siddiq and Muhammad (29) who obtained significant differences in the fertility percentage between the synthetic cultivars of maize. The treatment of Chitosan spray at a concentration of 100 mg L⁻¹ was superior and gave the highest mean fertility percentage reached 98.28% and 97.64% with an increase of 13.66% and 14.65% compared to the control treatment for both seasons respectively. There were significant increases in the effective fertility percentage when the seeds were soaked with distilled water, (4.93% and 4.26%) compared to the control treatment in both seasons respectively. This could be due to the role of Chitosan and Appetizer foliar treatments in maintaining the antioxidant enzyme systems (unpublished data), which enhances the flow of sugar and starch towards the developing ovaries at the pollination stage when maize plants were treated with Chitosan and Appetizer before the stage of tasseling, which significantly reduced the percentage of Abortion of maize ovaries under heat stress conditions (34). The interaction between the two factors of the study in the first season, The combination of Chitosan spraying at a concentration of 100 mg L⁻¹ for Baghdad 3 cultivar was superior in this trait (98.63%). In the second season, the combination of Chitosan spraying at a concentration of 100 mg L⁻¹ was superior to cultivar 5018, which gave the highest mean of 97.66%.

Table 1. Effect of cultivars, Chitosan and Appetizer on fertility percentage (%)of spring maize for the two growing seasons 2020 and 2021

Treatments	Spring Season 2020			Spring Season 2021		
	Baghdad3	5018	Mean	Baghdad3	5018	Mean
Control	87.00	85.94	86.47	85.17	85.15	85.16
seeds Soaking in distilled water	90.55	90.92	90.73	89.82	87.75	88.79
seeds Soaking in Chitosan 100 mg L ⁻¹	91.99	88.33	90.16	90.88	88.19	89.54
seeds Soaking in Chitosan 500 mg L ⁻¹	90.09	87.14	88.62	89.31	86.87	88.09
Spray Chitosan 100 mg L ⁻¹	98.63	97.94	98.28	97.63	97.66	97.64
Spray the Appetizer in two stages	98.00	97.94	97.97	97.31	97.07	97.19
Spray the Appetizer in three stages	96.53	96.83	96.68	96.62	97.57	97.10
LSD 5%	1.468		1.038	1.199		0.848
Mean	93.26	92.15		92.39	91.46	
LSD 5%	0.801			0.613		

Number of ears in plant (ear plant⁻¹)

The results in Table 2 show a significant effect of Chitosan and Appetizer treatments and the interaction between the two factors on the number of plant ear in both seasons, while the cultivars had significant effects in the first season only. Baghdad 3 cultivar was superior and gave the highest mean number of ears of 1.52 ears plant⁻¹. The reason for these differences between the genotypes in this trait could be due to the fact that most of the genotypes differed in the number of ears of plant, as the number of ears is determined since the early stages of crop growth, which is correlated with the appearance of initiators (28). This is consistent with what was found by Al-Temimi and Al-Hilfy (4) and Ali (5), they obtained significant differences in the number of ears per plant among the synthetic cultivars of maize. Appetizer spraying in three stages was superior and gave 1.77 ear plant⁻¹

, with no differ significantly with the treatment of Appetizer spray in two stages and Chitosan spray at a concentration of 100 mg L⁻¹, which gave the same mean of 1.73 ear plant⁻¹. In the second season, the treatment of Appetizer spraying in two stages was superior as it gave 1.92 ear plant⁻¹. There was a significant increase in the number of ears per plant when the seeds were soaked with distilled water, amounting to 17.70% and 15.04% compared to the control treatment in both seasons respectively. The increase in the number of ears when applying Chitosan and Appetizer treatments could be a result of the plants getting enough of their nutritional needs because they have a good source of nutrients .as well as its role in increasing the absorption of major elements NPK, which stimulated the growth of the secondary ear and the completion of its arrival to a full ear is included in the yield of the plant.

