Effect of kaolin and water stress conditions on the physiological and anatomical properties of bread wheat

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Abstract

A field experiment was conducted to study effect of spraying kaolin on some physiological and anatomical characteristics of bread wheat, Mawada variety, growing during the winter season 2023-2024. Using a randomized complete block design (RCBD) and a split plot arrangement with three replicates. The study included four spray levels, namely 2, 4 and 6%, in addition to the comparison treatment, which is 0% (without spraying), as well as three levels of water stress, namely irrigation at the depletion of 40, 60 and 80% of the available water, which occupied the main plots, while the kaolin spray levels occupied the secondary plots. The results showed that the plants treated with 40% depletion of available water were superior in the flag leaf index of chlorophyll pigment (SPAD) and the relative water content in the flag leaf (%) and the number of stomata (mm2) and the length and width of the stomatal opening (micron) by an increase rate of (22.93, 6.88, 3.08, 18.03 and 23.43%) for the studied traits in succession, compared to the irrigation treatment when 80% of the available water was depleted. The spraying treatments with kaolin also showed a significant superiority when spraying with a concentration of 6% in the flag leaf index of chlorophyll pigment (SPAD) and the relative water content by an increase rate of (14.71 and 11.36%) in succession, compared to the control treatment (without spraying), a concentration of 4% in the flag leaf index of chlorophyll pigment (SPAD). The spraying treatments with kaolin for all concentrations did not differ significantly when The length and width of the stomatal opening were measured, compared to the control treatment, which gave the highest average for these two characteristics.

Keywords. wheat, water stress, kaolin, physiological properties, anatomical properties

Introduction

Because its grains provide food for almost 35% of the world's population, it plays a crucial role in ensuring food security in developing nations. [1]. Some climate changes, such as high temperatures, low rainfall, low water releases from the Tigris and Euphrates rivers in the Iraqi environment, monopolization of water resources, and policies produced by neighbouring countries to control water releases, in addition to the high level of water pollutants, including CO2, as well as the increased need to use water in agricultural and industrial ways, all of these things led to an increase in dry areas, especially in the central and southern regions. It was necessary to think of several ways to reduce and rationalize water use, including the use of modern irrigation methods, according to scientific research, including irrigation scheduling. [2,3] indicated that irrigation scheduling is one of the irrigation management methods that aim to avoid excessive water use. Therefore, the amount and need of the plant for water must be known in order to provide it, in addition to using materials that reduce the seasonal water consumption of the crop, including the use of anti-transpiration, including kaolin.

Kaolin is an inert clay mineral that is mainly composed of aluminum silicate and its is Al2Si2O5(OH)2.. It is also found in the form of regular deposits in igneous rocks that have been exposed to erosion [4]. It is used in the agricultural field to combat agricultural pests and protect crops [5]. However, technological progress has made it possible to produce kaolin granules of a specific size and shape while maintaining the property of reflection and sedimentation on the plant surface, which has opened the way for its use in many agricultural uses and the possibility of employing its multiple and environmentally friendly properties, including its use in reducing environmental stress, including reducing water tension on the plant by reducing transpiration from the plant surface. Recently, the technology of fine particles (Pft) has appeared, which is based on the formation of a microscopic layer of mineral granules. Kaolin has been introduced into this technology and has become known as the granular kaolin layer technology. Kaolin-based particle film technology, commercially known as Surround WP, allows gas exchange and does not cause stomata to close [6]. Its physiological effectiveness has been attributed to its possession of a set of properties, the most important of which is that it has inert particles with a diameter of >2 micrometers and a structure that allows diffusion and does not hinder gas exchange. It also reflects infrared and ultraviolet rays and allows other effective wavelengths to penetrate to reach the carbon assimilation sites [7].

this study aims to achieve the following objectives:

Estimating the appropriate seasonal water consumption for wheat crops, Reducing the amount of water consumption for wheat crops by spraying kaolin and Getting the best production by adding transpiration resistors.

