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Effect of plant density and drought stress on seed productivity and biochemical composition of faba bean plants

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Abstract

Two field experiments were conducted at the Research and Experiment Centre, Faculty of Agriculture, Moshtohor, Benha University, Qaluibia Governorate, Egypt, during two growing winter seasons (2021–2022)&(2022-2023) to investigate the effect of four different water regimes, namely one, two and three irrigations, as well as conventional irrigations (four regular consecutive irrigations as a controller) applied at all vegetative and reproductive stages, and three different plant densities (20, 30 and 40 kg/ kg/fed), of faba bean (Cv. Marriott 2). The collected results demonstrated that plant density had a significant impact on various features,throughout the two winter growing seasons(2021/22 , 2022/23).The contents of nitrogen, crude protein, phosphorus, potassium, carbohydrates contents and the total seed storage protein dramatically increased when plant density was increased except calcium content. During the winter growing season(2021/22), all traits aside from the potassium and calcium contents were significantly impacted by interactions. Overall, the faba bean (Cv. Marriott 2) was presented in a respectable manner, and the findings demonstrated that different plant densities and water regimes had different chemical compositions. Crop composition may be slightly delayed by a water shortage (three irrigation treatments) of up to 75% of the water holding capacity; however, this does not limit the crop's ability to react to irregularities in its chemical composition. Additionally, in every attribute evaluated, faba beans performed well at the most advantageous plant density of 30 kg/fed+ three irrigations.

Keywords: water stress, plant density and faba bean.

INTRODUCTION

Faba bean plant (*vicia faba L.*), is the most significant legume crop in Egypt, which is valued for its high nutritional content for human consumption as well as its role as a break crop in the rotation of cereals. In the previous five seasons, a total of 216,000 feddans were cultivated, with an middling seed yield of 1.4 kg/fed [1].

It contains a lot of carbohydrates and protein. For both green and dry straw, the estimated protein level was 5.5% and 5.9%, respectively. Faba bean seeds have a high protein level that can reach 28%, as well as a high carbohydrate content of 58%. Faba bean may produce more than 6 tonnes of seed per hectare when maintained appropriately. Additionally, by supplying the soil with roughly 20–30 N units/fed after harvest, it aids with biological nitrogen fixation, which raises soil fertility in crop rotations.

Not just in dry and semi-arid areas, but also in areas with adequate rainfall, irrigation water is gradually becoming scarce. Due to its overall low precipitation, high evaporation, and uneven distribution of rainfall over time and space, Egypt suffers from a severe water shortage. In order to address the water fissure problem and support agricultural activities, which account for 85% of the total water utilised in the semi-arid region, water redeemable and preservation are therefore a key and critical requirement.

Due to water resource restrictions, the agriculture sector uses a lot of water, and standard irrigation systems only achieve an irrigation efficiency of about 60%.

In Egypt **[2]**, found that faba bean yields were higher when the plant density was lower 25 plants / m^2 because there were more branches, pods, and seeds per plant.as well as the seed yield and protein content than for the plant density of 33 plants / m^2. **[3]** found the highest seed and protein yield in faba bean for the plant density of 33 plants / m^2.

According to **[4]**, under optimal humidity, a high faba bean yield can be recorded for 20 plants per m^2, however under non-optimal conditions, the plant density can increase up to more than 60 plants per m^2, which points to a considerable plasticity of faba bean plants.

The main objectives of this investigation were to:

- Determine the ideal irrigation period for faba bean irrigation in the research location,
- Examine how one faba bean variety's (c.v. Marriott 2) chemical composition is affected by plant densities and intervals of trickle irrigation.

MATERIAL AND METHODS

During two growing winter seasons (2021–2022 and 2022–2023), this autopsy was conducted at the Agricultural Research and Experimental Centre, Faculty of Agriculture, Moshtohor, Benha University, Qaluibia Governorate, Egypt, to investigate the effects of water stress and plant density at different growth stages on the values of vegetative growth characteristics, yield, and yield components of faba bean plants.

The current study's treatments are as follows:

1-Irrigation regimes:

- At 60 days following planting, there will be one irrigation (high stress) (DAP).
- Two moderately stressful irrigations were performed 50 and 100 days after planting (DAP).
- Three light stress irrigations (days after planting), 40, 80, and 120 days later (DAP).
- Conventional irrigations: once every 30 days, four normal irrigations afterward as a control.

Triangular weirs (V-notch) were used to modify the irrigation outflow. The water's flow height was set at 30 centimetres. Water discharge was measured using the [5] equation, which is as follows:

Q is the water discharge, measured in m^3 per hour, and $Q = 0.0138 \times h^{2.5} \times 3.6$.

