

## **THE EFFECTIVENESS OF NITROGEN FERTILIZER, BIOFERTILIZER AND MICRONUTRIENTS ON YIELD OF FABA BEAN (VICIA FABA)**

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**ABSTRACT:** *Two field trials were carried out at Kafr El- Zaiyat center, Gharbia Governorate, during two successive seasons of 2008/ 2009 and 2009/ 2010 to identify the integrated effect of two nitrogen rates (0.0 and 20 kg N/fed.), Rhizobium inoculation which contain nitrogen fixing bacteria and some micronutrients mixture (Zn+ Mn+ Fe), added solely or together for improving the yield of faba bean variety i.e. Sakha 3. The tested micronutrients were in the form of EDTA, i.e. Zn- EDTA (12% Zn); Mn-EDTA (12% Mn) and Fe-EDTA (6% Fe) at a rate of 0.3, 0.15, 0.3 g/l, respectively as well as their solution were sprayed on the plants twice at volume of 400 l/fed. after 45 and 70 days from sowing. The obtained results demonstrated that increasing the level of nitrogen fertilizer from 0.0. to 20 kg N/fed. surpassed control in increasing significantly in either stems or leaves and roots dry weights. Roots were the most organs affected by different treatments. Significant increases in chlorophyll a, b and carotenoids after 60 days from sowing. Significant increases in faba bean yield, where the percentage of increase in seed yield reached to 39.72 and 40.74% for first and second seasons, respectively. Corresponding increases significantly in macronutrients, micronutrients concentrations and uptake, crude protein and carbohydrates in seeds and stems for two seasons.*

*Data also indicate that addition of the micronutrients at physiological stage; likewise, Rhizobium inoculation simultaneously induced increases significantly of all studied parameters as well as their values were maximized as results of the combination with nitrogen fertilizer at 20kgN/fed. Moreover, the beneficial effects of the applied treatment included full recommended dose of nitrogen fertilizer (20kgN/fed.) plus Rhizobium inoculation and micronutrients (Zn+ Mn+ Fe) cleared through enhancing the plants growth, and in turn increasing crop yield due to more available and easily mobile of the tested nutrients toward roots and their uptake by plants. Such favourable conditions reflected positively on developments of nutrients concentrations in the seed yield of faba bean.*

**Key Words:** *Faba bean, nitrogen fertilizer, Rhizobium inoculation and micronutrients fertilizer (Zn+ Mn+ Fe)*

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### **INTRODUCTION**

Faba bean (*Vicia faba* L.) is one of the main leguminous crops grown in different types of Egyptian soils (alluvial, sandy and calcareous soils). Also,

it is considering as one of the basic sources of protein in the Egyptian diet with relatively low price. So it seems important to optimize its nutritional program to obtain the maximum yield.

Plant growth depends on adequate supply of nitrogen (N) in order to synthesize the amino acids, protein, nucleic acid and other cellular constituents necessary for plant development (Esteban Sanches, *et al.*, 2004). Kandil (1985) reported that number of pods and seeds/ plants, 100-seeds weight, seed and straw yield were significantly increased by increasing N rates up to 20 kg N/fed. Application of nitrogen fertilizer resulted in: 1) an increase in seeds and straw yield of faba bean (EL- Zeiny *et al.*, 1990 and Hegab and Beshir, 1994) and 2) an increase in seed protein content of faba bean ( Stayanova *et al.*, 1992).

Promising effect of using micronutrients on faba bean yield was found by many workers Baza *et al.*, (1992) and Sabik *et al.*, (2001) found that Fe, Zn, Mn, B, and Cu increased faba bean yield. Also, Osman *et al.*, (1991), resulted in Zn, Mn, and Fe nutrients by coating methods of faba bean seeds grown in alkaline soil corrected the requirement and were suitable balance among such nutrients for growth, uptake and high yield production. Marouah *et al.*, (2003) found that the balanced fertilization including micronutrients ( Fe, Mn and Zn) used as foliar spray and macronutrients (NPK) could increased No of pods/ plant, seeds number/ pods, seeds weight/pods, seed weight/ plant and 100- seeds weight, yield (seeds & straw) and nutrients uptake (N, P, K, Mn, Zn, Cu, Fe) for both seeds and straw.

The micro-organisms colonize in the rhizosphere and enhance plant growth by providing it with nitrogen (Badr and Moawad 1988 and Saleh *et al.*, 2000).The highest yield was mostly associated with Rhizobium. This increase may be attributed to N<sub>2</sub>- fixation which reduced the soil pH especially in the rhizosphere, thereby increase the availability of most essential macro- and micronutrients (Mohamed *et al.*, 2001). Abd-El Kodoos *et al.*, (2002) found that inoculation with Rhizobium and fertilization with micronutrients significantly increased number and weight of nodules, dry matter yield and yield of soybean plant. Also, they found that seed inoculation of soybean ensure adequate N for soybean than application of combined nitrogen fertilizer.

The objective of this experiment was to study the effect of nitrogen, biofertilizer and micronutrients on dry matter, chlorophyll content, chemical composition and yield of faba bean.

## **MATERIALS AND METHODS**

Two field experiments were conducted during two successive seasons 2008/2009 and 2009/2010 at Kafr EL- Zayiat centre, Gharbia Governorate to study the efficiency of nitrogen fertilizer, Rhizobium inoculation and micronutrients mixture (Zn, Mn, and Fe) added either separately or in combination on faba bean genotype i.e. Sakha 3 in its growth, yield and its

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components and chemical compositions of seeds and stems. Representative surface soil samples (0-30 cm) were taken before performance of the experiment, where the physical and chemical analysis characteristics were determined using the standard methods according to Black (1965) and illustrated in Table (1).

**Table (1): Mechanical and chemical properties of the studied soil.**

Soil properties	First season	Second season
<b>Mechanical analysis</b>		
Sand %	13.59	13.82
Silt %	33.55	33.81
Clay %	52.41	52.61
Textural class	Clayey	Clayey
<b>Chemical analysis</b>		
CaCO <sub>3</sub>	2.3	2.45
EC (mmhos/cm 1:5,soil : water)	0.28	0.35
pH (1 : 2.5,soil : water suspension)	7.7	7.81
Available N (ppm)	70.3	75.76
Available P (ppm)	14.3	15.23
Available K (ppm)	435.66	430.67
Available Zn (ppm)	0.75	0.84
Available Mn (ppm)	3.10	3.45
Available Fe (ppm)	6.10	6.55

The faba bean seeds were cultivated on 5 and 10 October in the first and second seasons, respectively. Two seeds were in hill and 20 cm spacing, after emergency plants were thinned to one plant per hill.

