

ENDOGENOUS PHYTOHORMONES IN RELATION TO WATER POTENTIAL OF POTATOES (*Solanum tuberosum*, L.) LEAVES AS AFFECTED BY BIO-AND MINERAL FERTILIZERS .

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ABSTRACT

IAA, GA₃ and cytokinin concentrations in potatoes leaves as well as their leaf water potential; Ψ_w were decreased whereas ABA and total soluble phenol were increased due to decreasing NPK dose to 50% from the recommended dose during the two growing seasons. On the other hand, Bio-fertilizers used NFB, PDB and SB individually or in combinations recorded a decrease in the concentrations of ABA and total soluble phenol whereas caused an increased in IAA, GA₃ and cytokinin as well as leaf Ψ_w , overall NPK doses.

The interaction treatments indicate that, inoculation of potato tubers with the three strains of used bacteria, increased IAA, GA₃ and cytokinin concentrations as well as leaf Ψ_w , whereas decreased ABA and total soluble phenol during the two growing seasons at any of the NPK doses used. Inoculation with nitrogen fixing bacteria (NFB) showed high levels in IAA and cytokinin as well as leaf Ψ_w compared with plants inoculated with phosphate- dissolving bacteria (PDB) or silicate bacteria (SB). On the other hand, inoculation with silicate bacteria (SB) recorded high GA₃ values compared with the other strains used.

Potato plants which received only half the rate of the recommended dose of NPK fertilizers interacted with the dual inoculation did accumulated more IAA, GA₃ as well as cytokinin and leaf Ψ_w whereas, decreased that of ABA and total soluble phenol in potato leaves than that in plants received the recommended NPK dose without inoculation. Moreover, plants grown under 75% NPK showed highest promoters concentrations and leaf Ψ_w values as well as less values from ABA and total soluble phenol.

Keywords: Potato, bio-and mineral fertilizers, IAA, GA₃, TSP cytokinin and leaf water potential.

INTRODUCTION

The biological activity of the endogenous growth substances in the presence of mineral nutrition was reported. It was found that gibberellin activity clearly increased with decreasing nitrogen levels and increased with plant age (Salama and Helaly, 1981). However, the levels of inhibitors showed a noticeable rise with the decrease in nitrogen supply. Torelli *et al.*, (2000) concluded that, P nutrition up to 112.5 kg/ha increased zeatin riboside and IAA content in leek; Alliaceae plants. Ghallab and Salem (2001) found that, the concentrations of indol-3-acetic acid, gibberellic acid and cytokinin of wheat leaves were increased with increasing the different NPK fertilizer levels up to 160 kg N + 150 kg P₂O₅ + 100 kg K₂O/fed.

The effects of bio-fertilizers on phytohormones were also reported by Fayez *et al.*, (1985) who recorded that, potato plants treated with *Azospirillum*, *Azotobacter* and *Pseudomonas* individually or in combinations were higher in their content from GA₃, cytokinin and IAA than the untreated

ones. Amara, Mervat and Nasr, Sohair (1995) noted that, the inoculation with *Pseudomonas fluorescenc* and *Rhizobium leguminosarum* increased the amounts of growth promoters and decreased level of growth inhibitors in wheat plants. Kawthar *et al.*, (2002) found that, treated potato plants with bio-fertilizers (*Azospirillum chroococcum*, *Azotobacter lipoferum*, *Bacillus megatherium* and *Bacillus circulans*) increased total growth promoters (cytokinins, gibberellic acid and indole acetic acid) with reduction of abscisic acid content to the half. Ghallab and El-Ghadban (2004) recorded that, using bio-fertilizers (*Bacillus polymexa*, *Azospirillum brasilense* and *Azotobacter chroococcum*) of marjoram plants; Lamiaceae increased the growth promoters IAA, cytokinins and GA₃. Sadik and Neseim (2004) found that, bacterial inoculation (*Azospirillum brasilense*, *Azotobacter lipoferum*) increased the endogenous IAA and cytokinins concentration in the shoots of wheat.

