

Effect of the growth regulator Brassinolide on the Some plant hormones content of maize Zea mays. L seedlings affected by water stress

Kadum Mohammed Abdullah

Horticulture and landscape Department, College of Agriculture, University of Kerbala , Karbala , Iraq

Corresponding author E-mail: kadum.m@uokerbala.edu.iq

Received:	Abstract:
Octo. 18, 2021	A laboratory trail was carried out in a lab of the Seeds Examina- tion and Certification in Babylon. Governorate., according to a com- plete random design (CRD) with four replicates. The trail consisted
Accepted:	of three levels of water stress by adding PEG 6000 polyethylene gly-
Accepted: Nove. 22, 2021 Published: Des. 01, 2021	of three levels of water stress by adding PEG 6000 polyethylene gly- col (0, -6 and -9) bar and symbolized by S0., S1., and S2., while the other factor included three concentrations to stimulate the seeds by soaking them in the growth regulator Brassinolide for 24 hours (0, 2, and 4) mg. L-1 and symbolized by B0, B1, and B2. The outcomes revealed the negative significant impact of water stress in reducing content of maize seedlings hormones, as the level of water shortage at S2. gave less average of gibberellin content, indole-acetic acid and cytokinin, while it caused an increase in abscess content. On the other hand, the Brassinolide treatment produced a remarkable boost, as B2 dose recorded the best mean of gibberellin, indole-acetic acid and cytokinin content, and the abscisic acid content decreased com- pared to the control treatment without addition. It can be concluded from this study is the possibility of increasing the germination effi- ciency of corn crop under conditions of water stress by using plant hormones, including Brassinolide for its role in increasing plant hor-
	mones responsible for embryo development and germination. Key words: PEG 6000, Brassinolide, Growth regulators, Seedlings of maize.

Introduction :-

Yellow corn is *Zea mays*. L is one of the important crops that occupies the third place after wheat and rice, and is an essential component of animal nutrition, as well as for various other manufacturing purposes [1]. The utility of growth promoters to boost plant toleration to stress during seed germination is useful technique because it is easy to apply, cheap and less hazard, as well as its major importance through plant cycle by its role in adjusting growth such as boosting and germination lateness, flowering and ripening phase and has a role in plant response to environmental stresses [2]. The outcomes of the trail by [3] revealed that the exposure maize plants to water stress resulted a remarkable reduction in the leaves content of auxin, gibberellin and



cytokinin. Brassinolide growth regulator is one of the steroidal compounds, which is a natural growth hormone in plants, and it includes a wide range of components that produce a vital impact on botanical operations inside the plant by adjusting some ingredients related to growth and boosting plants, and it is classified within the sixth group of plant regulators [4, 5]. Brassinolide is present in various constituent of plant (pollen, flowers, seeds, leaves and roots), and their amount is higher in young plant tissues than in mature tissues [6]. Recent studies indicated that the use of Brassinolide would give good results in increasing the plant's tolerance to water stress. Therefore, the research aims to know the role of Brassinolide growth regulator in regulating the hormonal content of maize seedlings, and thus define the function of this hormone to overcome water shortage.

Materials and methods:

Trail was carried out in a lab. of the Seeds Examination and Certification in Babylon Governorate for the year 2018, using a factorial experiment according to a complete random design (C.R.D.) and repeated 4 times. The experiment included three water levels stress by exposing maize seeds to PEG 6000 solutions of polyethylene glycol (0, -6 and -9) bar, and its symbols were S0, S1, and S2, while the other treatments included stimulating seeds germination by soaking them for 24 hours in three concentrations (0, 2 and 4) mg. L⁻¹ of the growth regulator Brassinolide, and its symbols are B0, B1, and B2. To study the simulation of drought in the lab., solutions of polyethylene glycol were prepared according to [7]. The required concentrations of Brassinolide (American production from Sigma company) were prepared. Completed and clean grains were selected, rinsed with plain water, and then with hypochloride solution at concentration of 0.01 %, for 3 minutes, and then the sterilized grains were rinsed many times with distilled water. Fifty grains were distributed on filter paper in plastic Petri dishes, then PEG 6000 mixture were applied in an amount that ensured complete coverage of the seeds in the dishes, under controlled germination conditions in accordance with the recommendations of the [8].