Methods and Materials

1.1. Field Experimental

A winter-season field study (2023–2024) was carried out in the Sadat Al-Hindiyah District of Babylon Governorate, Iraq, at an experimental extension farm in the Al-Mahnawiyah area. This site, managed by the Babylon Extension Training Center, lies 8 km north of Babylon at coordinates 32.61°N latitude and 44.30°E longitude. The experiment employed a randomized complete block design (RCBD) with a split-plot arrangement and three replicates. Kaolin spray concentrations (0% control, 2%, 4%, and 6%) were assigned to subplots, while irrigation treatments based on water depletion levels (40%, 60%, and 80% of available water) occupied main plots.

The field, post-plowing and leveling, was divided into three blocks, each containing 12 experimental units. Main treatments and replicates were spaced 1.5 m apart to prevent water seepage, with 0.75 m between subplots. Each 4 m^2 experimental unit comprised eight 2-meter-long rows spaced 20 cm apart. Wheat seeds were sown on November 26, 2023, at a rate of 120 kg ha^{-1} (48 g per unit, 6 g per row), with a planting depth of 5 cm.

Fertilization and Management

Urea (46% N) was applied at 200 kg N ha^{-1} in three equal splits: at the three-leaf stage (Zadoks GS13), stem elongation (Zadoks GS32), and booting (Zadoks GS40). Triple superphosphate (46% P_2O_5) was incorporated pre-planting at 100 kg ha^{-1}. Weeds were manually removed, and crops were harvested at full maturity.

Soil Analysis Preparation

Pre-planting composite soil samples (0–30 cm depth) were collected from multiple randomized locations after removing the top 5 cm surface layer. Samples were air-dried, homogenized, sieved through a 2 mm mesh, and analysedUniversity of Baghdad / College of Agriculture, Department of Soil Analysis to determine physicochemical properties (Table 1).

Table 1 Some chemical and physical properties of field soil before planting

.No	Prosperity	Value		Unit
.1	(pH)	7.9		-
.2	EC	3		$ds.m^{-1}$
.3	O.M	0.57		$gm.kg^{-1}$
.4	Porosity	42		%
.5	Bulk density	1.39		$M\ gm.m^{-3}$
.6	K available	160		ppm
.7	p available	7.2		ppm
.8	available N	43.5		ppm
.9	Soil Particles	Sand	51.6	$gm.kg^{-1}$
		Silt	608.4	
		Clay	340	
.11	F.C	0.428		cm cm
.12	P.W.P	0.23		cm cm
.13	Aval. Water	0.198		cm cm

The soil retention capacity was estimated by estimating at tensions (33, 100, 500, 1000 and 1500) kPa, which was represented graphically in the soil moisture characterization curve (Figure 1).