Ghallab and Salem (2001) studied the effects of the interactions between mineral fertilizers and some of bio-fertilizers and found that, inoculation with bio-fertilizers (Cerealin and Nemales) combined with NPK (160 kg N + 150 kg P₂O₅ + 100 kg K₂O/fed.) increased growth hormones (IAA, gibberellic acid and cytokinins) in wheat plants. Kawthar *et al.*, (2002) indicated that, the bio-fertilizers (*Azospirillum chroococcum*, *Azotobacter lipoferum*, *Bacillus megatherium* and *Bacillus circulans*) bacterial strains combined with 50% from the recommended dose of mineral fertilizer (125 kg N/fed) increased growth promoters; GA₃, IAA and cytokinins whereas, decreased that of ABA content. Ghallab and El-Ghadban, (2004) on marjoram plants; Lamiaceae, mentioned that, bacterial strains bio-fertilizer (*Bacillus polymexa*, *Azospirillum brasilense* and *Azotobacter chroococcum*) combined with 50% from recommended dose of mineral fertilizer (2.8 g N + 2.1 g P₂O₅ + 1.4 g K₂O/pot) increased growth promoters, GA₃, IAA and cytokinins with reduction of ABA content.

Changes in leaf Ψ_w alter the level of growth regulators, most noticeably ABA levels (Wahon, 1980 and Helaley *et al.*, 2007). Generally there is a threshold value of leaf Ψ_w below which ABA rapidly increases. For example in pot grown sorghum this was found to be about 0.8 to 1.0 MPa (Beardsell and Cohen, 1975). Diurnal changes of leaf Ψ_w and ABA levels occur in cotton and peach with more pronounced fluctuations under nonirrigated than under irrigated conditions (Xiloyannis *et al.*, 1980).

The purpose of this investigation was to examine the changes in leaf Ψ_w , IAA, cytokinins, GA₃, ABA and total soluble phenol in field potato plants in relation to bio-and mineral fertilization.

MATERIALS AND METHODS

Two field experiments were carried out at the Agriculture Experimental Station, Faculty of Agriculture, Mansoura University, Egypt during the two growing seasons of 2001/2002 and 2002/2003. Different rates of the recommended NPK mineral fertilizers and three strains of non-symbiotic bacteria as a bio-fertilizers sources of N, P and K were used.

Potatoes tubers; Spunta cv (imported from Holland) were used in the present investigation and obtained from Agric. Res. Center (ARC), Ministry

of Agric., Egypt. Tubers were divided to pieces, averaging approximately 50 g weight.

Soil samples and analysis:

Twenty surface samples (0-20 cm depth) were taken at ten different locations before the experimental design, air dried, grounded, mixed and kept in plastic bags for the analyses. The mechanical and chemical analyses of the soil used were carried out in the two growing seasons as described by Jackson (1973) and Page *et al.*, (1982) and presented in Table (1).

Table (1): The physiochemical properties of the experimental soil used during the two growing seasons of 2001/2002 and 2002/2003.

Season	1. Mechanical Analysis				Organic Matter	Calcium carbonate	PH (1:2.5 soil: water suspension)	Soil texture	
	Soil Fraction %								
	Coarse sand	Fine sand	Silt	Clay					
2001/2002	2.43	21.43	27.66	48.29	0.99	2.09	7.80	Clayey	
2002/2003	2.58	22.50	25.92	49.00	1.10	2.12	7.65		
	2. Chemical Analysis								
	EC dsm ⁻¹ soil paste extract at 25 C ^o	CATIONS (meq/L)				ANIONS (meq/L)			
		Ca ⁺⁺	Mg ⁺⁺	Na ⁺	K ⁺	HCO ₃ ⁻	CO ₃ ²⁻	SO ₄ ²⁻	Cl ⁻
2001/2002	1.31	5.33	4.22	10.40	0.39	2.44	-	7.68	10.63
2002/2003	1.45	5.21	4.11	10.99	0.37	2.07	-	7.80	11.00
	3. Nutrients Analysis								
	mg/100 g soil								
	N		P		K				
2001/2002	25.00		8.30		268.91				
2002/2003	33.00		8.50		335.10				

Experimental design:

Farm yard, manure has been added during soil preparation as organic fertilization at dose (40 m³/fed.). The experiments comprised of 24 treatments included three different rates of the recommended NPK mineral fertilizers used individually or in combinations with three strains of non-symbiotic bacteria as a bio-fertilizer sources for N, P and K. The experiments design used was a two factor randomized complete block system distributed as a split plot combined with five replications. Each plot was (14 m²) included four ridges, each five meters long and 70 cm apart; the distance between hills was 25 cm apart.