Table 2. Effect of cultivars, Chitosan and Appetizer on the number of ears in plant (ear plant⁻¹) of spring maize for the two growing seasons 2020 and 2021

Treatments	Spring Season 2020			Spring Season 2021		
	Baghdad3	5018	Mean	Baghdad3	5018	Mean
Control	1.20	1.07	1.13	1.40	1.27	1.33
seeds Soaking in distilled water	1.27	1.40	1.33	1.47	1.60	1.53
seeds Soaking in Chitosan 100 mg L ⁻¹	1.47	1.33	1.40	1.70	1.60	1.65
seeds Soaking in Chitosan 500 mg L ⁻¹	1.27	1.27	1.27	1.47	1.40	1.43
Spray Chitosan 100 mg L ⁻¹	1.87	1.60	1.73	1.87	1.93	1.90
Spray the Appetizer in two stages	1.87	1.60	1.73	1.93	1.90	1.92
Spray the Appetizer in three stages	1.73	1.80	1.77	1.87	1.73	1.80
LSD 5%	0.162		0.114	0.122		0.087
Mean	1.52	1.44		1.67	1.63	
LSD 5%	0.071			n.s		

As for the interaction between the two factors of the study,. In the first season, the two combinations of two-stage Appetizer spraying and Chitosan spraying at a concentration of 100 mg L⁻¹ for Baghdad 3 cultivar were superior and gave the highest mean number of ears, (1.87 ear plant⁻¹) In the second season, the combination of Chitosan spray at a concentration of 100 mg L⁻¹ for cultivar 5018 and the two-stage Appetizer spray treatment for Baghdad 3 had the highest mean number of ear which was 1.93 ear of plant⁻¹.

Number of rows in the main ear(ear row⁻¹)
The results of Table 3 show that there is a significant effects of cultivars, Chitosan and Appetizer, and the interaction between the two factors on the number of rows in the ear in both seasons. Cultivar 5018, which gave the highest mean number of rows per ear, was 17.60 and 17.50, row ear⁻¹ in both seasons

respectively. This could be due to the fact that this trait is governed by a large number of gene pairs, which differed in the nature of their genetic action from one class to another, as environmental factors indirectly affect the preparation of the emerging classes since the early stages of the plant's life through their influence on the general activity of the plant in division, growth , representation and transfer the products of photosynthesis to the downstream of the grain. As well as being affected by climate factors (temperature and humidity), especially on pollination, fertilization and the success of grain formation (7). This is consistent with what was found by Al-Temimi and Al-Hilfy (4) and Ali (5), who obtained significant differences in the number of rows in the main ear among the synthetic varieties of maize.

Table 3. Effect of cultivars, Chitosan and Appetizer on the number of rows in the main ear (row ear⁻¹) of spring maize for the two growing seasons 2020 and 2021

Treatments	Spring Season 2020			Spring Season 2021		
	Baghdad3	5018	Mean	Baghdad3	5018	Mean
Control	14.80	15.87	15.33	14.73	15.70	15.22
seeds Soaking in distilled water	16.13	16.93	16.53	16.03	16.80	16.42
seeds Soaking in Chitosan 100 mg L ⁻¹	15.07	16.80	15.93	15.67	17.13	16.40
seeds Soaking in Chitosan 500 mg L ⁻¹	15.20	16.13	15.67	15.47	16.03	15.75
Spray Chitosan 100 mg L ⁻¹	18.93	18.93	18.93	18.73	19.17	18.95
Spray the Appetizer in two stages	18.00	19.20	18.60	18.00	19.00	18.50
Spray the Appetizer in three stages	18.67	19.33	19.00	18.47	18.63	18.55
LSD 5%	0.560		0.396	0.709		0.465
Mean	16.69	17.60		16.73	17.50	
LSD 5%	0.650			0.714		

In the first season, Appetizer spray in three stages, which gave the highest mean of 19.00 row ear⁻¹. There was a significant increases in the number of rows in the main ear when soaking the seeds with water amounted to 7.83% and 7.88% compared to the control treatment in both seasons respectively. Although this trait has a high heritability, the lack of material represented negatively affects it as a result of the failure to develop complete flowers of one or more rows in the ear (15). Increasing the activity of the antioxidant enzymes and the vitality of pollen grains (unpublished data) and it could be that the increase in the leaf area is led to an increase in the process of carbonic metabolism and then the representations resulting from the increase of these components may have contributed effectively to the development of the flowers constituting these rows. As for the interaction between the two study factors in the first season. The combination of the three-stage Appetizer spray treatment was superioered to cultivar 5018, which gave the highest mean of 19.33 row ear⁻¹. In the second season. the combination of Chitosan spraying at a concentration of 100 mg L⁻¹ was superioered to cultivar 5018, which gave the highest mean of 19.17 row ear⁻¹.