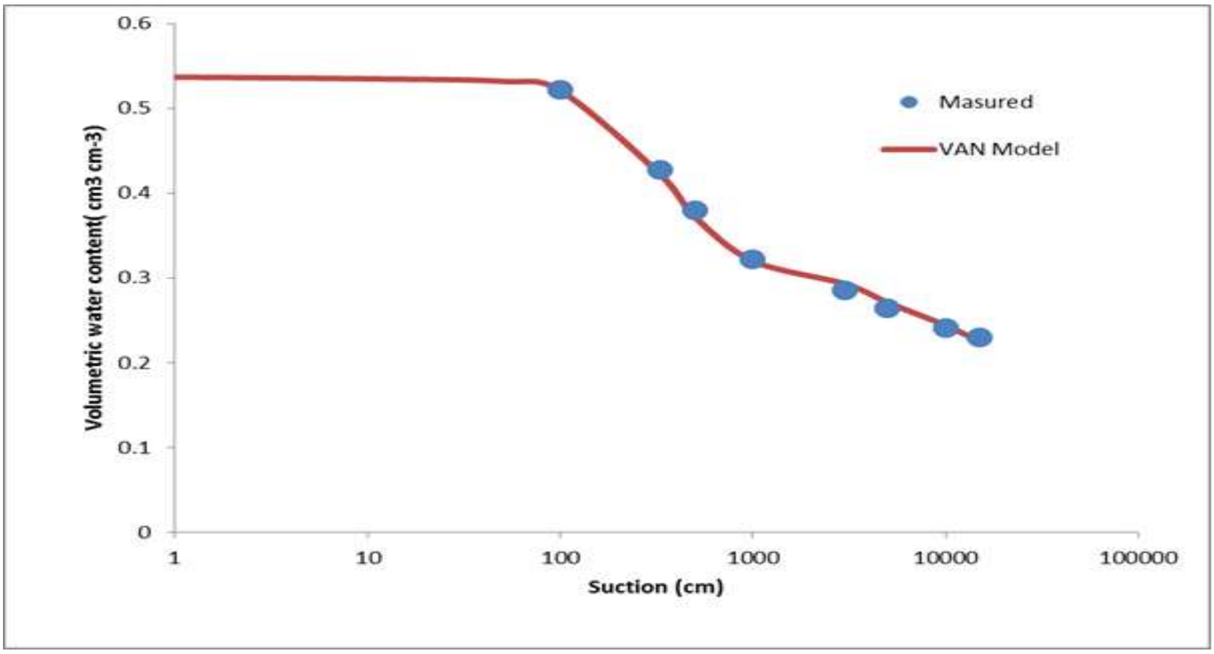


Figure 1 water content with tension soil used in the study

The following measurements were performed:

• Physiological characteristics:

- leaf Area index of chlorophyll pigment (SPAD):

- Relative water content in leaves (%):

• Anatomical characteristics:

- Number of stomata (mm^2):

- Length and width of stomatal opening (micron):

Statistical analysis:

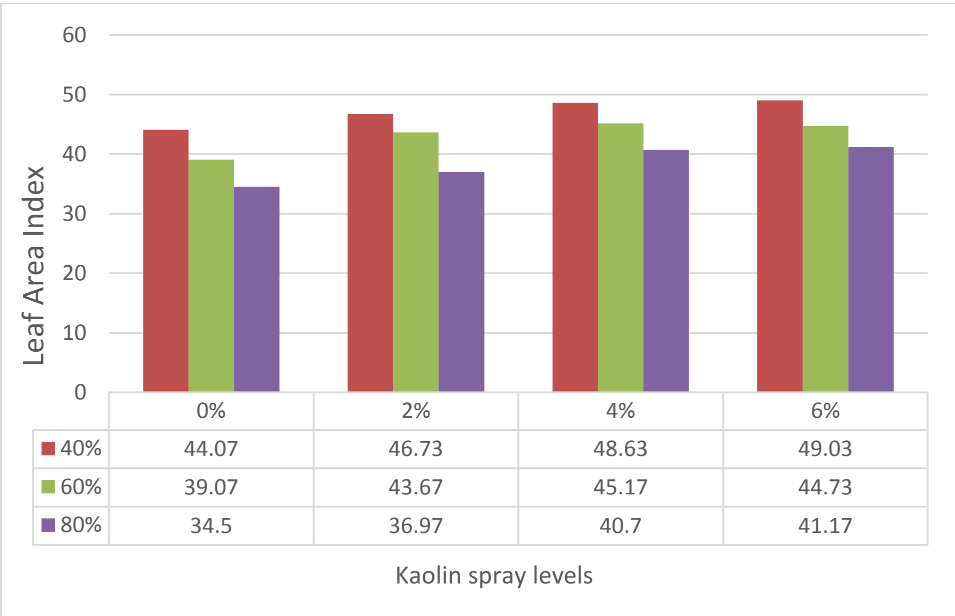
After collecting the data, the Available-made statistical program GenStat V.20 was used according to the method approved by (Al-Asadi, 2019), and the data are analyzed according to the ANOVA table, and the averages are compared according to the Least Significant Differences Test (LSD) at a probability level of 0.05 (Al-Rawi and Khalaf Allah, 2000).

Result and Discussion

3.1. Flag leaf index of chlorophyll pigment (SPAD):

The results in Figure (2) showed a significant effect of the depletion treatments, kaolin spraying and their interaction on the flag leaf index of chlorophyll pigment.