The treatments were as follow:

- 1- Control ( no nitrogen fertilizer, biofertilizer and micronutrients)
- 2- 1 N (20 kg N/fed.)
- 3- Zn+ Mn+ Fe (mixture)
- 4- 1 N+ Zn+ Mn+ Fe
- 5- Rhizobia
- 6- 1 N+ Rhizobia
- 7- Zn+ Mn+ Fe+ Rhizobia
- 8- 1 N+ Zn+ Mn+ Fe+ Rhizobia

All plots received phosphorus as calcium super phosphate (15% P<sub>2</sub>O<sub>5</sub>) at a rate of 15 kg P<sub>2</sub>O<sub>5</sub>/fed. before sowing. Potassium as potassium sulphate (48%) was applied at a rate of 24 kg/Fed. as a recommended dose after 35 days from cultivation. Basic application of nitrogen fertilizer at a rate of 20 kg N/fed. was added before the first irrigation (after thinning) directly in the form of ammonium nitrate (33.5%N) as an activating dose. Control treatment without adding either mineral N fertilizer or micronutrients or Rhizobium inoculation. The micronutrient mixture was applied in the form of EDTA compounds i.e. Zn-EDTA (12% Zn), Mn- EDTA (12% Mn) and Fe-EDTA (6% Fe) at a rate of 0.3, 0.15 and 0.3 g/l respectively and applied foliar at two times. The first spray was at 45 days from sowing and the second spray was at 70

days from sowing, at a rate of 400l/fed. Rhizobium contain nitrogen fixing bacteria were inoculated. Inoculation was performed through mixing seeds with the appropriate amount of Rhizobium using a suitable amount of Arabic gum before sowing. The other different field practices were followed in the usual manner for faba bean cultivation. Plants were grown till maturity and sampled twice. The first sample was taken after 60 days from sowing; the second sample was at harvest. At the first sample, Chlorophyll a, b and carotenoids concentrations in leaves spectrophotometer methods recommended by Metzner *et al.*, (1965) were recorded. Also, dry weights of stems, leaves and roots (g/plant) were recorded. The harvest data was on 2 and 4 May for two growing seasons, respectively. At harvest, agronomic trails were determined i.e. plant height (cm), number of branches/ plant, number of pods/plant, weights of pods (g/plant), weights of seeds/plant, 100-seeds weights, seeds yield (ardab/fed.) and straw yield (ton/fed.).

For chemical determination, plants were fine powdered, wet digestion for dry material was carried out according to Chapman and Pratt (1978). Nitrogen % was determined in seeds and stems by micro kjeldahl methods and Crude protein was estimated in such organs by Multipling N% by 6.25 as described by (A.O.A.C. 1990) Phosphorus %, potassium %, and carbohydrates in prior organs were determined using the procedures described by (A.O.A.C 1990). The atomic absorption spectrophotometer was used to determine zinc, manganese and ferrous concentrations in both seeds and stems according to the methods recommended by (A.O.A.C. 1990). Least significant differences test was used for comparing treatments means as described by Gomez and Gomez (1984).

## **RESULTS AND DISCUSSIONS**

### **Dry weights of stems, leaves and roots**

The obtained results in Table (2) revealed that control treatment (no, nitrogen fertilizer, biofertilizer and micronutrients) gave the lowest dry weights (g/plant) of stems, leaves, roots and whole plant that was true for two seasons. Increasing nitrogen fertilizer from zero to 20 kg N/fed. yielded in significant increase in stems dry weights by 54.90 and 55.35 % for first and second seasons, respectively as compared with control. Similar trends were found for leaves, roots and whole plant for sakha 3 faba bean cultivar.

From that Table, it could be noted that micronutrients mixture foliar spray associated with nitrogen fertilizer at 20kgN/fed. induced further significantly increases in studied parameters, where percentage increases in leaves dry weights reached to 12.14 and 12.83% for first and second seasons in a respective order as compared with nitrogen fertilizer used alone. Also more significant increases in dry weights of these organs were produced when Rhizobium inoculation combined with nitrogen fertilizer at prior rate, where percentage increase in dry weights of leaves raised to 58.81 and 58.92% for first and second season, respectively. In this connection, Sprent and Faria

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(1988) revealed that Rhizobium is a major group of heterotrophic nitrogen fixing organism which invade roots through wounds hairs and epidermis. Ishac (1988) found that Rhizobium leguminosarum had the effect of fixing nitrogen on leguminous plants.

Table (2): Effect of nitrogen fertilizer, Rhizobium inoculation and micronutrients either separately or in combinations on dry weights of stems, leaves and roots for Sakha 3 after 60 days from sowin[ 2008/2009 and 2009/ 2010 seasons].

Treatments	2008-2009				2009-2010			
	Dry weight(g/plant)				Dry weight(g/plant)			
	Stems	Leaves	Roots	Whole Plant	Stems	Leaves	Roots	Whole Plant
Control	3.37	2.70	1.20	7.27	4.30	3.57	1.40	9.45
1N	5.22	4.20	1.70	11.12	6.68	5.38	1.90	13.96
Zn+Mn+Fe	3.59	2.99	1.30	7.88	4.64	3.96	1.52	10.12
1N+Zn+Mn+Fe	5.79	4.71	2.74	13.24	7.50	6.07	3.06	16.63
Rhizobia	3.84	3.13	1.40	8.37	4.95	4.14	1.63	10.72
1N+Rhizobia	7.85	6.67	3.69	18.21	10.12	8.55	4.13	22.80
Zn+Mn+Fe+Rhizobia	4.04	3.37	1.51	8.92	5.20	4.45	1.77	11.42
1N+Zn+Mn+Fe+Rhizobia	9.12	7.51	5.59	22.22	11.68	9.63	6.25	27.56
L.S.D	5%	0.19	0.11	0.15	0.20	0.12	0.14	0.23
	1%	0.28	0.16	0.22	0.32	0.18	0.21	0.34
C.V.		10.11	11.24	11.01	10.44	11.21	10.41	11.34

1 N: 20 kg N/ fed.

Moreover, application 20 kg N/fed. of mineral nitrogen fertilizer with micronutrients and Rhizobium inoculation resulted in highly significant increases in dry weights of stems, leaves and roots. The percentage increase in roots dry weight amounted to 228.82 and 228.95 % for first and second seasons, respectively. Such treatment increased significantly whole plant by 99.82 and 97.42%, at a respective order. Those calculations are based on treatment that taken nitrogen fertilizer only. These results are in agreement with those obtained by Marouah *et al.*, (2003) they found that association nitrogen fertilizer together with Rhizobium inoculation and micronutrients produced the highest significant increases in dry weights of faba bean plants after 40 and 110 days from sowing.

