

Effect Of Spraying With Glutathione , Citric Acid And Appetizer On Some Growth Traits Of Two Wheat Varieties

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ABSTRACT

A field experiment was conducted during the winter seasons of 2022-2023 and 2023-2024 in the experimental fields of the Department of Field Crops - College of Agricultural Engineering Sciences, University of Baghdad - Al-Jadriya, with the aim of knowing the response of some growth traits of two wheat varieties under the effect of spraying with the antioxidant glutathione, organic citric acid and biostimulant appetizer. The experiment was applied according to the randomized complete block design RCBD and in the arrangement of split plots and with three replicates. It included two factors, the main plots included two wheat varieties, namely Buhooth 22 and Mawaddah. The secondary plots included spraying glutathione at a concentration of 1000 and 1500 mg L⁻¹, citric acid at a concentration of 75 and 150 mg L⁻¹, and appetizer at a concentration of 1.25 and 1.50 ml L⁻¹, in addition to the control treatment (without spraying). Spraying was done in two stages, the first at the beginning of tillering stage and the second at the heading stage. The results showed that the cultivar Buhooth 22 was superior in flag leaf area with averages of 44.48 and 44.58 cm², for the two study seasons respectively. As for the spray treatments, the Appetizer treatment at a concentration of 1.50 ml L⁻¹ was superior in plant height (109.31 and 103.20 cm), flag leaf area cm² (46.91 in the first season), dry weight of the plant (1142.4 and 1054.9 g m⁻²), and crop growth rate from elongation to flowering (37.42 and 40.40 g m⁻²). As for the treatment of spraying glutathione at a concentration of 1500 mg L⁻¹, it excelled with the highest average number of tillers, reaching 513.2 tillers m⁻².

Keywords: Growth traits, Glutathione, Citric acid, biostimulant .

INTRODUCTION

Given the economic and nutritional importance of wheat crops, as it is the leading grain crop in Iraq and the world and is considered one of the strategic and essential crops for achieving food security, it is necessary to intensify efforts to increase the efficiency of the production of this crop, which is still below the required level compared to the countries of the world. This comes from

several reasons, including the lack of adoption of sustainable agricultural techniques in the field of crop management and modern field practices, including the lack of use of good varieties with a high ability to exploit the available growth resources[1]. Knowing the performance and response of each variety to growth factors and modern field practices is one of the important matters for increasing production[2]. One of the modern and environmentally

safe methods is the use of antioxidants, including glutathione. It is a tripeptide consisting of three amino acids (glutamic, cysteine and glycine) and is responsible for the balance between oxidation and antioxidants and protects against free radicals in addition to regulating the work of enzymes[3] and has a role in metabolic processes, cell division and differentiation, flower development in plants and other functions[4]. Citric acid is one of the basic compounds that play an effective and influential role in the formation and production of active plant compounds, as it contributes to building the plant cell and forming its compounds such as fats, proteins and carbohydrates that the plant produces in its growth stages from germination, emergence and growth until reaching the grain yield, In addition to its role in the formation of chlorophyll, phytochromes and cytochromes[5]. Biostimulants are natural and environmentally friendly methods that have the ability (when added in small quantities) to enhance vegetative growth, mineral nutrient absorption, and plant response to different climatic conditions[6] [7]. Appetizer is one of these stimulants and is mainly composed of seaweed *Ascophyllum nodosum*, which is a rich source of many biologically active phenolic compounds such as phlorotannins. It is also rich in plant hormones such as cytokinins, auxins, and gibberellins, in addition to carbohydrates, proteins, and other compounds[8] [9]. In addition, Appetizer contains trace elements such as manganese and zinc. Appetizer is a physiological stimulant that plays a fundamental role in activating nutrient absorption by the plant and thus

increasing production. Due to the importance of the above, this study was conducted with the aim of knowing the response of growth characteristics of two wheat varieties to spraying with glutathione, citric acid and the biofertilizer Appetizer.