The results of the table showed the superiority of irrigation 40% of the Available water with an average of 47.21 (SPAD) compared to the irrigation treatment after depleting 80% of the Available water, which had the lowest average of 38.33 (SPAD), as the percentage of decrease reached 18.65%. The reason for the increase in the chlorophyll index when the irrigation treatment was depleted 40% of the Available water may be attributed to the efficiency of the roots in absorbing nitrogen, which is one of the components entering the pyroferin ring, which is one of the basic components of the pigment molecule [2]. Or in the water potential of the leaves and the relative water content (Table 3), which causes inhibition of the photosynthesis process as a result of reducing the openings of the stomata and the lack of CO_2 exchange, which affects the growth of the chloroplast membranes and reduces the concentration of pigments present in them, including chlorophyll pigment [10], especially when high temperatures accompany these conditions.



L.S.D=Depletion level 0.91 , Kaolin spray 0.61 , interction 1.15

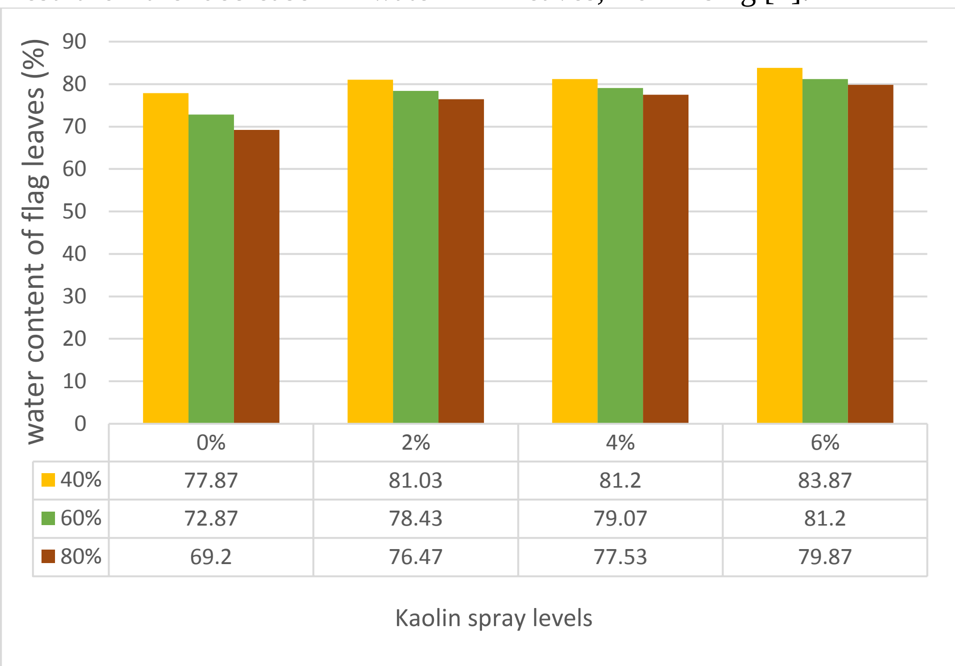
Figure 2. Effect of Available-to-use water depletion levels and kaolin on the science leaf index of chlorophyll (SPAD)

3.2. water content in flag leaves (%):

The results of Figure (3) showed a significant effect of depletion treatments and kaolin spraying on the relative water content in flag leaves and no significant effect of the interaction between them.

The results of Figure 3 showed the superiority of the irrigation treatment after depleting 40% of the Available water with an average of 80.99, compared to the irrigation treatment after depleting 80% of the Available water, which gave the lowest average for this trait of 75.77% and without a significant difference with the irrigation treatment after depleting 60% of the Available water. The reason for the decrease in the relative water content in flag leaves when the depletion level increases may be due to the decrease in water content in plant tissues as a result of the decrease in water potential in the soil, which led to a decrease in the plant's ability to absorb water, in addition to the loss of balance between absorption and transpiration, which negatively affected the water status of the plant [11].