Realising that: h = Water height or pressure head (cm); 0.0138 and 3.6 =constant values, where 3.6 was used to achieve Q in m3 hr^{-1}. Efficiency of water usage (WUE)

2- Faba bean densities: there are three plant populations with densities of 20, 30, and 40 kg/fed.

3- Source of plant seeds: Faba bean (Vicia faba L.), seeds of plants(Cv. Marriott 2) were provided from legumes department,ARC, Egypt.

4-Design of an experiment:

There were three replications of the split plot design. The main plots were assigned to the four irrigation treatments, whereas the sub plots were randomly assigned to three faba bean denisties. The subplot's dimensions were 3 x 3.5 m, or 10.5 m^2.

5-Cultural practices:

Before seeding, the proper soil preparation was carried out, and 150 kg/feddan of phosphorus fertilizer calcium super phosphate (15.5%) was used. 5 ridges measuring 3.5 metres long and 60 centimetres wide make up the experimental unit. Faba bean seeds were hand-drilled into the ridges. In the 2022–2023 season, planting took place on November 17 and in the 2022–2023 season on November 19. A 25 kg N/fed nitrogen fertiliser was divided into two equal dosages and applied both before the first irrigation of the two seasons and during planting. Urea (46.5%N) was employed as the N_2 carrier.

Climate data for both of the experiment's growing seasons (Table 1) came from the Agriculture Research Center's Climates Research Station.

6 -Yield and yield components :

During the harvest, ten guarded faba bean plants were taken randomly from the central ridge of each experimental plot for estimation of:

-weight of seeds(kg) plot-1.

7 -Chemical analysis:

Constituents of faba bean seeds chemically:

Seeds at harvest time in (2021-2022) season (the first growing season) were taken to determine their chemical constituents.

- Total nitrogen and crude protein content:(%)

Using [6] methods of wet digestion, the total nitrogen content of seeds was ascertained at the time of harvest. Microkjeldahel was then computed as a percentage of dry weight following the method outlined.[7]

Next, the following equation was used to compute the crude protein:

Crude protein is equal to total nitrogen x 6.25 [8].

Table (1) Qaluibia Governorate's predominant ambient climate parameters for each of the two growth seasons.

Climatic factors and weather average during two growing winter seasons (2021-2022 and 2022-2023)														
	growing winter seasons (2021-2022)							growing winter seasons (2022-2023)						
Factors per Months	NOV. 2021	DEC. 2021	JAN. 2022	FEB. 2022	MAR. 2022	APR. 2022	May 2022	NOV. 2022	DEC. 2022	JAN. 2023	FEB. 2023	MAR. 2023	APR. 2023	May 2023
Temp. (°C) Max.	25	20	19	20	23	28	32	24	19	19	20	23	28	32
Temp. (°C) Min.	15	11	10	11	13	15	19	14	10	10	11	13	15	19
Humidity (%)	63	63	63	58	56	50	49	64	63	63	58	56	50	49
Pressure(mbar)	101	101	101	101	101	101	101	101	101	101	101	101	101	101
	7	9	9	8	6	4	3	6	8	9	8	6	4	3
Precipitation(mm)	0.0	0.2	0.5	0.3	0.1	0.0	0.0	0.1	0.1	0.5	0.3	0.1	0.0	0.0

*The Ministry of Agriculture and Land Reclamation's Central Laboratory for Agricultural Climate (CLAC) at the Agricultural Research Centre (ARC) is the source of this data.

-Tissue ions concentrations measurement :

Calculated as a percentage (%).A sample of 0.5 g of plant seeds was burned at 600 °C for six hours.After adding 3 mL of HCl (10 N) to recover the generated ash, the solution was filtered and then diluted with distilled water to reach a final volume of 100 mL.

-Phosphorus content (%):

Using **Sandell's (1950)** colorimetric method, total phosphorus was measured and expressed as a percentage (%).

-Potassium content (%):

Using the approach outlined by **[7]**, the flame photometer model Carl-Zeiss determined potassium (K^+) and calculated it as a percentage (%).

-Calcium content (%):

With the use of a flame emission photometer (AFP100 Model, Biotech Management Engineering Co. Ltd., UK), the amount of calcium (Ca^{++}) in the resulting solution was determined. utilising the procedure outlined by **[7]** and expressed as a percentage (%).

-Total carbohydrates content (%):

Using the phenol-sulphoric acid method as described by **[8],** the total carbohydrate content was computed as a percentage (%).