MATERIALS AND METHODS

A field experiment was carried out in the fields of the College of Agricultural Engineering Sciences - University of Baghdad - Al-Jadriya during the winter seasons 2022-2023 and 2023-2024 with the aim of knowing the response of some growth traits of two wheat varieties (Mawaddah and Buhooth 22) under the effect of spraying with the antioxidant glutathione, organic citric acid and the biostimulant Appetizer according to the design of complete randomized blocks RCBD and in the arrangement of split plot and with three replicates. The experimental land was prepared by plowing the land twice perpendicularly using a rotary plow, and the soil was smoothed with a rotary plow (Rotovater) then leveled and divided into three replicates with 14 experimental units for each replicate with dimensions of 2x2 m. The experimental unit includes 10 lines with a length of 2 m, the distance between one line and another is 20 cm, The experimental land was planted on 2022-2023 for the first season and 2023-2024 for the second season, with a seed quantity of 120 kg ha⁻¹. The experimental land was fertilized with urea fertilizer (46% N) at a rate of 162 kg N ha⁻¹ in three equal batches for all experimental units, the first at the tillering stage (ZGS: 21), the second at the beginning of stem elongation (ZGS: 32), and the third batch at the heading

stage (ZGS: 40). DAP fertilizer (46% P₂O₅) was added at a rate of 100 kg ha⁻¹ in one batch before the smoothing process[10]. The experiment included two factors, the first factor (the main one) included two varieties of wheat, Mawaddah and Buhooth 22, while the second factor included spraying glutathione, citric acid and appetizer was applied separately, at concentrations of 1000 and 1500 mg L⁻¹ for glutathione, 75 and 150 mg L⁻¹ for citric acid and 1.25 and 1.50 ml L⁻¹ for the biofertilizer appetizer, in addition to the control treatment (without spraying glutathione, citric acid and appetizer). Spraying was done in two stages, the first at the beginning of tillering stage ZGS: 21 and the second at the heading stage 59: ZGS.

The Studied Traits

1.Plant Height (cm)

2.Flag Leaf Area (cm²)

The flag leaf area was measured according to the following equation:

$$\text{Area of flag leaf (cm}^2\text{)} = \text{Length of flag leaf} \times \text{Width at the middle} \times 0.95$$

3.Total Number of Tillers m⁻²

The total number of tillers at the harvest stage was calculated from the harvested area (0.3 m²) for each experimental unit and the results were then converted to square meters.

4.Dry Weight of the Plant (g m⁻²)

The dry weight was measured at the elongation stage by taking a sample from a distance of 30 cm in length from each experimental unit and cutting the roots to the crown area, then the sample was dried in an electric oven at 65 °C until

the weight was stable and weighed with a sensitive balance, then the results were converted to square meters.

5.Crop Growth Rate (g m⁻² day⁻¹)

According to the period from the planting stage to the plant entering the elongation stage (ZGS: 32 - ZGS: 69). According to the following equation [11]:

$$\text{CGR} = 1/A \text{ W}_2 - \text{W}_1 / \text{T}_2 - \text{T}_1$$

RESULTS AND DISCUSSION

Plant Height (cm):

The results in Table 1 indicate that the wheat varieties (Buhooth 22 and Mawaddah) did not differ significantly in plant height for both seasons. However, the spraying treatments of the materials were significant in both seasons, as the spraying of Appetizer at a concentration of 1.50 ml L⁻¹ was superior in plant height by giving the highest average of 109.31 and 103.20 cm for the two seasons respectively, compared to the comparison treatment (without spraying), which gave the lowest average of 94.78 and 86.20 cm respectively, with an increase rate of 15.33 and 19.72%, and did not differ significantly from the treatment of spraying citric acid at a concentration of 150 mg L⁻¹ and the treatment of spraying glutathione at a concentration of 1500 mg L⁻¹. In the second season, it did not differ significantly from the citric acid spray treatment at a concentration of 150 mg L⁻¹, which gave 100.95 cm. The reason for the superiority of Appetizer may be attributed to the positive role of seaweed extracts in activating auxin synthesis inside the plant by activating certain enzymes that

affect a series of cell divisions below the apical, leading to an increase in plant height. The results are consistent with what [12] reached.

The effect of the interaction was significant in the first season, as the cultivar Buhooth 22, which was sprayed with the Appetizer treatment at a concentration of 1.50 ml L⁻¹, achieved

the highest average of 110.99 cm, followed by the cultivar Mawaddah when sprayed with citric acid at a concentration of 150 mg L⁻¹, with a non-significant difference of 109.62 cm. As for the second season, the interaction between the two study factors was not significant.

Table 1. Effect of varieties, concentrations of glutathione, citric acid, appetizer and their interaction on plant height (cm) for the seasons 2022-2023 and 2023-2024.

