

The Integrated Use of Bio-inoculants and Chemical Nitrogen Fertilizer on Growth, Yield and Nutritive Value of Two Okra (*Abelmoschus Esculentus*, L.) Cultivars

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Abstract: Two field experiments were conducted during the two seasons of 2005 and 2006 at the experimental station of National Research Centre in Nubaria (North Egypt). The aim of these experiments is to study the integrated effect of bio-inoculants and chemical nitrogen fertilizer on okra plant growth, pods yield as well as its physical and chemical properties. Two chemical nitrogen fertilizer rates as ammonium sulphate (the recommended rate and half of it) and two bio-inoculants, i.e. *Azospirillum* and *Azotobacter*, all of these treatments were applied for two okra cultivars, i.e. Balady and Eskandarani. The important obtained results are as following: The cultivars of okra plant had no significant effect on the plant growth parameters, pods yield as well as its physical and chemical measurements, but the application of two bacterial strains, i.e. *Azospirillum* and/or *Azotobacter* enhanced the plant growth parameters and pods yield and its quality in both two cultivars. The fertilization of okra plant by chemical fertilizer gained less plant growth values as well as yield of pods and its physical and chemical properties if compared with that plants which fertilized by 50 % of the recommended chemical nitrogen fertilizer plus the inoculation by bio-fertilizer. Moreover, the better plant growth was associated with *Azospirillum* bacteria as bio-fertilizer.

Key words:

INTRODUCTION

Most of Egyptian agricultural lands are deprived of some of the essential nutrients for growth and development of crop plants. One of the major essential elements for growth of plants is nitrogen. Nitrogen is required in large quantities for plants to grow, since it is the basic constituent of proteins and nucleic acids. Nitrogen is provided in the form of synthetic chemical fertilizer. Such chemical fertilizers pose a health hazard and microbial population problem in soil besides being quite expensive and making the cost of production high. In such situation the bio-fertilizers play a major role (Tiwarly *et al.*, 1998). Bio-fertilizers are the formulation of living microorganism, which are able to fix atmospheric nitrogen in the available form for plants either by living freely in the soil or being associated symbiotically with plants (Subba Rao, *et al.*, 1979). Bio-fertilizers are inputs containing microorganism which are capable of mobilizing nutritive elements from non-usable form to usable form through biological processes (Tien *et al.*, 1979). Biological nitrogen fixation is carried out by both symbiotic and free living bacteria and blue green algae. Nitrogen fixing bacteria are very selective in choosing roots of particular legumes species to infect, invade form root nodules (Subba Rao, *et al.*, 1979). An unique blend of organic manure using micro nutrients and some beneficial micro organisms with sugarcane press mud as base reported as useful (Arangarasan *et al.*, 2000).

In general bio-fertilizer from associative N₂ fixing bacteria could be used especially for cash crops such as vegetables, fruits, flowers and medicinal or herbal crops. It is a breakthrough technology that promises very significant impact on the country's farmers in terms of increasing farm productivity and income as well saving the country's dollars reserve due to decreased importation of inorganic nitrogenous fertilizers. It is mainly composed of microorganisms that can convert the nitrogen gas into available form to sustain the nitrogen requirement of host plants. These bacteria once associated with roots of some vegetable plants can enhance their root development growth and yield.

In the present agricultural practices there are number of microbial inoculants used as bio-fertilizers. They induce *Azospirillum* and *Azotobacter* and phosphor-bacterium, which have been given much attention as they are responsible to plant growth and yield of crops under field inoculation. In the present investigation the promoting bacteria and synthetic fertilizers as ammonium sulphate were studied in combinations with two

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cultivars (Balady and Eskandarani) to establish the morphological biochemical nutritive value, yield and productivity of okra (*Abelmoschus esculentus*, L.).

MATERIALS AND METHODS

Field and Experimental Design:

The field experiments were conducted during spring 2005 and 2006 growing seasons at the Experimental Farm of the National Research Centre at El-Nubaria, (North Egypt). Three random soil samples were taken from the field and tested before starting the experiment to establish the pH, soil texture and the total amount of nutrients present in the soil (Table 1). The plants were spaced 50 X 100 cm apart and were supplied with the recommended dose of phosphorus (9.0 g. P₂O₅/m²) and potassium (9.0 g. K₂O. / m²). The dose of nitrogen (9.8 g. N sq.m.) was manipulated at various levels in combination with different bio-fertilizers. The experimental design was split – plots, which the okra cultivars were occupied in main plots, but the nitrogen fertilization were arranged randomly within the sub- plots with 3 replicates. The sub- plot area was 30 m² included 3 ridges (1 m. width) by 10 m long. The different treatment combinations were as follows:

- cv. Balady + 100 % N as ammonium sulphate.
- cv.Balady+50 %N as ammonium sulphate + Azospirillum Bacteria.
- cv.Balady +50 % N as ammonium sulphate + Azotobacter Bacteria.
- cv. Eskandarni + 100 % N as ammonium sulphate
- cv.Eskandarni+50%N as ammonium sulphate + Azospirillum Bacteria.
- cv.Eskandarni + 50 % N as ammonium sulphate+Azotobacter Bacteria.