Bio-fertilizer treatments:

Three strains of non-symbiotic bacteria were used in the present investigation as bio-fertilizers sources; "*Azospirillum brasilense*", nitrogen-fixing bacteria (NFB), "*Pseudomonas fluorescens*", phosphate-dissolving bacteria (PDB) and "*Bacillus circulans*", silicate bacteria (SB) which able to release K from clay minerals (Monib *et al.*, 1984). The two former strains were obtained from Microbiol. Res.Dept., Soil, Water and Environ. Res. Inst., ARC, Giza, Egypt, whereas the third organism was obtained from Microbiol. Dept., Fac. of Agric., Mansoura Univ. Egypt. All bacterial strains were

multiplied in nutrient liquid broth and centrifuged then prepared again in suspension. Liquid broth cultures contains 5×10^8 , 9×10^8 and 2.15×10^8 cells/ml of NFB, DPB and SB, respectively.

Microbial inoculum treatments:

As recommended by the Pathology Dept. Ministry of Agric. Egypt, potato tubers pieces were sterilized with Vitavax Kapetan 1% at the rate of 1.25 kg/ton. and then inoculated with bacteria suspension, individually or in combinations directly before planting to form the following treatments:

- 1- Without bio-fertilizers.
- 2- Inoculation with *Azospirillum brasilense* (NFB).
- 3- Inoculation with *Pseudomonas fluorescens* (PDB).
- 4- Inoculation with *Bacillus circulans* (SB).
- 5- Inoculation with (NFB + PDB).
- 6- Inoculation with (NFB + SB).
- 7- Inoculation with (PDB + SB).
- 8- Inoculation with (NFB + PDB + SB).

Mineral fertilizer treatments:

As recommended by the Agric. Res. Center, Egypt, nitrogen fertilizer in the form of ammonium nitrate (33.3% N) was used at the dose of 180 kg N/fed. at three equal doses. The first was used after emergence (18-21 days from planting), whereas the second and third doses were applied before the 2nd and the 3rd irrigations respectively (31 and 46 days from planting). Calcium superphosphate (15.5% P₂O₅), as a source of phosphorus, at the dose of 75 kg P₂O₅ /fed., was added to the soil before planting and during soil preparation. Potassium sulphate (48 % K₂O) was used as a source of potassium at the dose of 96 kg K₂O/fed. at two times, the first half was added with the first addition of N-fertilizer, and the second with the third doses of N-fertilizer.

The mineral fertilizer treatments were used at the three following different rates:

- 1- 100% NPK from the recommended dose (control).
- 2- 75% NPK.
- 3- 50% NPK.

These treatments were used with or without the bio-fertilizer treatments.

Planting procedure:

The treated potato pieces were planted in the ridges at 12-15 cm depth (25 cm apart) on 12nd October, 2001 and 15th October, 2002 growing season, respectively. Irrigation was done immediately. All usual cultural practices of potatoes cultivation were carried out according to the procedures that recommended by the Ministry of Agric. Egypt. At the active growth period (75 days from planting) samples were taken to determinations of N,P and K levels as well as nitrate-reductase activity and free amino acids.

Endogenous phytohormones:

To ensure uniformity and comparable physiological state in all treatments, the 3th terminal fully expanded compound leaf was collected at approximately mid-day, since there is some evidence that hormonal levels in the leaves showed diurnal fluctuations (Esmail, 2005). The leaf was excised

from the intact plants at its base, then weighted and washed with distilled water prior to the extraction. The samples were plugged immediately after washing into Dewar flasks containing liquid nitrogen, freeze dried and stored at -70°C until hormone analysis.

Extraction of the endogenous plant hormones was carried out according to Sadeghian (1971) and modified by Helaly and Salama (1985). The methanolic extract was methylated and used for estimation of cytokinins (Palmer *et al.*, 1981), ABA (Davis and Addicott, 1972) as well as gibberellins (GA_3) and auxins as indole acetic acid (IAA) (Fales and Jaouni, 1973). The identification and the quantification of the endogenous phytohormones were carried out by Computer controlled Gas Liquid Chromatography (GLC); ATI-Unicum- 610 series equipped with flame ionization detector according to the method described by Vogel (1975). The fractionation of the phytohormones was conducted using a coiled glass column (1.5 m x 4.0 mm) packed with 1% OV-17. Gases flow rates were 30, 30, 330 ml/min for nitrogen, hydrogen and air, respectively. The peaks identification and quantification of the endogenous phytohormones were performed by using external authentic hormones and a Microsoft program to calculate the concentration of the identified peaks (MacMillan, 1970 and Gaskin and Zeevaart, 1973).