The results of the table also showed that the concentration of kaolin 6% was superior, reaching 81.64%, , reaching 73.31%. Kaolin forms a thin white cover on the surface of the leaf that reflects the sunlight falling on it, thus reducing the temperature of the plant tissue, especially the leaves, which reduces the difference in vapor pressure between the plant tissues and the outside, thus reducing transpiration. As for paraffin wax, it forms a barrier that prevents direct water loss from the outer epidermis, as it reflects a large part of the incident sunlight, which prevents the temperature of the plant tissue, including the leaves, from rising [1].



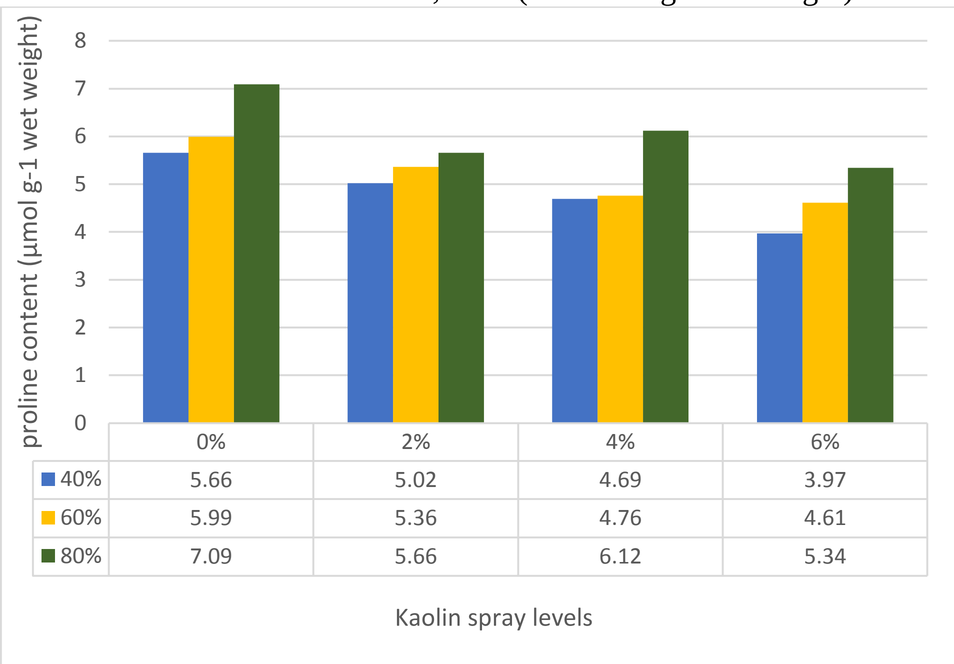
L.S.D=Depletion level 2.45 , Kaolin spray 1.93 , interaction N.S

Figure 3. Effect of Available water depletion levels and kaolin on water content of flag leaves (%)

3.3 Proline content of flag leaves (μmol g^{-1} wet weight):

Figure (4) shows a significant effect of the treatments of depletion, spraying kaolin and their interaction on the proline content of flag leaves. The results of the table showed that the irrigation treatment after depleting 80% of the Available water was superior to the proline content of flag leaves with an average of 6.05 (micromol g^{-1} wet weight), compared to the irrigation treatment after depleting 40% of the Available water, which had the lowest average for this trait, reaching 4.83 (micromol g^{-1} wet weight). The reason for the increase in proline content with the increase in the level of depletion may be attributed to the fact that protein-degrading enzymes such as Proteinase increase with the increase in water tension in the plant, which contributes to the increase in the accumulation of the amino acid proline, which is one of the solutes that play a role in the osmotic regulation process, as it works to reduce the water potential of the leaf cells, causing water to enter them and raise its content in the leaves, which in turn leads to mitigating the negative effects of water tension to which the plant is exposed [11]. This result is consistent with what was reached by [13], who indicated that the proline content increases with the increase in water tension.