Treatment	Season 2022-2023							
	Glutathione mg L ⁻¹	Appetizer ml L ⁻¹		citric acid mg L ⁻¹				
variety	0	1000	1500	1.25	1.50	75	150	Mean
Buhooth22	96.43	104.20	105.07	105.91	110.99	102.41	104.88	104.27
Mawaddah	93.12	103.32	108.91	103.11	107.64	105.16	109.62	104.41
L.S.D 0.05				4.91				N.s
Mean	94.78	103.76	106.99	104.51	109.31	103.79	107.25	
L.S.D 0.05				3.49				
Season 2023-2024								
Buhooth22	89.54	95.54	98.91	97.93	102.33	97.03	102.10	97.63
Mawaddah	82.86	93.78	97.12	99.82	99.57	92.44	104.30	95.70
L.S.D 0.05				N.S				N.S
Mean	86.20	94.66	98.02	98.87	103.20	94.73	100.95	
L.S.D 0.05				3.61				

Flag Leaf Area (cm²):

The results of Table 2 indicate that the Buhooth 22 variety gave the highest average flag leaf area of 44.48 and 44.58 cm² compared to the Mawaddah variety which gave the lowest average of 41.47 and 42.75 cm² for the two seasons respectively. This difference between the varieties in flag leaf area is due to the genetic difference between these varieties in this trait, and this is consistent with what [13], [14][15] and [16] reached, who indicated that the flag leaf area differs according to the varieties.

The results in Table 2 indicated that spraying Appetizer at a concentration of 1.50 ml L⁻¹ gave the highest average flag leaf area of 46.91 and 48.96 cm² compared to the comparison treatment which gave the lowest average of 36.71 and 36.10 cm² with an increase rate of 27.7 and 35.62% for the two seasons respectively, followed by the citric treatment at a concentration of 150 mg L⁻¹ with an increase rate of 21.22% in the first season and the Appetizer treatment at a concentration of 1.25 ml L⁻¹ in the second season which gave 46.40 cm² with an insignificant difference from the glutathione treatment

at a concentration of 1500 mg L⁻¹ which gave 45.37 cm² with an increase rate of 28.53 and 25.67% for the two treatments respectively.

This may be attributed to the content of the seaweed extract (the main component of the appetizer) on high levels of cytokinins, auxins, amino acids and a number of major and minor elements that stimulate cell division and expansion, in addition to increasing the ability of the roots to absorb water and dissolved elements in it. This result is consistent with what was reached by [17] and [12], who indicated that the area of the flag leaf differs according to the different concentrations of spraying the seaweed extract.

As for the interaction between the two factors, which was significant, we note from the same table the difference in the

response of the trait in the two seasons to spraying with these materials for the two cultivars, as the area of the flag leaf increased with increasing spraying concentrations of appetizer and citric acid in the first season, but when increasing the concentration of glutathione to 1500, this trait decreased, but this decrease was not significant, while for the cultivar Mawaddah, this trait increased with increasing spraying concentrations of all these materials. As for the second season, the area of the flag leaf increased for the two cultivars with increasing concentrations of glutathione, appetizer and citric acid, with the exception of the cultivar Buhooth 22, in which this trait decreased with increasing the concentration of citric acid to 150 mg L⁻¹, a significant decrease.

Table 2. Effect of varieties, concentrations of glutathione, citric acid, appetizer and their interaction on flag leaf area (cm²) for the seasons 2022-2023 and 2023-2024.

Treatment	Season 2022-2023							
	Glutathione mg L ⁻¹	Appetizer ml L ⁻¹		citric acid mg L ⁻¹				
variety	0	1000	1500	1.25	1.50	75	150	Mean
Buhooth22	35.94	45.42	44.30	45.00	50.21	43.71	46.76	44.48
Mawaddah	37.47	40.64	43.56	43.86	43.61	40.83	42.25	41.47
L.S.D 0.05				2.46				2.71
Mean	36.71	43.03	43.93	44.43	46.91	42.27	44.50	
L.S.D 0.05				1.53				
Season 2023-2024								
Buhooth22	36.85	45.61	47.19	48.36	50.14	44.52	39.37	44.58
Mawaddah	35.35	38.77	43.55	44.44	47.78	42.48	46.86	42.75
L.S.D 0.05				1.79				0.57
Mean	36.10	43.12	43.50	48.96	46.40	45.37	42.19	
L.S.D 0.05				1.35				

Number of Tillers m⁻²:

The results shown in Table 3 indicate that the Mawaddah variety is superior by giving the highest average of 497.6 and

473.5 tillers m⁻² for both seasons compared to the Buhooth 22 variety, which gave the lowest average of 448.5 and 441.1 for both seasons, respectively,

with an increase rate of 10.94 and 7.34%. The difference in the number of tillers between varieties may be due to the genetic nature of each variety and the determinant of the ability to branch, in addition to the response of each variety to environmental conditions. This is consistent with what was reached by [18] and [14], who indicated that the varieties differ significantly in the number of tillers characteristic.