For total soluble phenoles determination, the ethanolic extract was prepared and used. The colorimetric methods of Folin-Denis as described by Swain and Hillis (1959) was employed.

Leaf water potential:

Fresh segments, 4x1.5 cm from the 3rd terminal fully expanded compound leaf, excluding the main veins are rapidly inserted into 6 ml screw capped polypropylene vials. The vials were immediately brought on the laboratory, fitted to a thermocouple psychrometer of a type designed by Kaul (1976) and leaf Ψ_w was estimated after an equilibration for 3 h at 25°C in a water bath. This equilibration time were sufficient to produce a steady response (Kannangara, 1982).

RESULTS AND DISCUSSION

The effects of mineral nutrients level (Table 2-4), over all bio-fertilizers treatments, show that IAA, GA_3 and cytokinins concentrations as well as leaf Ψ_w in potatoes leaves were decreased whereas ABA and total soluble phenol were increased due to decreasing NPK dose to 50% NPK level from the recommended dose. Moreover, it was found that the changes in leaf Ψ_w alter the level of growth substances examined which most noticeably ABA level. Generally, there is a threshold value of leaf water potential below which ABA rapidly increases. Moreover, ABA levels showed a significant negative correlation with Ψ_w . These results were confirmed with the previous results obtained for growth characters in the present investigation (Helaly and ramadan 2009). In this context, Helaly and Salama (1985) found that, the deficiencies of mineral nutrients, seem to affect ultimately the endogenous auxins. Apparently, biosynthesis and/or the activity of the enzymes required for normal biosynthesis of auxins was restricted and withdrawal of the mineral nutrient facilitated the production and/or accumulation of growth inhibitors and

ethylene. They added that, mineral deficiencies considerably decreased the endogenous levels of auxins while increase both plant inhibitors and endogenous ethylene in sunflower plants. These changes in plant hormones may exert an important influence on the physiological process such as assimilation, transport and others, including the fomentation of anatomical and physiological characteristics, the promotion or retardation of senescence and ripening as well as well as restriction and resistance to the unfavourable condition, consequently the yield and its components. Kamangara, (1982) reported that the contents of PGS especially ABA may depend on the condition, consequently the yield and its components. Kamangara, (1982) reported that the contents of PGS especially ABA may depend on the environmental condition, plant species and irrigation treatment. He added that, the small fluctuation in IAA levels do not bear a significant relationship to either the changes in leaf water potential or ABA level.

Table (2): Effects of mineral fertilizers on endogenous phytohormones as well as leaf water potential concentrations ($\mu\text{g/g}$ F.Wt.) in the leaves of potato plant [Average of the two growing seasons of 2001/2002 (S1)and 2002/2003 (S2)].

Treatments	Endogenous phytohormones					Leaf Ψ_w
	IAA	GA ₃	Cytokinins	ABA	Total soluble phenol	
Mineral NPK						
Control 100%	0.604	10.34	19.14	0.007	3.371	-1.331
75%	0.520	8.04	16.71	0.013	4.341	-1.164
50%	0.415	6.80	13.89	0.023	5.063	-0.902
F.test	**	**	**	**	**	**
LSD at 5%	0.005	0.02	0.03	0.001	0.013	0.012

Table (3): Effects of bio- fertilizers on endogenous phytohormones as well as leaf water potential concentrations ($\mu\text{g/g}$ F.Wt.) in the leaves of potato plant [Average of the two growing seasons of 2001/2002 (S1)and 2002/2003 (S2)].