### Chlorophyll

Data in Table (3) show that significant increases in chlorophyll content as nitrogen fertilizer added at a rate of 20kg N/fed., over control. The percentage increase in chlorophyll a and b amounted to 50.61 and 62.34%, respectively for first season. In case of second season data were 63.03 and 64.56%, respectively.

Table (3) indicated that micronutrients mixture added solely increased significantly chlorophyll content in leaves with some exception for both seasons. According to Hu and Sparks (1991), zinc play a key role in photosynthesis affecting chlorophyll synthesis. The positive action of Mn on chlorophyll content could be attributed to its effect on the stability of the

chloroplast structure (Clarkson and Hanson, 1980), protection of the photosynthetic apparatus against the dexterous action of oxygen radical (Marschner, 1986). Combined this treatment with 20 kg N/fed. induced high significant increases in these contents and surpassed nitrogen fertilizer treatment. The percentage increases in chlorophyll a and b rose to 103.66 and 87.01%, respectively for first season and 109.1 and 91.14% for second season as compared with the control treatment and 35.22 and 15.20%, in order stated for first season and 28.25 and 16.15%, in order stated for second season, overcoming nitrogen fertilizer alone.

**Table (3): Effect of nitrogen fertilizer, Rhizobium inoculation and micronutrients on chlorophyll content for Sakha 3 variety after 60 days from sowing [2008/ 2009 and 2009/ 2010 seasons].**

Treatments	2008-2009			2009-2010		
	Chl a (ug/ml)	Chl b (ug/ml)	Carotenoids (ug/ml)	Chl a (ug/ml)	Chl b (ug/ml)	Carotenoids (ug/ml)
Control	3.28	0.77	1.07	3.30	0.79	1.08
1 N	4.94	1.25	1.56	5.38	1.30	1.60
Zn+Mn+Fe	3.51	0.85	1.13	3.61	0.89	1.14
1N+Zn+Mn+Fe	6.68	1.44	1.76	6.90	1.51	1.81
Rhizobia	3.71	0.90	1.20	3.80	0.93	1.22
1 N+Rhizobia	7.66	1.63	1.90	8.11	1.72	1.95
Zn+Mn+Fe+Rhizobia	3.98	0.95	1.24	3.99	0.96	1.26
1N+Zn+Mn+Fe+ Rhizobia	9.43	1.90	2.31	9.77	1.96	2.37
L.S.D. 5%	0.16	0.11	0.14	0.17	0.10	0.15
1%	0.24	0.16	0.21	0.25	0.15	0.22
C.V.	7.55	7.91	8.11	8.12	7.63	7.90

1 N: 20 kg N/fed.

Association nitrogen fertilizer with Rhizobium inoculation caused further significant increases in such parameters for both seasons, since percentage increase in carotenoids reached to 21.79 and 21.88% for first and second seasons in a respective order as comparing with nitrogen fertilizer added individually.

Furthermore, highly significant increases in these contents were obtained by application full treatments include nitrogen fertilizer +Zn+ Mn+ Fe+ Rhizobia, where percentage increase in carotenoides raised to 48.08 and 48.13% as compared with nitrogen fertilizer and 115.89 and 119.44% for first and second seasons at a respective order as compared with the control treatments.

### **Yield and yield components**

Pooled data in Tables (4&5), indicated that yield and yield components for Sakha 3 genotype was significantly increase with addition of nitrogen fertilizer at 20 kg N/fed., over Rhizobium inoculation or micronutrients added singly or in combinations as comparing with control. The percentage increase in plant height (cm) and number of branches / plant recorded 31.40 and 50.0%, respectively for first season. Also data were, 30.36 and 100.00%, respectively for second season. The positive role of nitrogen fertilizer was

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due to increase vegetative growth which could be considered as a criterion for the photosynthetic efficiency of the plant (Omar *et al.*, 2008). These results are in accordance with those reported by Hegab and Beshir (1994) and Ahmed *et al.*, (2005).

Results also indicated that action of nitrogen fertilizer was enriched by adding either foliar feeding Zn+ Mn+ Fe mixture or Rhizobium inoculation, where the efficiency of combinations was more than using nitrogen only, which translated in term of further significant increases in yield and yield attributes, where percentage increases in number of pods and weights of pods/ plant amounted to 45.45 and 63.64% respectively for number of pods and 30.85 and 47.20% for weights of pods /plant for first seasons .. Corresponding data for the second season were recorded as 54.55 and 72.73%, respectively for number of pods and 32.72 and 49.80 % respectively for weights of pods/ plant. It is obvious that nitrogen fertilizer associated with Rhizobium inoculation induced higher values more that its combination with micronutrients. The promotive effect of micronutrients is in agreement with those reported by EL- Sayim and Sawan (2007). Also, the beneficial effect of Rhizobia inoculate obtained under this study may be due to two reasons, the first is the promotive effect of non- symbiotic N<sub>2</sub>- fixing bacteria to plants which ascribed to their ability to fix free molecular atmospheric nitrogen. The second one is the role of these bacteria in improving the availability of soil nutrients through creator substances which are important for solubilizing sparingly soluble inorganic compounds and make easy forms available for plant uptake (Knany *et al.*, 2004). Fairy close agreement was obtained by several authors Badr and Estefanous (1997); Mohamed and El-Abbas (2005), and Yoho *et al.*, (2009).