-Seed storage protein (SDS-PAGE) in faba bean seeds:

The protein banding patterns for one variety of faba bean (*Vicia faba L.*) were examined using the sodium dodecyl sulphate polyacrylamide (SDS-PAGE) technique. The protein content of stressed and unstressed faba bean seeds was compared, and the plants'seeds overall health were assessed using method of **[9].**

-Analytical statistics: In accordance with **[10],** the analysis of variance was performed for the data from each of the two growth seasons. When comparing means, the 5% threshold of the L.S.D. test was employed.

RESULTS AND DISCUSSION

- Impact of varying densities of faba beans and water regimes, as well as their interactions, on:

A-Weight of seeds (kg) plot-1:

Fallouts in table (1) indicated that the mean values of seeds weight kg plot-1 of faba bean plants was significantly increased by increasing number of irrigations up to applied normal irrigation (control) in the corresponding first and second seasons.

The maximum value of seeds weight kg plot-1 were (0.160 and 1.300 kg plot-1) which were acquried from normal irrigation in the first and second seasons respectively, whereas the minimum ones were (0.094 and 0.791 kg plot-1) from one irrigation. It is typically observed that the largest seed weight kg plot-1 was produced by applied normal irrigation (control), which was followed by three irrigations in the first season, two irrigations in the second, and one irrigation in the third. The application of two irrigations of slightly different magnitudes and regular irrigation (control) appear to be similar in general, with no discernible changes. These results are in the line with those accomplished by . [11-26]

Data in table (2) exert that faba bean densities evidentiate there were significantly differences in seeds weight kg plot-1 in both seasons. The results indicates that density 20 kg/fed was the highest seeds weight (0.143 kg plot-1), followed by density 30 kg/fed (0.131 kg plot-1) then density 40 kg/fed (0.123 kg plot-1) in the first and in the second seasons density 30 kg/fed was the highest seeds weight (1.035 kg plot-1), followed by density 20 kg/fed (1.029 kg plot-1) then density 40 kg/fed (1.018 kg plot-1), respectively .

Also,the results illustrated that, applied 50 kg/fed produce highest seeds weight kg plot-1 in the 1st season and applied 30 kg/fed was the highest in the 2nd season respectively. These results concur with those that were published by. [27-36]

There were insignificant differentiations of the mean values of seeds weight kg plot-1 may be due to the interaction between irrigation treatments and densities of faba bean in the first and second seasons respectively, applied 20 kg/fed when irrigated at normal irrigation (control) gave the greatest seed yield kg plot-1 in the first and the second seasons, respectively. Meanwhile, the lowest results was produced from adding 40 kg/fed when irrigated by one irrigation treatment in the first season,and adding 20 kg/fed in the second season under same conditions, respectively. It could be concluded that applied 20 kg/fed with applied normal irrigation (control) gave the best values of seeds weight kg plot-1. These outcomes concur with those that were published by [28 & 37].

Table (2) Effect of water regimes and plant density on weight (kg) of seeds/plot of faba bean plants during each of two growing winter seasons (2021/22 and 2022/23).

Water regimes (W)	2021/2022 Season Faba bean densities kg/fed			
	Density=20kg	Density=30kg	Density=40kg	Means
One	0.105	0.092	0.083	0.094
Two	0.138	0.132	0.121	0.130
Three	0.156	0.143	0.139	0.146
Control	0.174	0.153	0.149	0.160
Means	0.143	0.131	0.123	

L.S.D. at 5% for water = 0.00, for Densities= 0.002 and for interaction = N.S

Table (2) Continue.

Water regimes (W)	2022/2023 Season Faba bean densities kg/fed			
	Density=20kg	Density=30kg	Density=40kg	Means
One	0.761	0.790	0.823	0.791
Two	0.888	0.933	0.966	0.929
Three	1.083	1.100	1.083	1.089
Control	1.383	1.317	1.200	1.300
Means	1.029	1.035	1.018	

L.S.D. at 5% for water = 0.09 ,for Densities = 0.04 and for interaction = N.S

One =One irrigation at beginning of flowering (B.F.).
Two =Two irrigations at vegetative growth (V.G) and beginning of flowering (B.F.).
Three =Three irrigations at vegetative growth (V.G.), beginning of flowering (B.F.) and full seed formation .
Control =Normal irrigation (Four irrigations as a control).at all vegetative and reproductive stages.

B: Chemical properties of faba bean seeds:

1.Nitrogen content (%) inseeds of faba bean:

Table (3), provides results about the impact of water regimes, faba bean densities, and their combinations on the percentage of nitrogen in faba bean seeds during the winter growing season (2021/2022).