Treatments	Endogenous phytohormones					Leaf Ψ_w
	IAA	GA ₃	Cytokinins	ABA	Total soluble phenol	
Bio-fertilizer						
Without	0.383	8.03	6.57	0.040	5.230	-0.623
NFB	0.432	8.27	13.92	0.013	4.379	-1.153
PDB	0.409	8.15	9.22	0.017	4.514	-0.963
SB	0.403	8.40	8.94	0.020	4.676	-0.792
NFP+PDB	0.607	8.41	23.34	0.006	3.733	-1.370
NFB+SB	0.513	8.64	23.32	0.007	3.977	-1.200
PDB+SB	0.446	8.53	20.16	0.008	4.113	-1.177
NFB+PDB+SB	0.909	8.70	27.15	0.004	3.446	-1.783
F.test	**	**	**	**	**	**
LSD at 5%	0.004	0.03	0.03	0.001	0.022	0.019

Thus, it seems unlikely that level of IAA will be regulated by ABA *in vivo* by decarboxylation or by inhibition of IAA transport (Kaldewey *et al.*, 1974) Bestford *et al.*, (1991) stated that, mineral constituents within the plant tissues in addition to DNA and RNA controlled the phytohormone metabolism and consequently other morphogenetic effects and growth. Moreover, it was found that, the positive effects of mineral fertilizer on endogenous phytohormones may be attributed to their effects on increase the merisimatic activity of plant tissues and their constituents from GA₃ and auxins (Helaly *et al.*, 1985; Maier *et al.*, 1995). Marschner (1995) added that, both P and K play an important role in biosynthesis of plant hormones.

On the other hand, the estimated phytohormones except that of ABA and total soluble phenol (IAA, GA₃ and cytokininis) as well as leaf water potential; Ψ_w gradually increased in plants inoculated with each of the bio-fertilizers used, overall the NPK doses during the two growing seasons (Table 2-4). However, ABA and total soluble phenol were decreased. Such results confirm the efficiency of the dual inoculation with all bio-fertilizers used as compared with the single ones under the same level of NPK fertilizers. The promotive effects of bio-fertilizers on phoytohormones and leaf water potential; Ψ_w may be attributed to the effects of the three bacterial strains used on their ability to release and produce growth promotive substances such as IAA, GA₃ and cytokinins (Ghulam *et al.*, 1998). Moreover, Al-Moshileh and Mofteh (2005) attributed the enhancing effect of the bio-fertilizers on endogenous phytohormones to their effects on the formation of a big root system.

The interaction treatments (Table 4) indicate that, inoculation of potato tubers with the three strains of used bacteria, increased IAA, GA₃ and cytokinins concentrations as well as leaf water potential; Ψ_w , whereas decreased ABA and total soluble phenol during the two growing seasons at any of the NPK doses used. Inoculation with nitrogen fixing bacteria (NFB) showed high value regarding IAA and cytokinins as well as leaf water potential; Ψ_w compared with plants inoculated with phosphate- dissolving bacteria (PDB) or silicate bacteria (SB). Moreover, it was found that, inoculation with silicate bacteria (SB) showed high values of GA₃ compared with the other used bacterial strains. Similarly, plants inoculated with either of NFB+PDB or mixed with the three bacterial strains used showed high values in their concentrations of IAA and cytokinins as well as leaf water potential; Ψ_w , and less values regarding ABA and total soluble phenol. In addition, plants inoculated with either NFB+SB showed high value in GA₃ and leaf water potential; Ψ_w . The obtained results clearly show the positive responses in the promoters hormonal concentrations and leaf water potential; Ψ_w which attained their maximum when the recommended dose (100% NPK ; control) inoculation with NFB+PDB+SB. Compared with the control (100% NPK recommended dose), data in the same tables indicate that, the addition of mineral fertilizers increased all the above mentioned growth promoters, while decreased ABA and total soluble phenol concentrations.

Table (4): Effects of mineral and/or bio-fertilizers on endogenous phytohormones as well as leaf water potential concentration ($\mu\text{g/g F.Wt.}$) in the leaves of potato plant grown in the two growing seasons of 2001/2002 (S1) and 2002/2003 (S2).