Plant Materials and Bio-fertilizers :

Seeds of okra plants (*Abelmoschus esculentus* L.) for the two selected varieties were seeded on the 15th and 21th of March in 2005 and 2006 seasons respectively. The bio-fertilizers used as inoculants for seed treatments were *Azospirillum* and *Azotobacter*.

Morphological Estimation:

The morphological characters such as height of the plant, number of leaves, number of shoots and shoots dry weight were estimated.

Chemical and Nutritive Value Estimation:

The composite samples were prepared for proximate analysis; Moisture was determined by oven dehydration method at 105°C up to the constant weight. Total crude protein was determined by estimating the nitrogen content of the pod materials using kjeldahl method. Chemical values, i.e. phosphorus and potassium were determined according the method which described by Troug and Mayer (1939) and Brown and Lilleland (1946) respectively. As well as Fe, Mn, Zn and Cu content were determined using flame ionization atomic absorption, spectrometer of Chapman and Pratt (1978).

Statistical Analysis:

All data values were subjected the analyses of variance according to Gomez and Gomez, 1984.

RESULTS AND DISCUSSIONS

A. Plant Growth Attributes:

Tables (2 and 3) clearly show that, the growth attributes of okra plant are influenced by the different cultivars and nitrogen treatments as well as the interaction within them in both two experimental seasons. However, the obtained data reveals that, all plant growth parameters, i.e. plant length, leaves and shoots number, fresh and dry weight of whole okra plant and its different organs all of these parameters responded no significantly at 5 % level within the two cultivars (Balady and Eskandrany). These findings are in good accordance in both experimental seasons. Concerning the effect of different nitrogen treatments on the plant growth character, the presented data in Tables (2 and 3) showed that, okra plants which supplied ammonium sulphate as 100 % of the recommended nitrogen fertilizer gained the poorest plant growth values if compared with other treatments. On the contrary, that okra plants which received its requirements of nitrogen fertilizer as 50 % of ammonium sulphate plus inoculation by *Azospirillum* as bio-fertilizer resulted in the vigor plant

Table 1: Physical and chemical properties of the experimental soil at Nubaria experimental station (Averages of 2005 and 2006).

Characters	Values
A. Physical properties:-	
Soil texture	Sandy Loam
Clay %	4.2
Silt %	23.4
Sand %	72.4
B. Chemical properties:-	
PH	7.9
E.C. (mmhos/cm)	3.12
CaCo3 %	6.6
Cations	
Ca ⁺⁺ (Meq/L)	1.2
Mg ⁺²	2.8
Na ⁺	1.35
K ⁺¹	0.21
HCo3 -	0.94
Cl -1	1.2

Table 2: Effect of the integrated use of Bio- inculants and chemical nitrogen fertilizers on growth characteristics of okra plant during the experimental season of 2005.

Characters		Plant length	No. plants		Fresh weight of plant (g)			Dry weight of plant (g)		
Treatments	N treatments	(cm)	Leaves	Shoots	Leaves	Shoots	Total	Leaves	Shoots	Total
Balady	(A)	68.06	17.20	3.33	107.07	84.37	191.43	19.00	18.87	37.87
	(B)	87.20	24.90	4.90	184.00	163.00	347.00	36.23	31.23	67.47
	(C)	83.87	24.00	4.57	174.67	140.67	315.33	32.00	27.23	59.23
	Mean	79.71	22.03	4.27	155.24	129.34	284.59	29.08	25.78	54.86
Esxandarani	(A)	73.53	19.37	3.97	120.00	92.33	212.33	25.78	20.87	41.27
	(B)	85.33	24.43	4.70	169.33	158.33	327.67	20.87	31.33	65.67
	(C)	82.33	23.87	4.57	150.33	147.33	327.67	31.33	32.07	65.07
	Mean	80.40	22.56	4.41	146.56	132.67	279.22	29.24	28.09	57.33
Average	(A)	70.80	18.28	3.65	113.53	88.35	201.88	82.09	19.87	39.57
	(B)	86.27	24.67	4.80	176.67	160.67	337.33	19.87	31.28	66.57
	(C)	83.10	23.93	4.57	162.50	144.00	3.6.50	31.28	29.65	62.15
L.S.D. at 5%	Cultivars	N.S	N.S	N.S	N.S	N.S	N.S	N.S	N.S	N.S
	N-treatments	2.86	1.64	0.32	9.95	9.12	10.66	2.52	2.69	3.68
	Interactions	N.S	N.S	N.S	14.07	N.S	15.07	N.S	N.S	N.S

(A) = 100% chemical N (B) = 50 % chemical + Bio – Azospirillum (C) = 50 % chemical + Bio - Azotobacter

Table 3: Effect of the integrated use of Bio- inculants and chemical nitrogen fertilizers on growth characteristics of okra plant during the experimental season of 2006.