Number of row grains in the main ear (grains row⁻¹): The results in Table 4 show that there are significant effects of Chitosan and Appetizer treatments and the interaction between the two factors in the number of rows

grains in the main ear in both seasons, while the cultivars had significant effects in the second season only. Baghdad 3 cultivar was superioered in the second season, which gave the highest mean, which amounted to 42.34 grains row⁻¹. This could be due to the presence of genetic differences between the cultivars, as each genetic structure has a genetic ability to produce a certain number of grains per row and it varies from one type to another (7). This is consistent with what was found by Mahmoud (22) and Ali (5). In the first season, the Appetizer spraying treatment with three stages, which gave the highest mean (45.23 grains row⁻¹). In the second season, the Chitosan spraying treatment with a concentration of 100 mg L⁻¹ was superioered, as it gave the highest mean of 44.63 grains row⁻¹. There were significant increases in the number of grains row in the main ear when soaking the seeds with distilled water amounted to 6.09% and 4.13% compared to the control treatment, which gave the lowest mean of 38.43 and 38.03 grains row⁻¹ in both seasons respectively. The decreases in the number of grains per row when compared to treatment would be attributed to the high temperatures during the flowering stage, which negatively affected the contract percentage through its effect on the degree of growth and development of anthers, low pollen vitality, drying stigmas, and the lack of opportunities for pollen to be fixed on them the rate of fertilization decreases, which negatively

affects the number of grains formed in the row (3). While the number of grains in the row increased when applying the Chitosan and Appetizer treatments as a result of the role of these treatments in increasing the number of rows in the ear (Table 4). The number of grains in each row is determined after determining the number of rows in the ear,

which develop into mature grains after fertilization (7). Such as plant height, leaf area and index, number of leaves, chlorophyll content, as well as the increases in soil nutrients uptake, pollen vitality, (unpublished data) and Increasing the fertility percentage (Table 1) due to the application of Chitosan and Appetizer treatments.

Table 4. Effect of cultivars, Chitosan and Appetizer on the number of (grains of a row in the main ear (grain row⁻¹) of spring maize for the two growing seasons 2020 and 2021

Treatments	Spring Season 2020			Spring Season 2021		
	Baghdad3	5018	Mean	Baghdad3	5018	Mean
Control	38.93	37.93	38.43	38.87	37.20	38.03
seeds Soaking in distilled water	41.40	40.13	40.77	39.80	39.40	39.60
seeds Soaking in Chitosan 100 mg L ⁻¹	43.00	40.33	41.67	41.87	41.67	41.77
seeds Soaking in Chitosan 500 mg L ⁻¹	42.53	41.93	42.23	41.40	42.00	41.70
Spray Chitosan 100 mg L ⁻¹	46.00	43.47	44.73	45.07	44.20	44.63
Spray the Appetizer in two stages	44.67	45.33	45.00	44.40	43.53	43.97
Spray the Appetizer in three stages	46.33	44.13	45.23	45.00	43.73	44.37
LSD 5%	1.208		0.854	0.707		0.500
Mean	43.27	41.90		42.34	41.68	
LSD 5%	n.s			0.179		

The interaction between the two study factors in the first season. The combination of Appetizer spraying with three stages for the Baghdad 3 cultivar gave the highest mean of 46.33 grains row⁻¹. In the second season, the combination of Chitosan spraying at a concentration of 100 mg L⁻¹ for Baghdad3 cultivar was superior in giving it the highest mean of 45.07 grains row⁻¹.

Number of grains in the main ear (grains ear⁻¹): The results of Table 5 indicate that there are non-significant effects for the cultivars with significant effects of Chitosan and Appetizer treatments on the number of grains in the main ear in both seasons, while the interaction was significant in the first season only. In the first season, the Appetizer spraying treatment with three stages, which gave the highest mean of 859.47 grains ear⁻¹.

In the second season, the treatment of Chitosan spray at a concentration of 100 mg L⁻¹ was superior, as it gave the highest mean of 845.75 grains ear⁻¹. There were significant increases in the number of grains in the main ear when the seeds were soaked with water, amounting to 14.47% and 12.37% in both seasons respectively. compared to the control that gave the lowest mean of 589.48 and 578.70 grains ear⁻¹. Due to the abortion of grains because heat stress, which is related to a lack of supply of representations to the growing grains, as the reason for the abortion of grains is the decrease in the activity of enzymes related to the synthesis of starch and sugar, and the decrease in starch levels, as well as that heat stress caused a decrease in the expression of genes Associated with starch synthesis (25).