Treatments		Endogenous phytohormones										Leaf water potential	
		IAA		GA ₃		Cytokinins		ABA		Total soluble phenole		Leaf Ψ_w	
		S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2
M-Mineral NPK	B-Bio-fertilizer												
Control 100%	Without	0.460	0.498	9.32	9.80	7.76	8.36	0.018	0.016	4.622	4.413	-0.700	-0.900
	NFB	0.470	0.522	10.15	10.51	16.56	17.22	0.007	0.007	3.604	3.398	-1.300	-1.500
	PDB	0.466	0.517	10.06	10.49	10.44	11.56	0.008	0.008	3.915	3.407	-1.100	-1.200
	SB	0.463	0.513	10.32	10.80	9.91	10.52	0.010	0.008	4.008	3.712	-0.900	-1.100
	NFB+PDB	0.652	0.708	10.22	10.62	27.08	27.58	0.004	0.005	3.028	2.366	-1.500	-1.100
	NFB+SB	0.524	0.672	10.57	10.97	26.42	26.97	0.002	0.003	3.217	2.705	-1.200	-1.900
	PDB+SB	0.478	0.543	10.52	10.93	23.16	23.92	0.003	0.003	3.405	2.922	-1.400	-1.300
	NFB+PDB+SB	1.060	1.112	10.61	11.01	29.08	29.61	0.001	0.001	2.910	2.308	-1.900	-2.100
Mean		0.572	0.636	10.13	10.55	18.80	19.47	0.007	0.006	3.589	3.154	-1.250	-1.387
75%	Without	0.339	0.353	8.06	8.52	6.03	6.52	0.040	0.035	5.475	5.219	-0.650	-0.630
	NFB	0.438	0.466	7.60	8.11	14.26	14.86	0.014	0.012	4.622	4.417	-1.100	-1.200
	PDB	0.374	0.407	7.51	7.89	9.00	9.49	0.016	0.014	4.698	4.501	-0.900	-1.000
	SB	0.360	0.400	7.71	8.20	8.96	9.45	0.021	0.018	4.819	4.604	-0.700	-0.810
	NFB+PDB	0.633	0.662	7.76	8.26	23.14	23.60	0.005	0.005	3.914	3.666	-1.440	-1.840
	NFB+SB	0.506	0.536	7.95	8.45	22.87	23.38	0.006	0.005	4.118	4.013	-1.080	-1.120
	PDB+SB	0.456	0.482	7.81	8.31	20.07	20.62	0.007	0.008	4.304	4.108	-1.180	-1.160
	NFB+PDB+SB	0.940	0.968	8.02	8.49	27.11	27.92	0.003	0.003	3.570	3.307	-1.860	-1.960
Mean		0.506	0.534	7.80	8.28	16.43	16.98	0.014	0.013	4.452	4.229	-1.114	-1.215
50%	Without	0.314	0.349	5.93	6.53	5.17	5.56	0.069	0.062	6.040	5.612	-0.420	-0.440
	NFB	0.331	0.366	6.33	6.92	10.09	10.51	0.020	0.020	5.330	4.904	-0.900	-0.920
	PDB	0.328	0.359	6.18	6.80	7.22	7.63	0.029	0.028	5.444	5.122	-0.750	-0.830
	SB	0.324	0.356	6.28	7.11	7.19	7.59	0.032	0.028	5.613	5.299	-0.600	-0.640
	NFB+PDB	0.477	0.512	6.51	7.11	19.68	20.13	0.009	0.010	4.908	4.516	-0.600	-1.060
	NFB+SB	0.392	0.448	6.64	7.25	19.33	19.78	0.012	0.010	5.116	4.588	-0.920	-0.980
	PDB+SB	0.342	0.375	6.50	7.11	16.38	16.82	0.013	0.012	5.237	4.702	-1.000	-1.020
	NFB+PDB+SB	0.670	0.703	6.74	7.33	24.34	24.82	0.006	0.007	4.460	4.119	-1.400	-1.480
Mean		0.397	0.433	6.49	7.11	13.68	14.11	0.024	0.022	5.269	4.858	-0.884	-0.921
LSD at 5% for: SxM		0.007		0.03		0.04		0.001		0.018		0.016	
SxB		0.006		0.04		0.04		0.001		0.030		0.027	
MxB		0.007		0.05		0.05		0.001		0.037		0.033	
SxMxB		0.010		0.07		0.07		0.002		0.052		0.048	

Another important finding is that potato plants which received only half the rate of the recommended dose of NPK fertilizers interacted with the dual inoculation did accumulated more IAA, GA₃ and cytokinins and showed less leaf water potential; Ψ_w whereas, decreased that of ABA and total soluble phenol in their leaves than that in the plants receiving the recommended NPK dose without inoculation. Moreover, plants grown under 75% NPK showed highest promoters values concentrations and leaf water potential; Ψ_w and lessees values from ABA and total soluble phenol. Therefore, the data indicate that, plants received mixed strains of used bacteria (NFB+PDB+SB) plus 75% NPK (from the recommended dose) showed high values regarding promoters (IAA, GA₃ and cytokinins as well as leaf water potential; Ψ_w) and low values of ABA concentrations and represented the best treatment in this respect.

