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Improving Growth, Yield and Nutritional Status of Pea Plants by Using Amino Acids under Different Levels of Nitrogen Fertilization

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Abstract: Two field experiments were carried out at the village of Faqous - Sharkia Governorate, Egypt, during successive winter seasons of 2019 and 2020. The experiment was designed to investigate the effect of three rates of nitrogen fertilization (20, 30 and 40 Kg N fed⁻¹) and foliar spray with four amino acids concentrations (0, 1, 1.5 and 2.5%) on growth, yield and its quality of pea (Lincoln cv.) plants under drip irrigation. Through the obtained results, it was found that spraying amino acids on pea plants greatly helped on improving the growth of plants, which were not affected as a result of reducing the added amount of nitrogen fertilization. The increase of nitrogen fertilization with amino acid sprayed in different concentrations led to a great improvement in yield characteristics of pea plants, especially with the high rate of nitrogen fertilization $(40 \text{ Kg N fed}^{-1})$ with the spraying of amino acids at the highest concentration (2.5%). The best treatment, which gave the highest values for the weight, length and width of the pea pods, was when spraying amino acids at a concentration of 2.5% with the addition of a high rate of nitrogen fertilization of 40 kg N fed⁻¹, in The same experimental treatment (40 Kg N fed⁻¹ + 2.5 % amino acid), which gave the highest values for the yield characteristic's, was the same treatment that contributed to improving the quality of pea yield (weight, length and width of pea pods) during the two successive seasons. As well as, the effect of increasing the foliar application treatment with amino acids from zero to 2.5 % was more effective and significant in increasing the nitrogen, phosphorous and potassium content in pea grain, especially with increasing the rate of nitrogen fertilization. Using amino acids as an alternative to the lack of nitrogen fertilization quantity did not decrease the growth, yield and quality of pea plants and can reduce the use of nitrogen fertilizers, which helpsin reducing environmental pollution and decreasing total cost.

Key words: Nitrogen fertilization • Amino acid • Pea plants • Growth • Yield • Nutritional state

INTRODUCTION

Pea (*Pisumsativum* L.) is one of the most important leguminous vegetable crops grown during winter season in Egypt for local consumption and exportation. The pods of pea contain a great amount of protein and carbohydrates [1]. The cultivation of pea plants preserve soil fertility meanwhile biological nitrogen fixation in organization with symbiotic rhizobium prevailing in its root nodules and consequently plays a pivotal role in enhancing prospective agriculture [2].

Nitrogen (N) is often the most limiting factor in agricultural system productivity [3]. Although the application of N fertilizer may increase yields, but it can

rise the environmental pollution through run-off into surface waters or leaching into ground waters [4]. A large amount of N added to soils are wasted because plants have low nitrogen use efficiency in using commercial N fertilizer [5]. Biological nitrogen fixation by legumes is an important pathway for N supplement and for improving soil fertility [6]. Using legumes in agricultural production systems can reduce N fertilizer dependence [7] and provide economic and environment benefits [8].

Amino acids are substances that promote plant growth and enhance plant quality [9]. Amino acids are not only getting popular for mitigating injuries caused by abiotic stresses [10], but also serve as hormone precursors [11]; signaling factors of different physiological progressions [12]; and regulators of nitrogen uptake [13]. Better root development supported by the addition of amino acids can boost nitrogen fixation, which induces an enhancement in root surface for nutrient uptake [14] and [15].

Thus this research aimed to study the response of pea plants growth, yield and nutritional status to different foliar concentrations of amino acids under different levels of nitrogen fertilization.

MATERIALS AND METHODS

Two field experiments were carried out at the village of Faqous - Sharkia Governorate, Egypt, during successive winter seasons 2019 and 2020. The experiment was designed to investigate the effect of three rates of nitrogen fertilization (20, 30 and 40Kg N fed⁻¹) as ammonium sulfate (20%N) and foliar spray with four amino acids concentrations (0, 1, 1.5 and 2.5 %) on

Table 1: Some physical and chemical properties of the soil used

growth, yield and its quality of pea plants(cv. Lincoln) under drip irrigation system. The amount of phosphorous and potassium fertilizers were added as recommended by the Ministry of Agriculture and Reclaimed Soil. Physical and chemical properties of the experimental soil are shown in Table (1) using the standard procedures outlined by Cottenie [16].