The results also showed an increase in the proline content in the plant when the control treatment (without spraying) was applied with an average of 6.24 (micromol g^{-1} wet weight), compared to a concentration of 6% kaolin, which had the lowest average for this trait, reaching 4.64 (micromol g^{-1} wet weight). The role of kaolin in reducing osmotic stress and water loss, improving plant water relations, and protecting plants from exposure to water stress [14], which does not require the production and accumulation of proline in large quantities to raise the osmotic stress, in addition to the increase achieved in the level of growth-regulating hormones IAA, GA, and CK and the decrease in ABA concentration when spraying kaolin [15], which stimulates plant growth and protein synthesis, and thus reduces the proline content. The table results also showed a significant effect of the interaction, as the table results showed the superiority of the 80% depletion treatment combination with the comparison treatment (without spraying) with an average of 7.09 (micromol g^{-1} wet weight), compared to the 40% and 6% concentration combination, which had the lowest average of 3.97 (micromol g^{-1} wet weight).

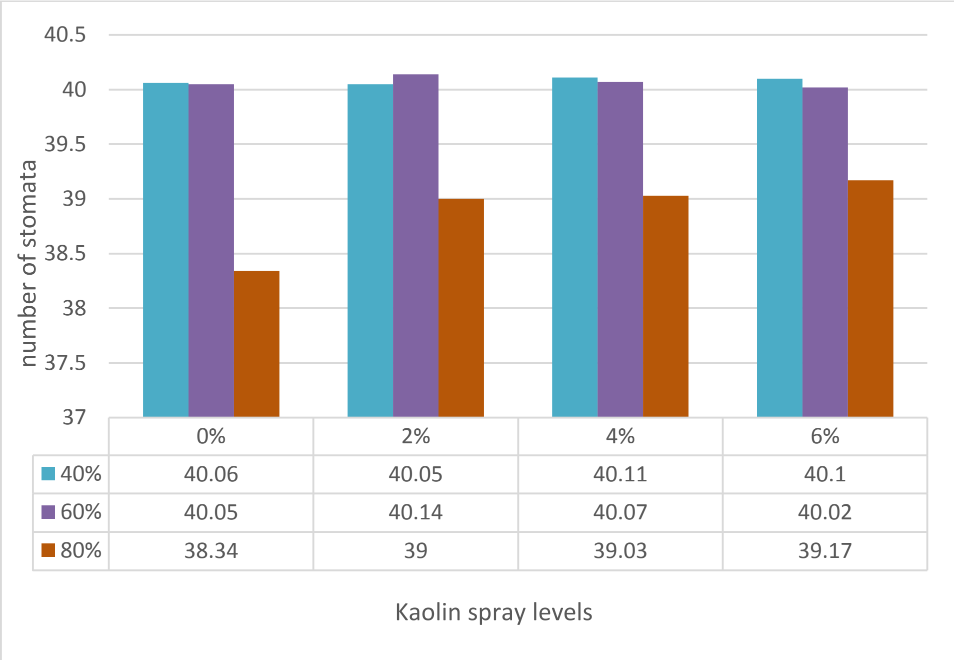


L.S.D=Depletion level 0.09 , Kaolin spray 0.27 , interaction 0.42

Figure 4. Effect of Available water depletion levels and kaolin on proline content of flag leaves (µmol g^{-1} wet weight).

3.4. Carbonate Rock Fragments

The results of Figure (5) indicate that there is a significant effect of the depletion treatments, while spraying kaolin and the interaction between them did not have a significant effect. The results of the table showed the effect of the depletion treatments on the average number of stomata in the flag leaf, as the irrigation treatment after depleting 40% of the Available water gave the highest average number of stomata, reaching 40.08 stomata mm^2, without a significant difference from the irrigation treatment after depleting 60%, with an average of 40.07 stomata mm^2, while the irrigation treatment after depleting 80% of the Available water gave the lowest average for this trait, reaching 38.88 stomata mm^2. The reason for the decrease in the number of stomata when the level of depletion increases may be attributed to the fact that the response of stomata is closely related to the soil moisture content, as stomata respond to chemical signals such as (ABA) produced by roots to confront water shortage conditions [16], especially when high temperatures accompany these conditions. These results are consistent with what was reached by [17,18], who showed that the number and density of stomata are reduced in wheat crops when exposed to water stress.