**Table (4): Effect of nitrogen fertilizer, Rhizobium inoculation and micronutrients on yield and yield components for Sakha 3 variety [ 2008-2009 season].**

Treatments	Plant height (cm)	No of branches /plant	No of pods/ plant	Weights of pods /plant (g)	Weights of seeds/ plant (g)	100- seeds weights (g)	Seeds yield ardeb /fed.	Straw yield ton/ fed.
Control	80.41	2	7	30.88	20.6	52.78	7.05	1.22
1N	105.66	3	11	48.98	34.77	79.71	9.85	1.76
Zn+Mn+Fe	84.96	2	7	31.29	21.80	54.61	7.69	1.31
1N+Zn+Mn+Fe	118.36	5	16	64.09	48.24	88.42	11.21	2.16
Rhizoir abbia	86.21	2	8	32.04	22.34	56.60	7.83	1.33
1N+Rhizobia	123.47	6	18	72.10	55.40	92.18	11.91	2.35
Zn+Mn+Fe+Rhizobia	87.46	2	8	32.51	22.97	58.12	7.90	1.38
1N+Zn+Mn+Fe+Rhizobia	131.23	6	20	83.80	63.85	103.54	12.74	2.46
L.S.D.	5%	4.16	0.14	0.11	2.61	1.12	3.66	0.31
	1%	6.14	0.21	0.16	3.85	1.65	5.31	0.46
C.V.		11.91	10.88	12.51	11.34	11.54	10.01	10.64

1N: 20kg N/ fed.

**Table (5): Effect of nitrogen fertilizer, Rhizobium inoculation and micronutrients on yield and yield components for Sakha 3 variety [ 2009-2010 season].**

Treatments	Plant height (cm)	No of branches /plant	No of pods /plant	Weight of pods /plant (g)	Weights of seeds /plant (g)	100 seeds weight (g)	Seeds yield ardad /fed.	Straw yield ton/fed
Control	81.41	2	7	30.91	21.01	52.91	7.07	1.23
1N	106.13	4	11	49.06	35.22	80.11	9.95	1.77
Zn+Mn+Fe	85.39	2	8	31.73	22.25	55.15	7.74	1.32
1N+Zn+Mn+Fe	119.31	6	17	65.11	49.21	89.67	11.68	2.18
Rhizobia	87.95	2	9	32.61	22.85	57.13	7.89	1.33
1N+Rhizobia	124.10	7	19	73.32	56.31	94.26	12.11	2.36
Zn+Mn+Fe+Rhizobia	88.84	3	9	33.49	23.48	58.89	7.95	1.39
1N+Zn+Mn+Fe+Rhizobia	132.54	7	21	87.24	65.44	104.47	12.92	2.48
L.S.D. 5%	6.18	0.16	0.13	3.63	2.15	2.67	1.27	0.30
1%	9.12	0.24	0.19	5.36	3.17	3.94	1.87	0.44
C.V.	12.67	11.31	11.97	10.82	9.99	9.18	10.16	11.22

1N: 20kg N/ fed.

Results also showed that nitrogen fertilizer action was greatly effective when associated with Rhizobium inoculation and micronutrients, where significantly highly increases in weights of seeds/plant and 100-seeds weights (g), by about 83.64 and 29.90%, at a respective order for first season (Table 4) while in case of second season, results were 85.80 and 30.41 %, in a respective order (Table 5) were recorded. Prior treatment increases significantly seeds yield and straw yield by about 29.34 and 39.77%, respectively for first season and 29.85 and 40.11%, respectively for second season. This holds was true since the applied treatment played an active role for building new merestemic cells, enhanced cell elongation and increased the ability of faba bean leaves for photosynthetic process (Yu *et al.*, 1999). These results are in agreement with results of Marouah *et al.*, (2003) and El-Sayim and Sawan (2007).

#### **Minerals concentrations and uptake:**

#### **Macronutrients concentrations and uptake**

Data in Tables (6, 7, 8 &9) clearly show increase significantly in nitrogen concentration in seeds and stems as the addition of nitrogen fertilizer at a rate of 20 kg N/fed., followed by micronutrients or Rhizobium inoculation alone or in associations. The percentage of increase reached to 46.08 and 41.79%, respectively for nitrogen concentrations and 104.38 and 104.65%, respectively for nitrogen uptake for first season. Also, in case of second season prior treatment recorded 47.19 and 41.18% for nitrogen concentrations and 107.19 and 103.23%, at a respective order for nitrogen uptake over that of control. The same trend was found for phosphorus and potassium concentrations and uptake for two cultivars. Similar results were obtained by Ahmed *et al.*, (2005).



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Abovementioned Tables, show that motivation of nitrogen fertilizer was highly pronounced when combined with either micronutrients foliar addition or Rhizobium inoculation for Sakha 3 genotype, where phosphorus concentration and uptake in seeds recorded 91.30 and 119.57% respectively for phosphorus concentrations and 204.78 and 271.31%, respectively for phosphorus uptake for first season (Table 6) and 88.00 and 112.80% respectively for phosphorus concentration and 210.58 and 264.60%, respectively for phosphorus uptake for second season (Table 8) as comparing with control and more than nitrogen fertilizer only by 15.79 and 32.89% at a respective order for phosphorus concentration and 31.90 and 60.69% for phosphorus uptake, at a respective order for first season and 17.50 and 33.00% at a respective order for phosphorus concentration, also data were 37.93 and 61.91% in the order state for phosphorus uptake for second season. Such treatments could not bring any significant effect for those nutrients concentrations but increases significantly their uptake except for phosphorus uptake in stems as compared with nitrogen fertilizer alone (Tables 7&9) .

**Table (6): Effect of nitrogen fertilizer; Rhizobium inoculation and micronutrients on macronutrients concentrations and uptake in seeds for Sakha 3 variety [ 2008-2009 season].**

Treatments	N		P		K		
	%	Uptake (kg/fed.)	%	Uptake (kg/fed.)	%	Uptake (kg/fed.)	
Control	2.3	25.13	0.23	2.51	0.49	5.35	
1N	3.36	51.30	0.38	5.80	0.81	12.37	
Zn+Mn+Fe	2.35	28.01	0.23	2.74	0.51	6.080	
1N+Zn+Mn+Fe	3.84	66.72	0.44	7.65	0.90	15.64	
Rhizobia	2.41	29.25	0.25	3.03	0.53	6.43	
1N+ Rhizobia	4.01	74.03	0.505	9.32	0.95	17.54	
Zn+Mn+Fe+Rhizobia	2.52	30.86	0.26	3.18	0.55	6.73	
1N+Zn+Mn+Fe+rhizobia	4.25	83.92	0.560	11.06	0.98	19.35	
L.S.D.	5%	0.05	2.56	0.07	1.31	0.1	2.13
	1%	0.07	3.78	0.10	1.93	0.15	3.15
C.V.		7.11	8.31	7.36	9.12	8.12	7.64

1N: 20 kg N/ fed.