Table (3) Effect of water regimes and plant density on nitrogen content (%) of faba bean seeds during the winter growing season (2021/2022).

Water regimes (W)	2021/2022 Season Faba bean densities kg/fed			
	Density=20kg	Density=30kg	Density=40kg	Means
One	5.90	5.99	6.15	6.01
Two	5.91	5.88	6.16	5.98
Three	5.10	5.69	6.05	5.61
Control	4.63	5.06	5.38	5.12
Means	**5.46**	**5.65**	**5.93**	

L.S.D. at 5% for water = 0.10 , for Densities = 0.07 and for interaction = 0.15

Table (3) displays the average nitrogen concentration in faba bean seeds as influenced by water regimes. Significant variations in the nitrogen content (%) of seeds during the winter growing season (2021/2022) were shown by irrigation regimens.The nitrogen content (%) in faba bean seeds is as follows: 6.01%, 5.98%, 5.12%, and so on when one irrigation treatment is applied, two irrigation treatments are applied, and then a regular irrigation treatment is applied, in that order. Similar results were also reported by **[17,21,22,26,38-40].**

The results presented in Table (3) demonstrated notable variations in the overall percentage of nitrogen present in seeds across the three densities over the winter growth season (2021/2022).

Adding 40 kg/fed yielded the highest N content (%) in seeds during the winter growing season (2021–2022). Conversely, adding 20 kg/fed yielded the lowest content (5.46%), meaning that the remaining density was in between as a single group. It was possible to draw the conclusion that growing 40 kg/fed resulted in seeds with a higher N content (%) when different irrigation techniques were applied. It is evident that the variations in nitrogen content (%) seen for each of the growing densities of faba beans may result from their unique genetic makeup, which interacts differentially with the environmental variables under investigation in diverse ways. These verdicts are in agreement with those reported by **[41-43].**

The interaction between faba bean densities and water regimes in table (3) had a substantial impact on the nitrogen content (%) in faba bean seeds.The maximum nitrogen concentration (6.16%) may have been achieved by applying two irrigations at the vegetative (V.G.) and beginning of blooming (B.F.) stages with 40 kg/fed, followed by one irrigation at the same density (6.15%). On the other hand, during the winter growing season (2021/2022), the use of a standard irrigation treatment with an addition of 20 kg/fed produced the lowest values of N content (4.63%).These grades are in harmony with those obtained by **[37&44]**

2.Protein content (%) in faba bean seeds:

Table (4), presents the findings about the impact of water regimes, faba bean densities, and their combination on the protein content (%) of faba bean seeds grown in the winter of 2021/2022.

Table (4) Effect of water regimes and plant density on protein content (%) of faba bean seeds during the winter growing season (2021/2022).

Water regimes (W)	2021/2022 Season			
	Faba bean densities kg/fed			
	Density=20kg	**Density=30kg**	**Density=40kg**	**Means**
One	36.92	37.44	38.43	37.60
Two	36.96	36.81	38.52	37.43
Three	33.97	35.56	37.81	35.78
Control	30.84	31.67	33.63	32.04
Means	**34.67**	**35.37**	**37.10**	
L.S.D. at 5% for water = 1.06 , for Densities = 0.85 and for interaction = 1.72				

Table (4) displays the values protein content (%) in faba bean seeds as impacted by water regimes. During the winter growing season (2021/2022), the protein content of seeds varied significantly depending on the irrigation regime. The application of one irrigation during the flowering stage produced the highest mean value of protein content (%) in seeds (37.60%), which followed by the application of two irrigations during the winter growth season (37.43%) (2021/2022). During the winter growing season (2021/2022), the application of normal irrigation treatment yielded the lowest mean value of protein content (%) in seeds (32.04%). Similar results were also obtained b **[17,21,22,26,38-40].**

In the winter growing season (2021/2022), the three densities under investigation were significant in terms of the protein content in seeds table (4). The density of 40 kg/fed had the highest protein content (%) at 37.10%, followed by 30 kg/fed at 35.37%, and 50 kg/fed at 34.67%. The differences between the three densities were nearly statistically significant. Additionally, during the 2021–2022 winter growing season. It is also evident that there was a minor variation in the three faba bean densities, with density 40 kg/fed having the highest protein content and density 30 kg/fed coming in second and 40 kg/fed in third place, with density 40 kg/fed having a significant advantage over the other two densities. In comparison to the other two densities, it was determined that the density of 40 kg/fed was better in assimilating the protein percentage content in faba bean seeds. It is abundantly evident that the variations in protein content observed for each of the faba bean densities under investigation may result from their unique genetic make up interacting in distinct ways with the environmental variables in place at the time of the study. These judgments are in agreement with those reported by **[41-43].**