The results of the same table showed significant differences between the spray treatments and their concentrations, as the glutathione spray treatment at a concentration of 1500 mg L⁻¹ in the first season gave the highest average of 513.2 tillers m⁻² compared to the comparison treatment which gave the lowest average of 436.1 and an increase rate of 17.67%. As for the second season, the appetizer spray treatment at a concentration of 1.50 ml L⁻¹ outperformed by giving an average of 487.3 tillers m⁻² which did not differ significantly from the glutathione spray treatment at a concentration of 1500 mg L⁻¹ which gave 481.5 tillers m⁻² compared to the comparison treatment which gave 419.9 and an increase rate of 16.05 and 14.67% for the two treatments respectively. The reason for the increase may be due to the fact that glutathione is

an antioxidant that plays a role in cell division and elongation and plant growth and development and it is agreed upon. This is in line with what [19] concluded, who indicated that spraying wheat plants with different concentrations of glutathione led to an increase in the number of tillers.

The same table indicates the difference in the response of the number of tillers for the two varieties with the difference in the concentrations of spraying materials. We notice that the number of tillers increased in the variety Mawaddah with the increase in the concentrations of spraying glutathione and appetizer. However, when the concentration of spraying citric acid increased to 150 mg L⁻¹, the number of tillers decreased significantly in the first season. As for the variety Buhooth 22, the increase in this trait continued with the increase in the concentrations of spraying materials. As for the second season, the opposite happened, as the number of tillers decreased with the increase in the concentrations of spraying citric acid to 150 mg L⁻¹. As for the Mawaddah variety, the increase in this trait continued with the increase in the concentrations of spraying all materials.

Table 3. Effect of varieties, concentrations of glutathione, citric acid, appetizer and their interaction on the number of tillers (m⁻²) for the seasons 2022-2023 and 2023-2024.

Treatment	Season 2022-2023							
	variety	Glutathione mg L ⁻¹	Appetizer ml L ⁻¹	citric acid mg L ⁻¹				Mean
	0	1000	1500	1.25	1.50	75	150	
Buhooth22	428.8	437.8	467.6	438.9	466.7	443.3	456.8	448.5
Mawaddah	443.4	531.1	558.9	464.4	482.2	520.1	482.7	497.6
L.S.D 0.05				32.88				32.39
Mean	436.1	484.4	513.2	451.7	474.4	481.7	469.7	

L.S.D 0.05				21.80				
				Season 2023-2024				
Buhooth22	415.7	447.2	459.6	444.9	471.4	430.2	418.6	441.1
Mawaddah	424.1	441.7	503.3	489.6	503.1	461.9	490.6	473.5
L.S.D 0.05				27.42				
Mean	419.9	444.4	481.5	467.3	487.3	446.0	454.6	32.04
L.S.D 0.05				16.19				

consistent with what was reached by [12] for the presence of a significant effect of seaweed extract treatment on the dry weight of the wheat plant.

Dry Weight of the Plant at the Elongation Stage g m^{-2} :

The results of Table 4 show that the varieties did not differ significantly among themselves, while the spray treatments differed significantly, as the Appetizer treatment at a concentration of 1.50 ml L^{-1} achieved the highest average of 1142.4 g and 1054.9 m^{-2} compared to the comparison treatment, which gave 819.9 and 773.4 g m^{-2} , with an increase rate of 39.33 and 36.39% , for two season , The reason for the increase in dry weight when treated with appetizer may be due to its role in increasing the rate of photosynthesis due to its containing marine algae, thus increasing the number of tillers, increasing the area of the flag leaf, and the dry weight in the elongation stage. These results are

As for the interaction between the two factors, we note from the same table that the dry weight at elongation increased with the increase in the spray concentrations of all materials for both varieties, but the maximum difference between the two varieties in this trait in the first season was when spraying citric acid at a concentration of 150 mg L^{-1} , reaching 92.7 g m^{-2} , as the average at this concentration and the Mawaddah variety reached 1032.1 g m^{-1} , while the lowest dry weight was recorded in the control treatment and the same variety (Mawaddah), reaching 772.3 g m^{-1} . As for the second season, spraying citric acid at a concentration of 75 mg L^{-1} recorded the maximum difference of 40.4 g m^{-2} for the Mawaddah variety, as the average reached 946.6 g m^{-1}

Table 4. Effect of varieties, concentrations of glutathione, citric acid, appetizer and their interaction on dry weight (g m^{-2}) at elongation for the 2022-2023 and 2023-2024 seasons.