In this connection, Dashti *et al.*, (1997) suggested that the promotive mechanism of growth and nitrogen fixing bacteria included direct and indirect effect. The direct one; included an increase in the mobilization of insoluble nutrients followed by enhancement uptake by the plants and production of plant growth regulators that stimulate plant growth. The indirect effect includes positive effect on symbiotic nitrogen fixation by improvement of root nodule number or mass. These results confirm that obtained by Mohamed *et al.*, (2001) and EL- Sayim and Sawan (2007).

**Table (7): Effect of nitrogen fertilizer, Rhizobium inoculation and micronutrients on macronutrients concentrations and uptake in stems for Sakha 3 variety [2008-2009season].**

Treatments	N		P		K		
	%	Uptake (kg/fed.)	%	Uptake (kg/fed.)	%	Uptake (kg/fed.)	
Control	0.67	8.17	0.058	0.71	0.85	10.37	
1N	0.95	16.72	0.087	1.53	1.26	22.18	
Zn+Mn+Fe	0.71	9.30	0.060	0.79	0.89	11.66	
1N+Zn+Mn+Fe	0.99	21.38	0.092	1.99	1.30	28.08	
Rhizobia	0.74	9.84	0.063	0.84	0.90	11.97	
1N+ Rhizobia	1.03	24.21	0.096	2.26	1.32	31.02	
Zn+Mn+Fe+Rhizobia	0.79	10.90	0.065	0.90	0.92	12.70	
1N+Zn+Mn+Fe+rhizobia	1.04	25.58	0.097	2.39	1.35	33.21	
L.S.D.	5 %	0.11	2.18	0.09	1.11	0.12	2.17
	1%	0.16	3.22	0.13	1.64	0.18	3.20
C.V.		9.14	8.74	9.34	8.12	8.44	7.81

1N: 20 kg N /fed.

**Table (8): Effect of nitrogen fertilizer; Rhizobium inoculation and micronutrients on macronutrients concentrations and uptake in seeds for Sakha 3 variety [2009-2010 season].**

Treatments	N		P		K		
	%	Uptake (kg/fed.)	%	Uptake (kg/fed.)	%	Uptake (kg/fed.)	
Control	2.31	25.31	0.25	2.74	0.50	5.48	
1N	3.40	5.44	0.40	6.17	0.82	12.65	
Zn+Mn+Fe	2.37	28.43	0.26	3.12	0.53	6.36	
1N+Zn+Mn+Fe	3.90	70.61	0.47	8.51	0.91	16.47	
Rhizobia	2.45	29.96	0.27	3.30	0.56	6.85	
1N+Rhizobia	4.06	76.21	0.532	9.99	0.97	18.21	
Zn+Mn+Fe+Rhizobia	2.54	31.30	0.28	3.45	0.59	7.27	
1N+Zn+Mn+Fe+Rhizobia	4.31	86.31	0.591	11.84	0.99	19.83	
L.S.D.	5%	0.13	3.60	0.09	1.34	0.06	2.17
	1%	0.19	5.31	0.13	1.98	0.08	3.20
C.V.		9.67	7.66	8.42	7.71	7.33	9.11

1N: 20 kg N/fed

Furthermore, highly significant effect in those concentrations and uptake in seeds under this study were induced by co-fertilization nitrogen fertilization with Zn+ Mn+ Fe as foliar feeding and seeds inoculation with Rhizobium, more nitrogen fertilizer used alone. Since the percentage in potassium concentration and uptake reached to 20.99 and 56.43%, respectively for first and second season. In case of second season, results were 20.73% and 56.76, at respective order. Such increases were accompanied by corresponding non significant effect in macronutrients concentrations in stems. However, prior treatment increased significantly their uptake except for phosphorus uptake in prior organ for two seasons as compared with nitrogen fertilizer used alone. Such trend might emphasize

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the translocation of those concentrations from stems to seeds and thereby increase in seed yield occurred under the above mentioned treatments (Tables 4 &5). The increases in macronutrients concentrations is of direct relevance in that the applied treatments have drastic effects on nutrients availability and their mobility in the soil, which reflect positively on the existed grown crops, in particular their growth and nutrients uptake ( Celik et al., 2004).

**Table (9): Effect of nitrogen fertilizer, Rhizobium inoculation and micronutrients on macronutrients concentrations and uptake in stems for Sakha 3 variety [2009-2010].**

Treatments	N		P		K		
	%	Uptake (kg/fed.)	%	Uptake (kg/fed.)	%	Uptake (kg/fed.)	
Control	0.68	8.36	0.06	0.74	0.86	10.58	
1N	0.96	16.99	0.09	1.59	1.31	23.19	
Zn+Mn+Fe	0.73	9.64	0.063	0.83	0.90	11.88	
1N+Zn+Mn+Fe	1.00	21.80	0.094	2.05	1.33	28.99	
Rhizobia	0.75	9.98	0.065	0.86	0.91	12.10	
1N+Rhizobia	1.04	24.54	0.098	2.31	1.35	31.86	
Zn+Mn+Fe+Rhizobia	0.80	11.12	0.069	0.96	0.92	13.07	
1N+Zn+Mn+Fe+Rhizobia	1.07	26.54	0.099	2.46	1.38	34.22	
L.S.D.	5%	0.14	2.26	0.05	1.18	0.08	2.19
	1%	0.21	3.34	0.07	1.74	0.11	3.23
C.V.		7.31	7.91	8.91	7.34	9.22	8.18

1N:20 kg N/fed.