Table 5. Effect of cultivars, Chitosan and Appetizer on the number of grains in the main ear (grain ear⁻¹) of spring maize for the two growing seasons 2020 and 2021

Treatments	Spring Season 2020			Spring Season 2021		
	Baghdad3	5018	Mean	Baghdad3	5018	Mean
Control	576.85	602.11	589.48	573.10	584.30	578.70
seeds Soaking in distilled water	668.96	680.56	674.76	638.26	662.31	650.28
seeds Soaking in Chitosan 100 mg L ⁻¹	648.40	678.37	663.39	656.68	714.33	685.51
seeds Soaking in Chitosan 500 mg L ⁻¹	646.96	677.33	662.15	641.39	673.61	657.50
Spray Chitosan 100 mg L ⁻¹	871.15	824.00	847.57	844.30	847.19	845.75
Spray the Appetizer in two stages	804.53	871.20	837.87	799.36	827.67	813.51
Spray the Appetizer in three stages	865.49	853.44	859.47	831.13	815.00	823.07
LSD 5%	31.729		22.436	n.s		23.325
Mean	726.05	741.00		712.03	732.06	
LSD 5%	n.s			n.s		

The increases in the number of ear grains when spraying with Appetizer could be attributed to the important role of plant hormones that enter into the formation of the bio-stimulator of marine algae extract Appetizer in regulating the distribution of manufactured materials between the source and the sink, especially at the start of the formation and appearance of grains sitting, as well as its role in encouraging root growth and increases the efficiency of water absorption and nutrients (17), and the reason for the increases in the number of main ear grains could be due to the increases in the number of ear rows and the number of grains in a row (Tables 2 and 3) in addition to the role of Chitosan and Appetizer spraying treatments in increasing the fertility percentage (Table 1), which were reflected in the increases in the number of grains in the main ear, and this is consistent with what was found by Mondal et al. (24) they obtained significant increases in the number of grains in the ear when spraying Chitosan. As for the interaction, it was significant in the first season only. Two-stage Appetizer spray treatment for cultivar 5018, giving it the highest mean of 871.20 grains ear⁻¹.

Weight of 500 grain (g)

The results of Table 6 indicate that there are non-significant effects for the cultivars, while the effects of Chitosan and Appetizer

treatments and the interaction between the two factors were significant in the weight of 500 grains in both seasons. The weight of 500 grains decreased more when Chitosan and Appetizer were treated. The control which gave the highest mean of 132.12 and 132.37g. As the greater the number of grains, the lower their weight due to the increased competition for photosynthesis products between grains. In the first season, the two treatments of soaking seeds with Chitosan at a concentration of 100 and 500 mg L⁻¹ did not differ between them significantly, they gave means of 128.97 and 129.57g for both concentrations respectively. They also did not differed significantly with the treatment of seed soaking with water. The lowest mean weight of 500 grains was when Chitosan spraying at a concentration of 100 mg L⁻¹ gave an mean of 118.58 g. In the second season, two Chitosan seed soaking treatments at a concentration of 100 and 500 mg L⁻¹ gave means of 131.39 and 126.65 g for both consecutive concentrations. The Appetizer spraying treatment in three stages gave the lowest mean weight of 500 grains amounted to 118.21 g and did not differed significantly with the two treatments of Appetizer spray in two stages and Chitosan spray at a concentration of 100 mg L⁻¹, which gave 119.33 and 120.26g for both treatments respectively.

Table 6. Effect of cultivars, Chitosan and Appetizer on the weight of 500 grain (g) of spring maize for the two growing seasons 2020 and 2021

Treatments	Spring Season 2020			Spring Season 2021		
	Baghdad3	5018	Mean	Baghdad3	5018	Mean
Control	132.19	132.06	132.12	132.96	131.78	132.37
seeds Soaking in distilled water	131.51	130.11	130.81	131.30	132.32	131.81
seeds Soaking in Chitosan 100 mg L ⁻¹	127.00	130.93	128.97	131.10	131.69	131.39
seeds Soaking in Chitosan 500 mg L ⁻¹	127.11	132.02	129.57	121.12	132.18	126.65
Spray Chitosan 100 mg L ⁻¹	117.04	120.12	118.58	120.46	120.07	120.26
Spray the Appetizer in two stages	116.00	121.37	118.68	119.98	118.67	119.33
Spray the Appetizer in three stages	119.09	122.02	120.55	116.78	119.64	118.21
LSD 5%	3.35		2.187	4.568		3.230
Mean	124.28	126.95		124.82	126.62	
LSD 5%	n.s			n.s		