The treatments were arranged in a split plot design with three replicates. Where, nitrogen fertilizer rates were randomly arranged in the main plots and the concentrations of amino acid wererandomly distributed in the sub plots. Each plot (15 m^2) contained six dripper lines with 6 m length and 50 cm between each two dripper lines. Pea seeds were inoculated with root nodules bacteria and sown on 25th October in both seasons on both sides of dripper lines on hills at 7 cm between each two hills. Foliar application with amino acids was added at 35 days after sowing. The chemical composition of amino acids are presented in Table (2).

Soil property	Value	Soil property	Value
Particle size distribution %		pH (1:2.5 soil suspension)	8.10
Sand	92.65	EC (dS m ⁻¹), soil paste extract	2.00
Silt	5.07	Soluble ions (mmol L^{-1})	
Clay	2.28	Ca ⁺⁺	8.02
Texture	Sandy	Mg^{++}	3.23
CaCO ₃ %	2.20	Na ⁺	3.92
Saturation percent %	22.50	K+	0.91
Organic matter%	0.11	CO_3^-	-
Available N (mg kg ⁻¹)	20.2	HCO ₃ -	2.20
Available P (mg kg ⁻¹)	3.50	Cl	3.98
		SO_4^-	9.90
Available K (mg kg ⁻¹)	66.4	CEC (cmol kg^{-1})	7.00

Amino acids	Content (%)
Isoleucine	0.6
Leucine	0.4
Lycine	1.2
Methionine	0.9
Phenylalanine	0.8
Theroninie	0.9
Valine	1.0
Histidine	0.3
Cysteine	0.1
Alanine	1.7
Aspartic	1.8
Serine	0.8
Glycine	1.0
Tyrosine	0.6
Proline	0.8
Glutamine	2.1
Argnine	1.0
Total amino acids	16

Data Recorded

Plant Growth Measurements: A random sample of five plants from each experimental treatment was taken at 60 days after sowing for measuring the vegetative growth parameters; i.e., plant height (cm.), number of leaves per plant. After that, the plants from each treatment were dried at 70°C till constant weight, then total dry weight/plant (g) was recorded.

Yield and its Components: A random sample of twenty pods from each experimental treatment, from the second harvest, was randomly taken to determine average pod weight(g), number of pods per plant, number of seeds per pod, weight of 100 seeds (g), pod length, pod width and total yield.

Seed Chemical Constituents: Seeds from the second harvest were dried in an oven at 60°C till constant weight. Seeds were finely grinded anddigested, then nitrogen, phosphorus and potassium percentages were determined (on dry weight basis) according to the methods advocated by [17], [18] and [19], respectively.

Total Carbohydrate (%): It was determined (on dry weight basis) according to the methods described by Dubois*et al.* [20].

Total Protein (%): It was determined as nitrogen content and converted to protein % by multiplying N% by 6.25.

Statistical Analysis: All the obtained data were subjected to statistical analysis of variance according to Snedecor and Cochran [21] and means separation were done by L.S.D.at 0.05 level of probability.

RESULTS AND DISCUSSION

Data presented in Table (3) showthe effect of spraying pea plants with different concentrations of amino acids under several levels of nitrogen fertilization on the vegetative growth characters. It was found that the increase of foliar application concentration of amino acids was accompanied by a significant increase in growth characteristics i.e., plant height, number of leaves and plant dry weight of pea plants with the increase of nitrogen fertilization levels. The use of spraying pea plants with amino acids reduced the negative effect caused by the lack of the added amount of nitrogen fertilization on plant growth and therefore spraying amino acids with the appropriate concentration, reduced the amount of nitrogen fertilization and raised the efficiency of nitrogen fertilization use and also allowed to reduce pollution resulting from the excessive addition of nitrogen fertilizers.