**Micronutrients concentrations and uptake**

Results in Tables (10, 11, 12&13) indicate that application nitrogen fertilizer at a rate of 20 kg N/fed. greatly increased significantly micronutrients concentrations and uptake in seeds and stems, more than micronutrients and Rhizobia inoculate. That was true for both seasons. The percentage increase in ferrous concentration and uptake amounted to 59.88 and 63.10%, at a respective order for ferrous concentrations and 123.39 and 135.28, respectively for ferrous uptake for first season. In case of second season corresponding significant increase were 61.27 and 61.63% at a respective for ferrous concentration, also data were 126.97 and 132.59, respectively for ferrous uptake. The significant favourable effect of the nitrogen fertilizer may be explained on the basis of the beneficial effect of nitrogen fertilizer in improving the absorption and translocation abilities of Fe, Mn, Cu and Zn to a certain extent in plants (Hu-Lin et al., (2007). Similar results were obtained by Monged et al., (2004).

**Table (10): Effect of nitrogen fertilizer, Rhizobium inoculation and micronutrients on micronutrients concentrations and uptake in seeds for Sakha 3 variety [2008-2009].**

Treatments	Fe		Zn		Mn		
	(ppm)	Uptake (kg/fed.)	(ppm)	Uptake (kg/fed.)	(ppm)	Uptake (kg/fed.)	
Control	172	187.95	26	28.41	57	62.29	
1N	275	419.86	45	68.70	95	145.04	
Zn+Mn+Fe	178	212.17	27	32.18	59	70.33	
1N+Zn+Mn+Fe	303	526.48	57	99.04	127	220.67	
Rhizobia	173	209.96	27	32.77	57	69.18	
1N+Rhizobia	295	544.58	52	95.99	116	214.14	
Zn+Mn+Fe+Rhizobia	180	220.41	28	34.29	59	72.25	
1N+Zn+Mn+Fe+Rhizobia	326	643.75	63	124.41	135	266.58	
L.S.D.	5%	4.66	20.16	2.53	2.66	1.72	5.44
	1%	6.88	29.76	3.73	3.92	2.54	8.03
C.V.		7.11	8.21	7.56	8.55	8.71	7.77

1N: 20kg N /fed.

From that Tables, it could be noted that combination 20kgN/fed. with micronutrients produced further significant increases in studied concentrations and uptake in seeds, and stems more than nitrogen fertilizer alone for both growing seasons, where percentage increase in zinc concentration recorded 26.67 and 20.00%, for first season and 27.08 and 18.75%, at a respective order. In case of zinc uptake, corresponding significant increases were 44.16 and 47.27%, in order state for first season and 49.17 and 46.26%, respectively for second season. Another significant increase in such concentrations in seeds reached to 15.56 and 16.67% for first season and second seasons, respectively non significant effecting such concentrations in stems for both seasons by combining nitrogen fertilizer with Rhizobium inoculation (11&13).

**Table (11): Effect of nitrogen fertilizer; Rhizobium inoculation and micronutrients on micronutrients concentrations and uptake in stems for Sakha 3 variety [ 2008-2009 season].**

Treatments	Fe		Zn		Mn		
	(ppm)	Uptake (kg/fed.)	(ppm)	Uptake (kg/fed.)	(ppm)	Uptake (kg/fed.)	
Control	84	102.48	7	8.54	11	13.42	
1N	137	241.12	15	26.40	24	42.24	
Zn+Mn+Fe	89	116.59	9	11.79	15	19.65	
1N+Zn+Mn+Fe	141	304.56	18	38.88	28	60.48	
Rhizobia	86	114.38	8	10.64	12	15.96	
1N+Rhizobia	139	326.65	16	37.60	25	58.95	
Zn+Mn+Fe+Rhizobia	90	124.20	9	12.42	16	22.08	
1N+Zn+Mn+Fe+Rhizobia	139	341.94	29	71.34	29	71.34	
L.S.D.	5%	3.44	8.13	1.67	1.16	1.11	2.33
	1%	5.08	12.00	2.46	1.71	1.64	3.43
C.V.		9.43	8.63	7.63	8.10	7.29	9.11

1N: 20kgN/fed.

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**Table (12): Effect of nitrogen fertilizer; Rhizobium inoculation and micronutrients on micronutrients concentrations and uptake in seeds for Sakha 3 variety [ 2009-2010 season].**

Treatments	Fe		Zn		Mn	
	(ppm)	Uptake (kg/fed.)	(ppm)	Uptake (kg/fed.)	(ppm)	Uptake (kg/fed.)
Control	173	189.58	26	28.49	58	63.56
1N	279	430.29	48	74.03	97	149.60
Zn+Mn+Fe	180	215.95	27	32.39	60	71.98
1N+Zn+Mn+Fe	310	561.22	61	110.43	129	233.54
Rhizobia	174	212.79	28	34.24	59	72.15
1N+Rhizobia	299	561.24	56	105.11	117	219.61
Zn+Mn+Fe+Rhizobia	183	225.5	28	34.50	60	73.94
1N+Zn+Mn+Fe+Rhizobia	337	674.88	68	136.18	138	276.36
L.S.D. 5%	5.61	21.41	1.44	2.71	1.69	5.71
1%	8.28	31.61	2.13	4.00	2.49	8.43
C.V.	8.11	7.64	9.16	8.55	7.41	9.14

1N: 20kg N/fed.

In case of zinc uptake in seeds and stems, corresponding significant increases were 39.72 and 42.42%, respectively for first season, also data were 41.98 and 41.67% at a respective order for second season by such treatment. Application combined treatment of nitrogen fertilizer + Zn+ Mn+ Fe+ Rhizobia inoculate caused highly significant increases in micronutrients concentrations and uptake in seeds for Sakha 3 variety. Since manganese relative increase reached to 42.11 and 42.26% for such nutrient concentrations, while data 83.80 and 84.73% for its uptake for first and second seasons at order state, over nitrogen fertilizer alone. Prior treatment increased significantly such nutrient concentrations in stems by about 20.83 and 20.00% for first and second seasons, respectively. Corresponding significant increases were 68.89 and 68.14% for studied nutrient uptake. These results are in agreement with those reported by Marouch *et al.*, (2003).