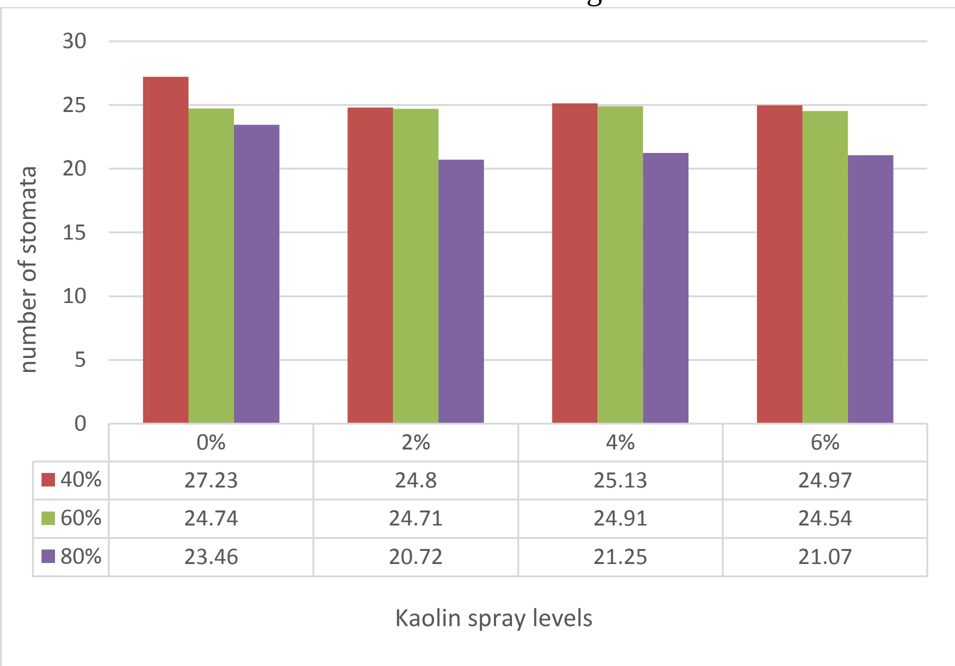


L.S.D=Depletion level 0.99 , Kaolin spray NS , interaction N.S

Figure 5. Effect of available water depletion levels and kaolin on the number of stomata in the flag leaf (mm^2)

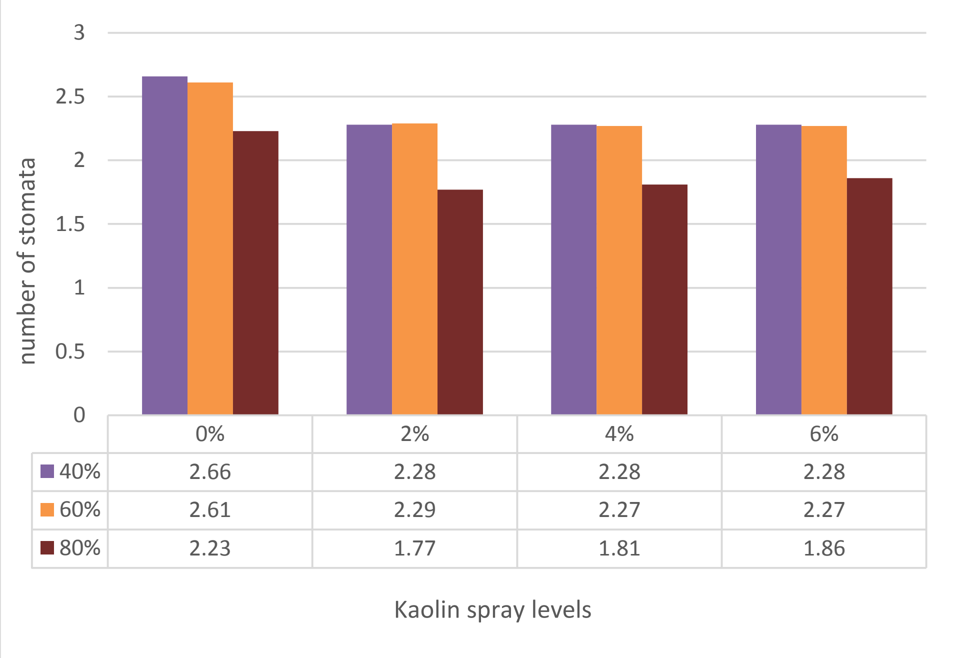
3.5 Length and width of the stoma opening (microns):