Several suppositions have been proposed to expound the role of amino acids in plants growth. Evidence suggests several alternative routes of indole acetic acid synthesis in plants, all starting from amino acids [22]. Amino acids can influence the physiological activities in plant growth and development such as exogenous application of amino acids have been reported to modulate the growth of tomato in plastic greenhouse [23]. Saeed et al. [24] found that treatments of amino acids significantly improved growth parameters of shoots and fresh weight as well as pod yield of soybean plants. Näsholm et al. [25] reported thatamino acids are crucial to activating cell growth, act as buffers, provide a source of carbon and energy and protect the cells from ammonia toxicity. The implementation of amino acids can spur the performance of plant [26]. Ghaith and Galal [27] reported that, spraving pea plants with mixture of aminoacid at 100 ppm significantly increased plant growth characters.

Through the results obtained from Table (4), which showed the response of pea plants yield characteristic's to spraving with amino acids under different levels of nitrogen fertilization. It was found that the increaseof amino acids foliar application concentration reduced the effect of using low levels of recommended nitrogen fertilization, which proved that the spraving of amino acids compensated the decrease in the amount of added nitrogen fertilizer, which confirmed the effective role of amino acids and their stimulating effect on plant growth and increasing plant yield. Also, the increase of nitrogen fertilization with amino acid foliar application with different concentrations led to a great improvement of pea plants yield characteristics, especially with the high rate of nitrogen fertilization (40 Kg N fed⁻¹) with the spraying of amino acids at the highest concentration (2.5%).

Amino acids can improve and raise the efficiency of the added fertilizer, as well as improve the absorption of water and nutrients and stimulate and raise the rates of photosynthesis and thus increase the yield [28]. Khalilzadeh *et al.* [29] reported that, spraying amino acid significantly increased the seed weight, seed yield and biological yield of bean plants.

The quality of pea pods continued to be improved with different concentrations of amino acids foliar application under various levels of nitrogen fertilization during the two successive seasons as shown in Table (5).

Treatments	1st season			2 nd season			
N fertilization Kg N fed ⁻¹	Amino acids %	Plant height cm	No. of leaves	Dry weight plant ⁻¹ g	Plant height cm	No. of leaves	Dry weight plant ⁻¹ g
20	0	28.5	9.77	9.54	26.6	9.76	9.66
	1	33.8	11.1	10.9	35.0	10.9	11.2
	1.5	39.5	12.8	13.1	38.7	12.7	13.9
	2.5	43.7	14.2	14.2	41.1	13.9	14.6
30	0	30.6	10.8	9.83	31.5	10.8	9.61
	1	35.7	13.8	11.8	34.2	13.6	11.2
	1.5	44.7	15.0	14.2	41.9	14.9	13.5
	2.5	48.1	17.2	16.4	46.1	16.9	15.0
40	0	37.0	12.5	11.7	35.8	12.3	11.0
	1	38.4	15.3	14.7	37.2	15.1	15.0
	1.5	49.0	16.7	16.8	48.1	16.6	17.0
	2.5	50.0	17.3	17.2	52.0	17.2	17.8
LSD _{0.05 %}		0.41	0.22	0.50	0.41	0.22	0.50

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Table 3: Effect of nitrogen	fertilization rates ar	nd amino acid	concentration of	on pea plan	ts growth