The reason for the decreases in grains weight when treated with Chitosan and Appetizer could be attributed to the fact that these treatments gave the highest means in the number of rows in the ear, the number of grains in a row and the number of grains in the ear (Tables 3, 4, 5), which were reflected in increases in the number of grains in the ear and increases in competition for building outputs Photosynthesis between the grains, which led to a decrease in their weight. As for the interaction between the two factors of this study, in the first season. The control combination of Baghdad 3 cultivar was superioered by giving it the highest mean weight of 500 grains that reached 132.19 g. In the second season. The control combination was superioered to Baghdad 3, which gave the highest mean weight of 500 grains, which reached 132.96 g.

Grain yield (Mg ha⁻¹)

The results of Table 7 reveal that there are significant effects of cultivars, Chitosan and Appetizer treatments, and interaction between factors on grains yield in both seasons. Cultivar 5018, which gave the highest mean grain yield of 11.82 and 11.08 Mg ha⁻¹, was superioered in both seasons, respectively. The grain yield per unit area varies from one structure to another according to the different stages of growth and combinations of genes it

possesses or the physiological processes that regulate the evolutionary stages of the cultivar or the genetic structure (21 , 34). This is consistent with what was found by Al-Temimi and Al-Hilfy (4) and Ali (5), they obtained significant differences in grain yield per unit area among the structural varieties of yellow maize. As for the Chitosan and Appetizer treatments, it is found that the Appetizer spray treatment with three stages was superioered by giving it the highest grains yield of 13.70 and 12.97 Mg ha⁻¹ with an increase of 55.15% and 59.14% compared to the control for both seasons respectively. In the second season. The increase in the grain yield when spraying Appetizer could be attributed to its content of nutrients, growth regulators and vitamins that enter into plant nutrition and improving growth characteristics and yield, which was reflected in the transfer of nutrients from the source (represented by the leaf) to the sink (represented by the grain). There were significant increases on grain yield when soaking the seeds with distilled water amounted to 4.42% and 7.85% in both seasons respectively, compared to the control that gave the lowest mean of 8.83 and 8.15 Mg ha⁻¹. The decreases in grain yield at control could be attributed to heat stress that led to many negative effects including These include reduced pollen viability, increased grain

abortion rate, decreased rate of cell division in storage tissues (endosperm), decreased starch synthesis, reduced downstream capacity of grain development, faster grain development, and reduced activities of enzymes responsible for determining the number of grains per row under heat stress (2 , 26). The increases in grain yield may also be attributed to the role of Chitosan and Appetizer in improving plant growth by increasing growth characteristics, macronutrients, pollen vitality and crop growth rate (unpublished data). and the fertility

percentage (Table 1), which was reflected in the increase in the number of active ear in the plant and the number of grains in the ear (Tables 2 , 5) and was reflected in the increase in the grain yield When spraying Chitosan. The interaction between the two factors, the combination of the Appetizer at three-stages was superior with cultivar 5018 (14.20 Mg ha⁻¹) In the second season. The combination of Chitosan spraying at a concentration of 100 mg L⁻¹ for cultivar 5018 gave the highest mean of 13.40 Mg ha⁻¹

Table 7. Effect of cultivars, Chitosan and Appetizer on grain yield (Mg ha⁻¹) of spring maize for the 2020 and 2021 growing seasons

Treatments	Spring Season 2020			Spring Season 2021		
	Baghdad3	5018	Mean	Baghdad3	5018	Mean
Control	8.59	9.06	8.83	7.86	8.43	8.15
Soak seeds in distilled water	9.34	9.11	9.22	8.67	8.91	8.79
Soak seeds in Chitosan 100 mg L ⁻¹	10.17	11.76	10.97	10.58	10.75	10.67
Soak seeds in Chitosan 500 mg L ⁻¹	9.74	11.49	10.62	10.64	10.22	10.43
Spray Chitosan 100 mg L ⁻¹	13.02	13.49	13.26	12.49	13.40	12.94
Spray the Appetizer in two stages	13.05	13.60	13.32	12.51	12.58	12.54
Spray the Appetizer in three stages	13.19	14.20	13.70	12.66	13.28	12.97
LSD 5%	0.329		0.233	0.266		0.188
Mean	11.01	11.82		10.77	11.08	
LSD 5%	0.174			0.048		

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