These results strongly confirm the previous conclusion drawn with the different other constituents; carbohydrates, nutrients (Helaly and Ramadan, 2009). The promotive effects of the bio-fertilizers, especially the dual treatment with NFB+PDB+SB may be attributed to the production of growth promotive substances from rhizospheric microorganisms such as IAA

and GA₃ (Ghulam, *et al* 1998). In this respect, *Azospirillum* produce several plant hormones in liquid cultures, mainly IAA (Fallik *et al* 1989). Other hormones detected at much lower concentration, indole 3-butyric acid (IBA) (Fallik *et al* 1989), indole 3-ethanol and indole 3-methanol (Crozier *et al* 1988), several gibberellins (Bottini *et al* 1989), Abscic acid; ABA (Kolb and Martin 1985) and cytokininis (Horemans *et al* 1986 and Tien *et al* 1979).

Several investigators showed that, application of external hormones either synthetic or purified from bacterial culture, to seedlings completely reproduced the effects of *Azospirillum* caused on root development and morphology (Kucey, 1988 and Zimmer and Bothe 1988). In particular, root length (Morgenstern and Okon, 1987), more root hairs (Kapulnik *et al.*, 1985), and branching root hairs (Jain and Patriquin, 1984) or produced more lateral roots (Barbieri *et al.*, 1986) and enhanced the rate of cell division and differentiation in meristimatic tissues (Fallik *et al.*, 1989).

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الهرمونات النباتية والجهد المائي في أوراق نباتات البطاطس تحت تأثير التسميد الحيوي والمعدني.

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أدى النقص فى نسبة التسميد المعدنى ،الى 50% من الجرعة الموصى بها ، الى نقص فى تركيزات كل من اندول حمض الخليك والجبريلينات والسيتوكينينات بينما زادت تركيزات حامض الأبسيسيك والفينولات الذاتية الكلية فى اوراق نبات البطاطس خلال موسمى الزراعة. وعلى العكس من ذلك فقد أدى استخدام السماد الحيوى الى نقص تركيز حامض الأبسيسيك والفينولات الذاتية الكلية ، وزيادة تركيز اندول حمض الخليك والجبريلينات والسيتوكينينات والجهد المائي فى الأوراق.

ولقد أوضحت معاملات التفاعل بين المعاملات ، ان البطاطس الملقحة بمخلوط السلالات البكتيرية المستخدمة الثلاث كانت أعلى تركيزاً فى محتواها من اندول حمض الخليك والجبريلينات والسيتوكينينات والجهد المائي وأقل تركيزاً من حامض الأبسيسيك والفينولات الذاتية الكلية ، تحت أى نسبة من التسميد المعدنى المستخدم ، خلال موسمى النمو. وقد سجلت معاملة التلقيح بالبكتيريا المثبتة للنيتروجين ، أعلى القيم فى تركيز اندول حمض الخليك والسيتوكينينات والجهد المائي ، مقارنة بالبكتيريا المثبتة للفوسفات ، أو البكتيريا الميسرة لإطلاق البوتاسيوم. وكانت الأخيرة الأكثر تأثيراً على تركيز الجبريلينات.

كما أدى التفاعل بين التسميد المعدنى ، عند إستخدام نسبة 50% من الجرعة الموصى بها ، مع التلقيح البكتيرى ، بأى من السلالات الثلاث المستخدمة ، الى زيادة تركيز اندول حمض الخليك والجبريلينات والسيتوكينينات ، ونقص تركيز حامض الأبسيسيك والفينولات الذاتية الكلية ، بأوراق النبات. وقد أعطت النباتات التى نمت تحت نسبة 75% من الجرعة الموصى بها من السماد المعدنى مع التلقيح البكتيرى أعلى القيم فى هذا الشأن من حيث تركيز المنشطات والجهد المائي وأقلها فى حمض الأبسيسيك والفينولات الذاتية الكلية.