The results of (Figures 6 and 7) showed a significant effect of the depletion treatments and kaolin spraying on the length and width of the stomatal opening. At the same time, the interaction between them was significant at the length of the stomatal opening only. Table 6 showed the superiority of the irrigation treatment after depletion of 40% of the Available water with an average of 25.53 (microns), compared to the irrigation treatment after depletion of 80% of the Available water with an average of 21.63 (microns). The results of Table 7 also showed a significant superiority of the depletion treatment when irrigating 40% of the Available water and without a significant difference from the depletion treatment when irrigating 60% of the Available water with averages of 2.37 and 2.36 (microns) for the two treatments, respectively, compared to the depletion treatment when irrigating 80% of the Available water, which gave the lowest average of 1.92 (microns). The increase in the level of depletion led to a reduction in the length and width of the stomatal opening, which affected the entry of carbon dioxide and reduced its fixation in the photosynthesis process, and thus its effect on plant growth and reduced yield. This result is consistent with previously reached by [2,2], who found that the length and width of the stomatal opening were reduced when wheat plants were exposed to water stress. Figure 6 also showed a reduction in the stomatal length in the kaolin spray treatments without a significant difference in average compared to the comparison treatment (without spraying), which had the highest average of 25.14 (microns). Figure 7 also showed a reduction in the stomatal opening width in the kaolin spray treatments without a significant difference, compared to the comparison treatment (without spraying), which gave the highest average of 2.50 (microns). The reason for reducing the length and width of the stomatal opening due to the effect of spraying kaolin may be attributed to the fact that kaolin is an anti-transpiration agent and is a white clay deposit that is added by spraying on the leaves. It works to reflect part of the incident light rays, thus preventing the leaf temperature from rising and controlling the water balance inside the plant cell. Thus. It works to reduce water loss from the green surface (leaves) by increasing their relative water content, especially when exposed to severe water stress levels, as it works to partially close the stomata, thus reducing the size of the stoma without affecting the effectiveness of carbon metabolism and maintaining water potential, thus increasing their tolerance to drought by increasing their relative water content [20]. The interaction in Table 6 also showed a significant effect, as the combination outperformed the irrigation after depleting 40% of the Available water with the comparison treatment (without spraying) with an average of 27.23 (microns), compared to the combination of 6% concentration with all depletion treatments, which had the lowest averages.



L.S.D=Depletion level 0.71 , Kaolin spray 0.43 , interaction 0.85

Figure 6. Effect of Available water and kaolin depletion levels on stomatal aperture length (microns)



	0%	2%	4%	6%
40%	2.66	2.28	2.28	2.28
60%	2.61	2.29	2.27	2.27
80%	2.23	1.77	1.81	1.86

L.S.D=Depletion level 0.02 , Kaolin spray 0.04 , interaction N.S

Figure 7. Effect of available water depletion levels and kaolin on the width of width opening (microns)

Conclusions

The field experiment demonstrated that moderate irrigation at 40% available water depletion significantly improved a range of physiological and anatomical traits in wheat. Additionally, the application of 6% kaolin foliar spray resulted in improvements in plant characteristics compared to the control group. Combining moderate irrigation with kaolin foliar spray effectively enhanced water retention and chlorophyll efficiency, suggesting its potential in drought management. These results highlight the significant physiological benefits of kaolin, which may surpass traditional structural adaptations, warranting further research on integrated drought-mitigation strategies.

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