The interaction between water regimes and plant densities over the winter growth season (2021/2022) had a substantial impact on the protein content (%) in faba bean seeds table (4).The application of two irrigations at the flowering stage treatment with density 40 kg/fed during the winter growing season (2021/2022) produced the highest protein content (38.52%), while the application of a normal irrigation treatment with density 20 kg/fed during the winter growing season (2021/2022) produced the lowest protein content value (30.84%).These ranks are in harmony with those obtained **[37, 44].**

3.Phosphorus content (%) in Seeds of faba beans:

Table (5) presents the findings on the impact of water regimes, faba bean densities, and their combination on the phosphorus content (%) in faba bean seeds during the winter growing season (2021/2022).

Table (5) Effect of water regimes and plant density on phosphorus content (%) of faba bean seeds during the winter growing season (2021/2022).

Water regimes (W)	**2021/2022 Season**			
	Faba bean densities kg/fed			
	Density=20kg	**Density=30kg**	**Density=40kg**	**Means**
One	2.10	2.55	3.04	2.56
Two	1.80	1.84	1.95	1.86
Three	1.71	1.84	1.82	1.79
Control	1.68	1.65	1.47	1.60
Means	**1.82**	**1.97**	**2.07**	
L.S.D. at 5% for water = 0.04 , for Densities = 0.03 and for interaction = 0.07				

Table (5) displays the averages phosphorus content (%) in faba bean seeds as influenced by water regime. During the winter growing season (2021–2022), irrigation regimes had a notable impact on the phosphorus content (%) of seeds. In the winter growing season (2021/2022), the application of two irrigation treatments yielded the highest mean value of phosphorus content in seeds (2.56%), whereas one irrigation treatment yielded the lowest mean values (1.86%). However, during the winter growing season (2021–2022), the administration of standard irrigation treatments resulted in the lowest mean value of phosphorus concentration in seeds (1.60%). These results authorize what were reported by **[26,31,38,40&41]**

The findings shown in table (5) unequivocally show that, during the winter growing season (2021–2022), there were no appreciable variations in the phosphorus content (%) of the seeds of the faba bean varieties under study. During the winter growth season (2021/2022), density 40 kg/fed had the greatest values (2.07%), followed by density 30 kg/fed and density 20 kg/fed by (1.97%) and (1.82%), respectively. It is abundantly evident that the variations in phosphorus content (%) seen for each of the cultivated faba bean densities may result from their unique genetic make up, which interacts differently with the environmental variables under investigation in a variety of distinct ways. These results are in agreement with those obtained by **[42,43].**

Water regimes and densities in both seasons had a substantial impact on the percentage of phosphorus in faba bean seeds. table (5). The results, however, generally show that the application of one irrigation treatment with a density of 40 kg/fed during the winter growing season (2021/2022) produced the highest phosphorus content (3.04%), while the application of normal irrigation at the flowering stage treatment with a density of 20 kg/fed during the winter growing season (2021/2022) produced the lowest values of phosphorus content (1.68%). These fallouts are at one with those obtained by **[44].**

4.Potassium content (%) in faba bean seeds:

Table (6) provides information on the effects of water regimes, faba bean densities, and their interactions on the percentage of potassium in faba bean seeds during the winter growing season (2021/2022).

Table (6) displays the average potassium content (%) in faba bean seeds as influenced by water regime. During the winter growing season (2021/2022), notable variations in the potassium content of seeds were observed across irrigation regimes. In the winter growing season (2021/2022), the application of one irrigation treatment yielded the highest mean values of potassium content in seeds (5.28%), whereas the application of two irrigation treatments yielded the highest mean values (4.19%). While regular watering during the winter growing season (2021–2022) produced the lowest mean value of potassium concentration in seeds (3.89%). The obtained marks are compatible with those obtained by **[26,31,38,39,40&45].**

During the winter growing season (2021/2022), there was a substantial variation in the potassium content (%) of seeds among the three densities under investigation.According to table (6), density 40 kg/fed had the highest potassium content (4.43%), followed by density 30 kg/fed (4.36%). By density 20 kg/fed, the lowest value was found to be (4.30%). It is abundantly evident that there were minor but noteworthy variations in the densities. In the meantime, the same pattern of the three densities' arrangement was seen in the winter growth season (2021/2022), with density 40 kg/fed having the highest potassium content, followed by density 30 kg/fed and density 50 kg/fed, with density 40 kg/fed showing a notable advantage over the other two densities. It is evident that the variations in potassium concentration observed for each of the faba bean densities under cultivation could potentially be attributed to These upshots are likeminded obtained by **[42-43].**