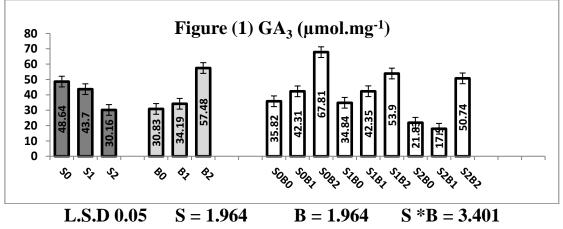
Determination of the plant hormones content of leaves (µmol. mg⁻¹ dry weight):

The quantitative determination of the plant hormones auxin (IAA), gibberellin (GA3), abscisic acid (ABA) and cytokinin (CK) was carried out according to the method used by [9]. Samples of dry ground leaves weighing 1 g were taken and extracted with 60 ml of (12 ml methanol, 5 ml chloroform and 3 ml ammonium hydroxide), then 25 ml of distilled water was added to this composition and the pH was adjusted to pH 2.5 by drop of Hydrochloric Acid (1N) and Sodium Hydroxide (1N), The combination was separated with 15 ml of Ethyl Acetate for the evaluation of (IAA, GA₃, and ABA) at wavelengths (263, 254, 280 nm) respectively, then the pH of the lower aqueous layer was regulated to 7 and the mixture was separated again with 15 ml of Ethyl Acetate to measure cytokinin (CK) on the wavelength 269 nm using a spectrophotometer.



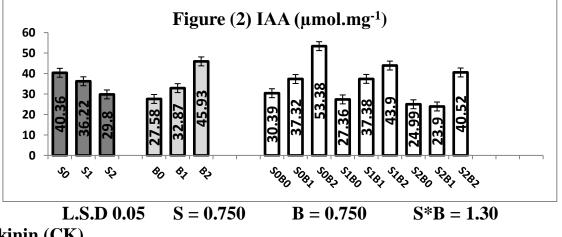
Results and discussion: Gibberellins (GA3).

The data at Figure (1) indicates that there are remarkable variations among water stress treatments, as the treatment of water stress S2 significantly affected the reduction of the content of leaves from the growth regulator gibberellins, and the lowest average was recorded at 30.16 μ mol. The figure also shows the significant influence of the Brassinolide treatment, as the B2 treatment recorded a higher average of 57.48 μ mol than to the control plates, which reached 30.83 μ mol. The combined treatments caused a reasonable increase in this property, as o S0 B2 treatment gave the highest mean of 67.84 μ mol.



Indole Acetic Acid (IAA):

Figure (2) shows the appreciable variations among water stress treatments, as the treatment of water stress S2 significantly affected the reduction of the content of leaves from indole acetate lion, so the lowest average was recorded at 29.80 μ mol. The figure also shows the significant impact of the Brassinolide treatment, as the B2 treatment recorded a higher average of 45.93 μ mol than the control seedlings, which produced 27.58 μ mol. The factor's overlap caused a significant increase in this trait, because S0 B2 produced more values of 53.38 μ mol.

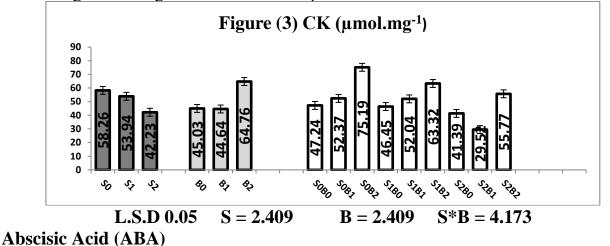


Cytokinin (CK)

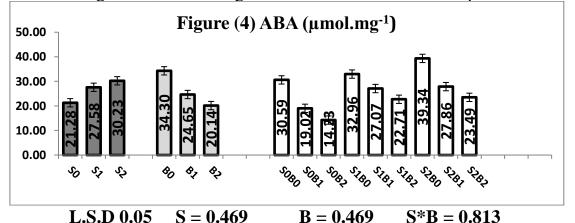
The values at Figure (3) shows the considerable variance caused by water stress exposure, as the treatment of water stress S2 significantly affected the reduction of



cytokinin content of leaves, so the lowest average was recorded at 42.23 μ mol. The figure also shows the moral effect of the Brassinolide treatment, as the B2 treatment recorded a higher value of 64.76 μ mol in relation to the control plants, where they reached 45.03 μ mol. The combined factors caused a remarkable boost in this property, as S0 B2 gave the highest mean of 75.19 μ mol.



The means represented at Figure (4) indicates the notable alterations resulted from water stress exposure, as the treatment of water stress S2 significantly affected the increase in the leaves content of abscisic acid, so it produced the highest rate of 30.23 μ mol in comparison with S0 where it recorded only 21.28 μ mol. Same figure also shows a clear effect of Brassinolide treatment reducing leaves' content of abscisic acid because B2 concentration revealed less rate of 20.14 μ mol in comparison with control plots, where it was only 34.29 μ mol. Combined treatments a prominent response in reducing ABA , as S0 B2 gave the lowest mean of 14.23 μ mol.



The effects of water stress were studied in the laboratory to simulate field conditions, as solutions of polyethylene glycol were added, to study the characteristics of wheat seedlings under water tension exposure. The reason could be referred to the most influential physiological impact of water shortage to reduce plant growth-regulating hormones, including auxins, gibberellins and cytokinins, which have a reflection in reducing plant growth processes [10]. Studies indicate that water stress significantly reduced the content of plant growth-promoting hormones (auxins, gibberellins, and cytokinins), while at the same time raising the content of ABA and ethylene [10, 11]. It



is also believed that water stress leads to a deficiency of the coenzyme Acetyl-CoA molecules required for Mevalonic acid to produce gibberellins [12].