**Table (13): Effect of nitrogen fertilizer, Rhizobium inoculation and micronutrients on micronutrients concentrations and uptake in stems for Sakha 3 variety [2009-2010].**

Treatments	Fe		Zn		Mn	
	(ppm)	Uptake (kg/fed.)	(ppm)	Uptake (Kg/fed.)	(ppm)	Uptake (kg/fed.)
Control	86	105.78	7	8.61	12	14.76
1N	139	246.03	16	28.32	25	44.25
1N+Zn+Mn+Fe	91	120.12	9	11.88	16	21.12
Rhizobia	144	313.92	19	41.42	29	63.22
1N+Rhizobia	88	117.04	7	9.31	13	17.29
Zn+Mn+Fe+Rhizobia	141	332.76	17	40.12	26	61.36
1N+Zn+Mn+Fe+Rhizobia	92	127.88	9	12.51	17	23.63
143	354.64	22	54.56	30	74.40	
L.S.D. 5%	3.14	8.83	1.22	1.21	1.32	2.42
1%	4.63	13.03	1.80	1.78	1.95	3.57
C.V.	9.67	7.78	8.34	9.52	7.63	8.49

1N: 20 kg N/fed.

**Crude protein**

Seed and stems protein in Sakha 3 cultivar for first season data are presented in Table (14) reflect that addition nitrogen fertilizer, and Rhizobium inoculation increased significantly crude protein in such organs, surpassed the control treatment. While micronutrients could not bring any significant improvements in crude protein in seeds for both seasons. The relative increases amounted to 46.04, 2.16 and 4.73%, respectively for seeds and 41.77, 5.97 and 10.50%, respectively for stems. While in case of second season, data were 47.16, 2.56 and 6.02%, respectively for seeds and 41.18, 7.29 and 10.35% respectively for stems. These results indicate that nitrogen fertilizer produced more crude protein percent than other treatments.

Percentage of crude protein in seeds showed that nitrogen fertilizer was more modified when combined with either micronutrients or Rhizobium inoculation to be 14.29 and 19.33%, respectively for first season, more than nitrogen fertilizer only. But in case of second season where prior treatments recorded 14.73 and 19.44%, respectively, over that of nitrogen fertilizer (Table14). Similar trends was found in stems for both seasons. Supportive evidence with these results were reported by Ragab (1998) and Ahmed *et al.*, (2002), they detected an increase in crude protein due to inoculation with Rhizobia.

Table (14) reveals that micronutrients association with Rhizobium inoculation increased significantly crude protein in seeds and stems over the control treatment by 9.53 and 17.90%, respectively for first season and 9.97 and 17.65%, respectively for second season. While nitrogen fertilizer combination with such treatment pear the highly significant increases in crude protein in such organs by about 84.70 and 55.13%, respectively for first season, and 86.57 and 57.41% , respectively for second season, as comparing with the control treatment and overcoming nitrogen fertilizer by 26.48 and 9.43%, respectively for first season. Also data were 26.78 and 11.50%, respectively for second season.

**Table (14): Effect of nitrogen fertilizer, Rhizobium inoculation and micronutrients on crude protein percent in seeds and stems for Sakha 3 variety [ 2008-2009 and 2009-2010 seasons].**

Treatments	2008-2009		2009-2010	
	Seeds	Stems	Seeds	Stems
Control	14.38	4.19	14.44	4.25
1N	21.0	5.94	21.25	6.0
Zn+Mn+Fe	14.69	4.44	14.81	4.56
1N+Zn+Mn+Fe	24.00	6.19	24.38	6.25
Rhizobia	15.06	4.63	15.31	4.69
1N+Rhizobia	25.06	6.44	25.38	6.50
Zn+Mn+Fe+Rhizobia	15.75	4.94	15.88	5.00
1N+Zn+Mn+Fe+Rhizobia	26.56	6.50	26.94	6.69
L.S.D.				
	5%	0.33	0.22	0.40
	1%	0.49	0.32	0.59
C.V.		8.27	8.66	7.61
				9.45

1N: 20 kg N/fed.

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**Carbohydrates**

It is quite from data presented in Tales (15 &16) that total carbohydrates and soluble sugars fractions had none significantly effect as a results of application of micronutrients or Rhizobium inoculation either singly or in combination, While mineral nitrogen fertilizer at a rate of 20kgN/fed. produced significant increases. That was true in different parts of the plants for both seasons. Since percentage increase in total carbohydrates in seeds and stems reached 53.61 and 48.02%, respectively for first seasons. Also data were 53.75 and 49.14%, respectively for second season.

The most interesting finding data in Tables (15 &16) indicated that when plant received nitrogen fertilizer at the same rate in combination with Zn+ Mn+ Fe mixture achieved high significant increases in those parameters, where relative increases in soluble sugars amounted to 14.11 and 15.03% in seeds for first and second seasons, respectively overcoming nitrogen fertilizer used only. Corresponding more significant increases in such parameter by 20.54 and 20.82% for first and second season, respectively were obtained when nitrogen fertilizer combined with Rhizobium inoculation. It is important here to mention that the beneficial effect of bio fertilizer inoculation with appropriate strains might be attributed to increase: 1) the symbiotic N fixation in the soil. 2) Level of free amino acids in the cell sap development and physiological enzymatic activity and growth rate of roots.

**Table (15): Effect of nitrogen fertilizer, Rhizobium inoculation and micronutrients on carbohydrates in seeds and stems for Sakha 3 variety [ 2008-2009 season].**

Treatments	Total carbohydrates		Carbohydrates fractions						
			Soluble sugars %						
			Total soluble		Non reducing sugars		Reducing sugars		
	Seeds	Stems	Seeds	Stems	Seeds	Stems	Seeds	Stems	
Control	39.79	29.32	5.03	2.96	4.43	2.54	0.60	0.42	
1N	61.12	43.40	10.42	4.81	9.75	3.82	0.67	0.99	
Zn+Mn+Fe	40.51	29.45	5.19	3.01	4.50	2.55	0.69	0.46	
1N+Zn+Mn+Fe	64.73	43.64	11.89	4.86	10.60	3.85	1.29	1.01	
Rhizobia	41.34	29.76	5.31	3.10	4.59	2.58	0.72	0.52	
1N+Rhizobia	67.46	43.91	12.56	4.88	11.26	3.87	1.30	1.01	
Zn+Mn+Fe+Rhizobia	42.45	30.10	5.48	3.13	4.74	2.60	0.74	0.53	
1N+Zn+Mn+Fe+Rhizobia	72.46	44.11	13.10	4.90	11.45	3.88	1.65	1.02	
L.S.D.	5%	2.16	1.33	1.11	0.33	1.65	0.71	0.22	0.17
	1%	3.19	1.96	1.64	0.49	2.44	1.04	0.32	0.25
C.V.		7.11	8.42	7.64	8.11	9.32	8.62	7.94	8.32

1N: 20kgN/fed.