Table 4: Effect of nitrogen fertilization rates and amino acid concentration on pea yield 1season 2nd season Number of Treatments Number of N fertilization Kg N fed-1 Amino acids % Weight of 100 se Pods plant Weight of 100 see Pods plant Seeds pod Total yield (ton fed) Seeds pod Total yield (ton fed) 20 0 0.839 34.78 10.33 6.333 0.844 34.77 10.34 6.312 1 0.967 35.33 10.80 6.622 0.972 35.34 10.79 6.612 6.692 0.973 0.981 1.5 36.00 11.00 6.700 35.80 11.01 2.5 0.982 37.66 11.50 6.955 0.990 37.59 10.91 6.954 30 0 0.991 38.68 10.92 7.062 1.000 38.67 38.67 7.061 1.150 39.70 7.555 39.80 39.71 7.560 1 11.12 1.155 1.5 1.270 41.22 11.82 8.200 1.279 41.23 41.21 8.195 1.422 2.5 44.50 12.92 8.466 1.460 44.42 44.45 8.467 48.63 40 0 1.133 12.00 7.500 1.128 47.62 48.61 7.521 1.400 50.16 13.10 8.622 1.388 50.06 50.12 8.621 1.5 1.550 53.23 13.80 9.000 1.500 53.13 53.22 9.000 2.5 1.680 56.66 14.22 9.500 1.690 56.59 56.59 9.451 LSD₀₀ 0.160 1.150 0.860 0.511 0.160 1.150 0.860 0.511

The length, width and weight of the pea pod increased significantly with the increase in nitrogen fertilization from 20 to 40 kg N fed⁻¹, with an increase in the concentration of amino acid foliar application from zero to 2.5%. The best treatment, which gave the highest values for the weight, length and width of the pea pods, was when amino acids were sprayed at the concentration of 2.5% with the addition of a high rate of nitrogen fertilization of 40 kg N fed⁻¹ duirng the two growing seasons.

The requirement of amino acids in base quantities is well famed as a mean to increase yield and quality of all crops [26]. Ghaith and Galal [27] reported that, spraying pea plants with mixture of amino acid at 100 ppm significantly increased total pods yield and pods quality.Mohsen and Jasim [30] reported that foliar fertilizer with amino-acids caused a significant increase in plant pods number, pod seeds number and total dry seed yield of pea plants.

Data presented in Table (6) show the effect of amino acid foliar application on protein and carbohydrate content of pea seeds under different levels of nitrogen fertilization. The spraying of amino acids increased the content of pea grain protein and carbohydrates, which was evident with the increase in nitrogen fertilization rates. The high values obtained from protein and carbohydrates content inside pea seeds are obtained at the high concentration of amino acids (2.5 %) foliar application with the addition of high fertilization rate of nitrogen (40 kg N fed⁻¹) in both growing seasons.

Kandi et al. [31] who reported that spraying wheat plants withamino acids limited the rapidity of the nutrient deficiency asthey are absorbed easily and used directly for proteinsynthesis, where Glycine is a major component of buildingchlorophyll within plant as it helps to increase chlorophyllconcentration leading to the highest degree of photosynthesis.Some amino acids are characterized by containing sulfur as amajor element participating in many plant proteins andkeeping the stereoscopic structure or forming the active sites of enzymes [32]. Spraying amino acids on plants increase both vegetative growth and leaf area as they are easily absorbed by leaves to create protein and increase the leaf chlorophyll content resulting in improving the crop yield and quality [33]. EL-Bassiouny [34] found an increment in protein content by 18.52 % resulted from spraying 50 mg L^{-1} tryptophan, in addition

		1 st season			2 nd season		
Treatments			Pod (cm)			Pod (cm)	
N fertilization Kg N fed ⁻¹	Amino acids %	Pod weight (g)	Length	Width	Pod weight (g)	Length	Width
20	0	2.73	6.50	0.90	2.81	6.60	0.94
	1	3.43	6.82	0.95	3.52	6.89	0.97
	1.5	3.50	6.92	1.00	3.62	7.00	1.05
	2.5	3.72	7.00	1.08	3.81	7.22	1.09
30	0	3.10	7.25	1.00	3.18	7.56	1.08
	1	3.72	8.03	1.12	3.91	8.32	1.26
	1.5	4.11	8.88	1.24	4.20	9.15	1.29
	2.5	4.56	9.30	1.37	4.60	9.48	1.45
40	0	5.50	9.69	1.19	5.56	9.77	1.21
	1	5.66	9.82	1.22	5.87	9.99	1.26
	1.5	6.42	10.1	1.30	6.45	10.4	1.33
	2.5	7.05	10.5	1.32	7.10	10.9	1.35
LSD _{0.05 %}		0.16	0.40	0.16	0.16	0.40	0.16