The interaction between water regimes and densities in the second season did not significantly impact the potassium content (%) in fababean seeds table (6). During the winter growing season (2021/2022), the application of a single irrigation treatment with a density of 40 kg/fed resulted in the highest potassium content (5.37%), while the application of a regular irrigation treatment with a density of 20 kg/fed produced the lowest potassium content values (3.84%).These domino effect are at one with those obtained by **[44].**

Table (6) Effect of water regimes and plant density on potassium content (%) of faba bean seeds during the winter growing season (2021/2022).

Water regimes (W)	2021/2022 Season Faba bean densities kg/fed			
	Density=20kg	Density=30kg	Density=40kg	Means
One	5.20	5.25	5.37	**5.28**
Two	4.12	4.18	4.26	**4.19**
Three	4.03	4.13	4.16	**4.11**
Control	3.84	3.90	3.95	**3.89**
Means	**4.30**	**4.36**	**4.43**	

L.S.D. at 5% for water = 0.07 , for Densities = 0.02 and for interaction = N.S

Table (7) Effect of water regimes and plant density on calcium content (%) of faba bean seeds during the winter growing season (2021/2022).

Water regimes (W)	2021/2022 Season Faba bean densities kg/fed			
	Density=20kg	**Density=30kg**	**Density=40kg**	**Means**
One	5.80	5.34	5.11	**5.41**
Two	4.95	4.86	4.78	**4.86**
Three	3.56	3.24	3.15	**3.32**
Control	2.70	2.11	2.10	**2.30**
Means	**4.25**	**3.89**	**3.78**	

L.S.D. at 5% for water = 0.13 , for Densities = 0.08 and for interaction = N.S

5.Calcium content (%) in faba bean seeds:

Table (7) provides accessible results about the effects of water regimes, faba bean densites, and their interactions on the calcium content (%) in seeds during the winter growth season (2021/2022).

Table (7) displays the average calcium content (%) in faba bean plant seeds as influenced by water regime.During the winter growing season (2021/2022), there were notable variations in the calcium content (%) of leaves based on irrigation regimes. During the winter growing season (2021/2022), the application of one irrigation treatment produced the greatest mean values of calcium content (%) in seeds *i.e.* 5.41% , followed by the administration of two irrigation treatments (4.86%).

While regular irrigation during the winter growing season (2021/2022) produced the lowest mean value of calcium content (%) in seeds (2.30%). Such obtained results are along the same line as those of **[31,38,39&40].**

The calcium content (%) of seeds at the three densities under investigation fluctuated significantly during the winter growth season (2021/2022). Table (7) usually suggests that density 20 kg/fed had the highest calcium content (%) at 4.25%, followed by density 30 kg/fed at 3.891% and density 40 kg/fed at 3.78%. The variations in density were nearly statistically significant. It is abundantly evident that the variations in calcium content (%) observed for each of the faba bean densities under investigation may result from their unique genetic makeups interacting in distinct ways with the environmental conditions in place at the time of the study. Such obtained results are along the same line as those of **[42&43].**

The calcium content (%) of seeds at the three densities under investigation fluctuated significantly during the winter growth season (2021/2022). Table (7) usually suggests that density 20 kg/fed had the highest calcium content (%) at 4.25%, followed by density 30 kg/fed at 3.891% and density 40 kg/fed at 3.78% . The variations in density were nearly statistically significant. It is abundantly evident that the variations in calcium content (%) observed for each of the faba bean densities under investigation may result from their unique genetic make up interacting in distinct ways with the environmental conditions in place at the time of the study.

6.Total Carbohydrates content (%) in faba bean seeds:

Table (8) provides results about the impact of faba bean densities and water regimes on the percentage of total carbohydrates in seeds during the winter growth season (2021/2022).

Table (8) shows the average total carbohydrate content (%) in faba bean seeds as influenced by water regime.During the winter growing season (2021/2022), there were notable variations in the total carbohydrates content (%) of seeds based on the irrigation regime.

The normal irrigation treatment had the highest mean value of total carbohydrates content (%) in seeds (44.69%), followed by the application of three irrigation treatments in the winter growing season (2021/2022) with a mean value of (36.20%). Additionally, during the winter growing season (2021–2022), the administration of a single irrigation treatment resulted in the lowest mean value of total carbohydrates content in seeds (27.62%). These findings showed that during the winter growing season (2021/2022), the total carbohydrate content (%) in seeds was dramatically decreased by drought stress. Similar comparative studies were conducted by **[11,15,17,22,26,40,46&47].**

Table (8) Effect of water regimes and plant density on carbohydrates content (%) of faba bean seeds during the winter growing season (2021/2022).