Data also show that the best treatment in that design of fertilizer treatment including the addition mineral nitrogen fertilizer + micronutrients mixture + Rhizobium inoculation, which recorded the highly significant improvement in studded parameters in seeds. Since the percentage increases in none reducing sugars amounted to 17.44 and 18.26% for first and second season,

respectively. Such treatment bear the highly significant increase in reducing sugars in seeds by about 146.27 and 122.5% for first and second seasons at a respective order ,over nitrogen fertilizer only.

However, different combination treatments produced could not influence significantly in studded parameters in stems for two seasons as compared with nitrogen fertilizer, indicating more carbohydrate translocation through hydrolyzing more glucosides. Such trend might emphasize the increase in seed yield (Tables 4&5).

**Table (16): Effect of nitrogen fertilizer, Rhizobium inoculation and micronutrients on carbohydrate in seeds and stems for Sakha 3 variety [ 2009 – 2010].**

Treatments	Total carbohydrates		Carbohydrate fraction					
			Soluble sugars%					
	Seeds	Stems	Total soluble		Non reducing sugars		Reducing sugars	
Seeds			Stems	Seeds	Stems	Seeds	Stems	Seeds
Control	39.85	29.51	5.15	2.98	4.61	2.55	0.54	0.43
1N	61.27	44.01	10.71	4.95	9.91	3.85	0.80	1.10
Zn+Mn+Fe	40.64	29.83	5.33	3.06	4.70	2.57	0.63	0.49
1N+Zn+Mn+Fe	65.01	44.38	12.32	5.02	10.79	3.89	1.53	1.13
Rhizobia	41.49	29.93	5.45	3.13	4.80	2.59	0.65	0.54
1N+Rhizobia	67.94	44.78	12.94	5.05	11.52	3.94	1.42	1.11
Zn+Mn+Fe+Rhizobia	42.60	30.37	5.61	3.17	4.94	2.61	0.67	0.56
1N+Zn+Mn+Fe+Rhizobia	73.19	45.10	13.50	5.10	11.72	3.95	1.78	1.15
L.S.D. 5%	2.88	1.45	1.29	0.61	1.71	0.81	0.31	0.21
1%	4.25	2.14	1.90	0.90	2.52	1.20	0.46	0.31
C.V.8.25	8.25	8.61	9.11	9.20	7.15	7.67	8.41	8.29

1N: 20kg N/fed.

In brief, it is concluded that application of nitrogen fertilizer at a rate of 20kg N/fed. encouraged roots and vegetative growth, increased photosynthesis and enable the faba bean to absorb more nutrients and produced the highest seed yield. Also, it is fairly to mention that mineral nitrogen fertilizer combined with micronutrients as foliar spray at specific physiological growth stage of the plants is actually of great importance, especially in case of the experimental soil which suffering from nutrients deficient, whether such nutrients may become a limiting factor concerning soil productivity for different crops. Likewise micronutrients enhanced the activity of most metabolic processes in the plant, especially Mn and Fe in photosynthesis and Zn in the synthesis of the hormone IAA. Adding Rhizobium inoculation to the mineral nitrogen fertilizer not only can alleviate the harmful effects on environment and seeds composition, but also can improve seed quality and yield. We can not neglect the importance combined treatment of 20kgN/fed. plus Rhizobium inoculation and micronutrients which improve the physical and chemical soil media and help faba bean to sustain deficiency and provide seeds yield much greater extent more than nitrogen fertilizer added alone.



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## تأثير التسميد النيتروجيني والمخصبات الحيوية والعناصر الصغرى على محصول الفول البلدي جاكلين جرجس صادق

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### الملخص العربي

اجريت تجربتان حقليتان في مركز كفر الزيات - محافظة الغربية خلال موسمي ٢٠٠٨/٢٠٠٩ و ٢٠٠٩/٢٠١٠ بهدف تأثير إضافة مستويين من التسميد النيتروجيني (صفر و ٢٠ كجم نيتروجين/ فدان) والمغذيات الصغرى (حديد - زنك - منجنيز) رشاً على النباتات بالإضافة إلي التلقيح البكتيري باستخدام مثبت النيتروجين الريزوبيا إما في صورة مفردة أو متحدين علي الفول البلدي صنف سخا ٣. اضيفت المغذيات الصغرى في صورة مخلبية بمعدل ٣,٣ جم/ لتر لكل من عنصري الحديد والزنك و ١٥,١ جم/ لتر للمنجنيز في شكل محلول مغذي بحجم ٤٠٠ لتر/ فدان عن طريق الرش علي النباتات وذلك بعد ٤٥ و ٧٠ يوماً بعد الزراعة. وتشير النتائج التي تم الحصول عليها أن إضافة التسميد النيتروجيني بمعدل ٢٠ كجم / الفدان تفوق علي الرش بالعناصر الصغرى والتلقيح البكتيري في إحداث إستجابة معنوية في الأوزان الجافة للسيقان والأوراق والجذور في كلا الموسمين. زيادة مؤكدة إحصائياً في محتوى الأوراق من الكلوروفيل أ و ب والكاروتينات وذلك بعد ٦٠ يوم من الزراعة. زيادة معنوية لمحصول الفول البلدي ومفرداته حيث وصلت الزيادة النسبية ٣٩,٧٢ و ٤٠,٧٤ % في كل من الموسم الاول والثاني علي التوالي مقارنة بالكنترول. زيادة معنوية في محتوى البذور والسيقان من العناصر الكبرى ( نيتروجين - فوسفور - بوتاسيوم) والعناصر الصغرى ( حديد - زنك - منجنيز) بالإضافة الي البروتين الخام والكاربوهيدرات. كما تعظم هذه الزيادات معنوياً عندما يضاف التسميد النيتروجيني بالإضافة مع المغذيات الصغرى والتلقيح الحيوى (الريزوبيا) وذلك من خلال تنشيط وزيادة النمو الخضري ومن ثم إنتاجية المحصول كنتيجة لتيسر وسهولة حركة المغذيات المدروسة في إتجاه الجذور وامتصاصها ومثل هذه الظروف الجيدة قد انعكست بصورة إيجابية علي تركيز هذه المغذيات في محصول البذور لصنف سخا ٣.