The boosted values of gibberellin and indoleacetic acid (Figures 1 and 2) after treated with Brassinolide could be due to the role of Brassinolide in gene expression in the metabolism of Mevalonic acid, that is one of the pathways of hormone formation [13]. This result agrees with the findings of [14] on seedlings of wheat. Moreover, Brassinolide could enhance and protect cells against environmental stresses by providing defence and boosting growth by stimulating gene expression that reveals defence, regulation, antioxidant responses, and the output of H_2O_2 concentration, which is an outcome of NADPH oxidase activity [13].

Conclusion:

It can be concluded from this study that drought and water stress certainly have negative influence on the growth and income of different crops, including corn, from seedling to production by affecting plant hormones that regulate the life cycle of the crop. On the other hand, synthetic plant hormones (including Brassinolide) can be used to counteract the negative effects of drought and push the plant to better germination and optimum growth. The study recommends studying these combinations on other crops with more advanced stages of growth in order to adapt to the risk of water shortage facing the world and to maintain the adequacy and quality of production.

References:

- Molazem, D and A. Jafar. (2011). Proline reaction, peroxide activity and antioxidant enzymes in varieties of maize. (*Zea mays L.*) under different levels of salinity. Australian Journal of Basic and Applied Sciences. 5(10): 1248-1253.
- 2) Jaleel, C. A., P.Manivannan. A.Wahid. M. Farooq. H. J. ALJuburi. R. Somasundar, and R. Pannerersel. (2009). Drought stress in plants: A review on mrphological characteristics and pigments composition. J. Agric. Biol. 11: 100–105.
- 3) Ansary, M. H., H. A. Rahmani, M. R. Ardakani, F. Paknejad, D. Habibi and S. Mafakheri .(2012). Effect of Pseudomonas fluorescent on proline and phytohormonal status of maize (*Zea mays* L.) under water deficit stress. Annals of Biological Research, 3(2): 1054-1062.
- 4) Favero, D.S., K. N. Le and M. M. Neff. (2017). Brassinosteroid signaling converges with suppressor of photochromic B4-#3 to influence the expression of small auxin up RNA genes and hypocotyl growth. Plant Joranal Author Manuscript; available in PMC; 89(6): 1133–1145.
- 5) Manoli, A., S. Trevisan, S. Quaggiotti, and S. Veretto. (2018). Identification and characterization of the BZR transcription factor family and its expression in



response to abiotic stresses in *Zea mays* L. Plant growth regulation, 84(3): 423-436.

- 6) Kim, S.K, S.C. Chang, E.J. Lee, W.S. Chung, Y.S. Kim, S Hwang, J.S. Lee. (2010). Involvement of brassinosteroids in the gravitropic response of primary root of Maize. Plant Physiology, 123: 997-1004.
- **7) Michel BE, Kaufmann MR. (1973).** The osmotic potential of polyethylene glycol 6000. Plant Physio., 51: 914-917.
- 8) ISTA. International Rules for Seed Testing. (2008). International Seed Testing Association Chapter5: germination test. P.1-57.
- 9) Ŭnyayar, S. Topcuoğlu, Ş.F. and Ŭnyayar, A. (1996). A modified method for extraction and identification of indole-3-acetic acid (IAA), gibberellic acid (GA3), abscisic acid (ABA) and zeatin produced Phanoerochate chrysosporium ME446. Bulg. J. plant Physiol., 22 (3-4): 105-110.
- Aldesuquy, H. S. (2014). Glycine Betaine and Saliylic acid induced modification in water relation and productivity of drought Wheat plants. J. stress. Physio. Bioche., 10(2): 55-73.
- 11) Zhang, L.X., G.Mei, L.Shiqing, L. Shengxiu and L. Zongsuo. (2011). Moddulation of Plant Growth, Water Status and AntIioxidantive system of two Maize (*Zea mays* L.) cultivars induced by exogenous Glycinebetaine under long term mild drought stress. Pak. J. Bot., 43(3): 1587-1594.
- 12) Aroca, R. (2012). Plant Responses to Drought Stress from Morphological to Molecular Features. Springer, Heidelberg, Berlin. PP.466.
- 13) Naveen, N., Kumari, N., Avtar, R., Jattan, M., Ahlawat, S., Rani, B., ... & Singh, M. (2021). Evaluation of Effect of Brassinolide in Brassica juncea Leaves under Drought Stress in Field Conditions. Horticulturae, 7(11), 514.
- 14) Toman, S. S., Jasim, A. H., Kadhim, Z. K., Hassan, A. A. H., & Hamzah, R. M. (2019). Effect of barassinolide on growth characteristics of wheat (Triticum aestivum L.) under water stress. In IOP Conference Series: Earth and Environmental Science (Vol. 388, No. 1, p. 012045). IOP Publishing.