Water regimes (W)	2021/2022 Season Faba bean densities kg/fed			
	Density=20kg	**Density=30kg**	**Density=40kg**	**Means**
One	26.86	27.47	28.53	27.62
Two	31.40	32.88	34.58	32.95
Three	35.37	36.41	36.83	36.20
Control	43.23	44.27	46.56	44.69
Means	**34.21**	**35.26**	**36.63**	

L.S.D. at 5% for water = 1.13 , for Denisties = 0.58 and for interaction = 1.16

The total carbohydrate content of the seeds from the three densites under investigation fluctuated significantly during the winter growth season (2021/2022) table (8). It is more likely to be true that density 4 kg/fed (36.63%) had the highest total carbohydrate content, followed by density 30 kg/fed (35.261)%. Density 50 kg/fed has the lowest observed values (34.21%). During the winter growth season (2021/2022), there were nearly substantial differences among the densites. The three densites were frequently seen in their optimal arrangement, with density 40 kg/fed having the highest percentage of total carbs, followed by density 30 kg/fed and density 20 kg/fed, with density 40 kg/fed showing a notable advantage over the other two. It is abundantly evident that the variations in total carbohydrate content (%) observed for every cultivated faba bean densite may be due to their individual specific genetical make up that interact differently with the prevailing environmental conditions under this study in various specific patterns.Such results agree with those reported by **[43].**

During the winter growth season (2021/2022), the interaction between densities and water regimes had a substantial impact on the percentage of total carbohydrates in faba bean seeds table (8). During the winter growing season of (2021/2022), the application of a normal irrigation treatment with a density of 40 kg/fed yielded the highest total carbohydrates content (46.56%), while the application of a single irrigation treatment with a density of 20 kg/fed produced the lowest total carbohydrates content (26.86%). In general, it can be claimed that morphological and biochemical markers could be able to distinguish between the densities of faba beans based on the background of drought tolerance. Analogous results were also obtained by **[27].**

7.Seed Storage Protein pattern of faba bean seeds on SDS-PAGE:

Total seed storage protein of faba bean seeds as affected by water regimes (control and levels of drought stress) had been performed using one-dimensional SDS-PAGE analysis, and optical differences were obtained between the well-watered plants and drought stressed plants in order to identify biochemical markers associated with the above findings.We hypothesised that irrigation regimes showed substantial changes in the total seed store protein by applying two and three irrigation treatments and regular irrigation treatment throughout the winter growing season (2021/2022) based on the morphological results in figures (1-a&b), a density of 30 kg/fed was shown to be the most tolerant of the drought stress, while a density of 20 kg/fed was the most sensitive, in contrast to the regular watering treatment administered during the winter growth season (2021/2022). Figure 1-a provides a practical display of these findings. During the winter growing season (2021/2022), as shown in figure (1-b), three different irrigation treatments were applied. The results indicated that density 20 kg/fed was the most resistant of the drought stress, while density 30 kg/fed was the most sensitive. This is not the case when a standard irrigation treatment is applied in the winter growing season (2021/2022). Many protein bands that were not seen in the protein pattern of the standard irrigation treatment were seen in the protein pattern of the two and three irrigation treatments during the winter growing season (2021/2022) (i.e., appeared at molecular weights of 25, 35, and 180 kDa), figure (1-a&b).The findings also showed that the protein patterns of the drought-tolerant and susceptible densities of faba beans varied from one another and that each was distinguished by the presence of particular protein bands. For instance, bands with a molecular weight of 25 kDa were present in the density of 20 kg/fed with the application of two irrigation treatments (figure 1-a). These bands emerged in the control pattern and vanished in the stressed pattern (three irrigation treatments), Figure1-b.