Treatment		Season 2022-2023						
		Glutathione mg L ⁻¹		Appetizer ml L ⁻¹		citric acid mg L ⁻¹		
variety	0	1000	1500	1.25	1.50	75	150	Mean
Buhooth22	867.5	909.0	1062.2	996.0	1131.2	922.1	939.4	975.3
Mawaddah	772.3	883.8	1091.1	959.7	1153.6	905.8	1032.1	971.2

L.S.D 0.05				76.34				N.S
Mean	819.9	896.4	1076.6	977.8	1142.4	913.9	985.7	
L.S.D 0.05				45.41				
			Season 2023-2024					
Buhooth22	759.1	877.2	1061.3	924.6	1055.4	906.2	1005.3	941.3
Mawaddah	787.8	872.1	1015.8	886.9	1054.4	946.6	1033.9	942.5
L.S.D 0.05				69.92				N.S
Mean	773.4	874.6	1038.5	905.7	1054.9	926.4	1019.6	
L.S.D 0.05				23.62				

Crop Growth Rate $\text{gm}^{-2} \text{day}^{-1}$ for the Stage from Elongation Stage to the flowering 100% :

It is noted from Table 5 that the varieties did not differ significantly among themselves in both seasons, while the spray treatments differed, as the Appetizer treatment at a concentration of 1.50 ml L^{-1} had the highest growth rate at this stage, reaching 37.42 and $40.40 \text{ g m}^{-2} \text{ day}^{-1}$ for both seasons, compared to the control treatment, which recorded the lowest growth rate, reaching 23.71 and $24.86 \text{ g m}^{-2} \text{ day}^{-1}$, with an insignificant difference from the Glutathione spray treatment at a concentration of 1500 mg L^{-1} , 36.62 and $40.02 \text{ g m}^{-2} \text{ day}^{-1}$, and the Appetizer treatment at a concentration of 1.25 ml L^{-1} for the first season, $37.31 \text{ g m}^{-2} \text{ day}^{-1}$. This is attributed to the fact that bio stimulants improve chlorophyll synthesis because they contain multiple growth regulators and hormones, thus leading to increased plant growth, as in plant height, flag leaf area, and dry weight, Tables 1, 2 and 4 respectively. As for the interaction between the two study factors, the Buhooth 22 variety outperformed when sprayed with glutathione at a concentration of 1500 mg L^{-1} with the highest growth rate of $39.88 \text{ g m}^{-2} \text{ day}^{-1}$, which did not differ significantly from the treatment of spraying with glutathione at a

concentration of 1000 mg L^{-1} , the treatment of spraying Appetizer at concentrations of 1.25 and 1.50 ml L^{-1} , and the treatment of spraying citric acid at a concentration of 75 mg L^{-1} in the first season. As for the second season, the treatment of Appetizer at a concentration of 1.50 ml L^{-1} outperformed the Mawaddah variety with the highest crop growth rate with an average of $41.43 \text{ g m}^{-2} \text{ day}^{-1}$ compared to the lowest average for the Mawaddah variety with the control treatment, which recorded 20.81 and $22.55 \text{ g m}^{-2} \text{ day}^{-1}$.

Table 5. Effect of varieties, concentrations of glutathione, citric acid, appetizer and their interaction on crop growth rate g m⁻² day⁻¹ from elongation stage to the flowering 100% for the seasons 2022-2023 and 2023-2024.

Treatment		Season 2022-2023						Mean
Variety	0	Glutathione mg L ⁻¹		Appetizer ml L ⁻¹		citric acid mg L ⁻¹		
		1000	1500	1.25	1.50	75	150	
Buhooth22	26.61	36.07	39.88	39.06	38.79	35.53	32.11	35.44
Mawaddah	20.81	31.73	33.37	35.55	36.05	28.92	33.34	31.39
L.S.D 0.05				4.958				N.S
Mean	23.71	33.90	36.62	37.31	37.42	32.22	32.73	
L.S.D 0.05				2.523				
		Season 2023-2024						Mean
Variety	0	Glutathione mg L ⁻¹		Appetizer ml L ⁻¹		citric acid mg L ⁻¹		
		1000	1500	1.25	1.50	75	150	
Buhooth22	27.18	31.57	39.18	37.31	39.37	34.58	35.34	34.93
Mawaddah	22.55	30.32	40.85	37.19	41.43	34.44	35.24	34.57
L.S.D 0.05				3.073				N.S
Mean	24.86	30.95	40.02	37.25	40.40	34.51	35.29	
L.S.D 0.05				1.827				

CONCLUSIONS

We conclude that wheat varieties varied in some growth characteristics and that spraying biostimulant appetizer at a concentration of

1.50 ml L⁻¹ had a positive effect in improving growth traits.

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