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Table 5: Effect of nitrogen fertilization rates and amino acid concentration on pea yield quality

Table 6: Effect of nitrogen fertilization rates and amino acid concentration on protein and carbohydrate of pea grain

		1 st season		2 nd season	
Treatments		Protein	Carbohydrate	Protein	Carbohydrate
N fertilization Kg N fed ⁻¹	Amino acids %		- %		- %
20	0	13.8	40.1	14.1	40.2
	1	14.5	41.4	15.3	41.0
	1.5	14.7	42.0	15.6	42.0
	2.5	15.9	42.8	16.2	41.8
30	0	14.8	40.1	14.9	40.3
	1	16.6	41.8	17.2	41.6
	1.5	17.8	42.1	18.0	42.2
	2.5	18.1	43.0	19.1	43.1
40	0	17.9	41.2	17.6	41.4
	1	19.3	42.0	20.4	42.5
	1.5	20.7	44.7	21.6	44.6
	2.5	21.3	46.0	22.1	46.1
LSD _{0.05 %}		0.33	4.60	0.33	4.60

Table 7: Effect of nitrogen fertilization rates and amino acid concentration on N, P and K content of pea grain

		1 st season		2 nd season	2 nd season		
Treatments		 N	Р	К	 N	р	К
N fertilization Kg N fed ⁻¹	Amino acids %		•			- 0/	
20	0	1.56	0.34	1.31	1.57	0.33	1.32
	1	1.68	0.35	1.33	1.71	0.35	1.35
	1.5	1.71	0.35	1.40	1.72	0.36	1.39
	2.5	1.74	0.37	1.41	1.80	0.38	1.42
30	0	1.89	0.35	1.39	1.86	0.35	1.37
	1	2.01	0.36	1.46	2.00	0.36	1.44
	1.5	2.18	0.38	1.49	2.17	0.39	1.49
	2.5	2.26	0.41	1.50	2.29	0.40	1.51
40	0	2.22	0.40	1.51	2.20	0.38	1.48
	1	2.45	0.41	1.62	2.44	0.40	1.61
	1.5	2.52	0.42	1.74	2.50	0.41	1.73
	2.5	2.60	0.42	1.81	2.59	0.42	1.79
LSD _{0.05 %}		0.07	0.03	0.04	0.07	0.03	0.04

carbohydrate percentage increase by 64% when 100 mg L^{-1} was used.Ruta *et al*, . [35]Confirmed that spraying liquid fertilizer of nitrogenamide containing 3% amino acids at the milk maturitystage increased the protein content in grains from 0.52% to0.87%.

Resultspresented in Table (7) show the improvement of pea seeds content of macro nutrients (N, P and K) which were estimated in pea seedsthrough different experimental treatments. The effect of increasing the foliar application concentration of amino acids from zero to 2.5 % was more effective and significant in increasing the nitrogen, phosphorous and potassium content in peaseeds, especially with increasing the rate of nitrogen fertilization than other concentrations. It was found that spraying amino acids can contribute to reducing the added amounts of nitrogen fertilizers without causing a shortage of nutrients within the plant tissues.

Application of amino acids can modulate N uptake and its assimilation; this phenomenon is mediated by enzymes engaged with N assimilation [36] and [37]. Additionally, application of amino acids was also found to increase K^+ in plants either in the presence of salt stress or without salt application [28]. Teixeira *et al.* [38] showed that foliar implementation with amino acids had different effects on soybean plants. An amino acid applied individually acts as a signaling component causes efficient nutrients uptake. Abo Sedera *et al.* [39] revealed that spraying strawberry plants with amino acids significantly increased total nitrogen, phosphorus and potassium in plant foliage compared with control treatment.

CONCLUSION

The use of amino acids as a spray on pea plants that were fertilized with different rates of nitrogen fertilization led to a significant improvement in the growth and yield of peas as well as its content of nutrients. It can also be replaced by amino acids as an alternative to the decreasing amounts of nitrogen fertilization, thus lowering production costs as well as reducing environmental pollution.

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