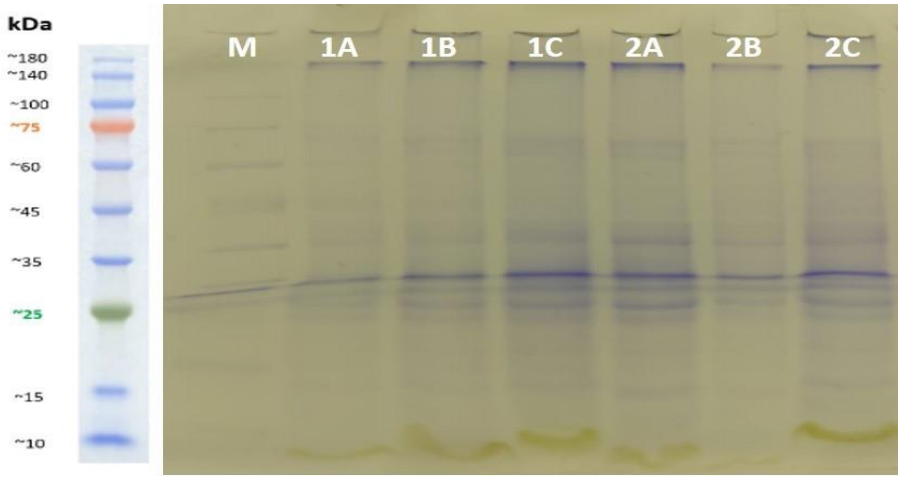

Fig (1-a) Sodium dodecyle sulfate polyacrylamide gel electrophoresis protein pattern of drought stressed faba bean densities separated on 10% SDS-PAGE.

1A=Density 20kg , 1B=Density 30kg , 1C=Density 40kg with (one irrigation treatment).
2A=Density 20kg , 2B=Density 30kg , 2C=Density 40kg with (two irrigations treatment).

The aforementioned results demonstrate that a density of 20 kg/fed can withstand drought stress through the use of two watering treatments. This publication represents nearly the first of its kind on faba bean research. According to **[49]** the increased synthesis of other polypeptides during stress may have compensated for the elimination of other polypeptides.

For the other densities, such as tolerant densities like 2 kg/fed and sensitive densities like 30 kg/fed, similar outcomes might be achieved. figure (1-a) shows that the two most distinct bands were detected at a molecular weight of 14 kDa for the susceptible density and 25 kDa for the tolerant density.

It was the most drought-susceptible variety, even though density 20 kg/fed with application of two watering treatments had the lowest mean through the densities for most of the morphological features. Conversely, a density of 20 kg/fed produced a medium mean across all densities, but it was the variety most resistant to drought. Therefore, it may be concluded that in faba beans, there is a negative link between yield and drought tolerance.

It is noteworthy that morphological and biochemical markers have a strong correlation and that biochemical markers the seed protein profile of seeds are an effective tool for analysing the genetic diversity of faba beans.

Finally,it could be cocluded that, in comparison to the other faba bean densities, the variety density of 20 kg/fed+ application of 3 watering treatments appears to be genetically distinct and may be drought-tolerant.And on the other hand,the economic value for this result is to saving up to 25% from total budget water that consumed in irrigation.

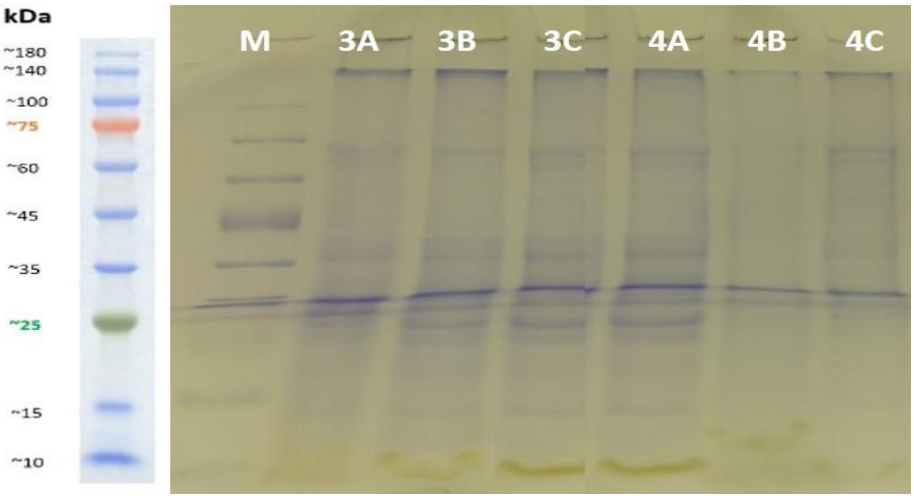

Fig (1-b) Sodium dodecyle sulfate polyacrylamide gel electrophoresis protein pattern of drought stressed faba bean densities separated on 10% SDS-PAGE.

3A=Density 20kg , 3B=Density 30kg , 3C=Density 40kg with (three irrigation treatment).
4A=Density 20kg , 4B=Density 30kg, 4C=Density 40kg with (four irrigations treatment).

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