Characters		Plant length	No. plants		Fresh weight (g) / plant			Dry weight (g) / plant		
Treatments	N treatments	(cm)	Leaves	Shoots	Leaves	Shoots	Total	Leaves	Shoots	Total
Balady	(A)	70.20	16.37	3.20	107.0	118.87	225.70	16.87	20.97	37.83
	(B)	90.77	26.57	3.90	192.23	164.33	356.57	41.00	38.23	79.23
	(C)	85.13	23.53	3.87	164.87	157.63	322.50	30.30	31.84	62.14
	Mean	82.03	22.16	3.66	154.72	146.87	301.59	29.39	30.35	59.73
Esxandarani	(A)	71.67	18.00	4.00	99.97	122.67	222.63	18.23	23.90	42.13
	(B)	84.43	23.80	3.40	173.90	171.07	344.97	35.77	32.80	68.57
	(C)	79.20	22.33	3.67	154.87	136.67	291.53	32.43	27.53	59.97
	Mean	78.43	21.38	3.69	142.91	143.47	286.38	28.81	28.08	56.89
Average	(A)	70.93	17.18	3.60	103.52	120.65	224.17	17.55	22.43	39.98
	(B)	87.60	25.18	3.65	183.07	167.70	350.77	38.38	35.52	73.90
	(C)	82.17	22.93	3.77	159.87	147.15	307.02	31.37	29.69	61.05
L.S.D. at 5%	Cultivars	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.
	N-treatments	2.72	0.79	N.S.	11.04	9.80	16.17	1.32	2.07	2.33
	Interactions	N.S.	1.12	0.38	N.S.	N.S.	N.S.	1.87	2.92	3.29

(A) = 100% chemical N (B) = 50 % chemical + Bio – Azospirillum (C) = 50 % chemical + Bio - Azotobacter

growth. These were true in the two experimental seasons., Generally, it could be concluded that, addition of the plant growth promoting bacteria as *Azospirillum* plus addition of 50% of the total nitrogen fertilizer as ammonium sulphate had the best plant growth values followed by that treatments which treated by bio-fertilizer as *Azotobacter* plus half of its total nitrogen required as ammonium sulphate. It is evident from the above results that the bio-fertilizers had a beneficial effect on growth attributes, this may be attributed to the plant

Table 4: Effect of the integrated use of Bio- inoculants and chemical nitrogen fertilizers on pods yield of okra and some its physical properties during the seasons of 2005 and 2006.

Characters		Pods				Pods					
Treatments					Total yield					Total yield	
Cultivars	N treatments	Length (cm)	Diameter (cm)	Average weight (g)	No./ plant	(ton/ fed.)	Length (cm)	Diameter (cm)	Average weight (g)	No./ plant	(ton/ fed.)
		2005				2006					
Balady	Control	3.17	2.02	204.87	71.98	1.434	2.34	2.41	291.40	95.20	1.725
	(B)	4.11	2.37	432.30	112.21	4.871	2.20	2.07	421.30	115.58	4.596
	(C)	4.61	2.48	417.10	102.21	2.579	2.39	2.48	368.13	110.63	3.816
	Mean	3.96	2.29	351.42	95.47	2.961	2.31	2.32	360.28	107.14	3.379
Esxandarani	(A)	3.96	2.16	261.53	94.46	1.793	1.63	2.51	222.50	92.43	1.739
	(B)	3.96	2.36	349.47	109.30	3.798	2.25	2.35	475.03	107.63	3.727
	(C)	3.96	2.37	444.77	109.30	3.397	2.55	2.32	365.77	102.58	3.004
	Mean	3.97	2.30	351.92	104.35	2.996	2.14	2.39	354.43	100.88	2.724
Average	(A)	3.56	2.09	233.20	83.22	1.614	1.99	2.46	256.95	93.82	1.732
	(B)	4.05	2.36	390.88	110.75	4.335	2.22	2.21	448.17	111.61	4.012
	(C)	4.29	2.42	430.93	105.76	2.988	2.47	2.40	366.95	106.61	3.410
L.S.D. at 5%	Cultivars	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	0.187
	N-treatments	0.31	0.10	61.54	61.54	0.137	0.12	0.14	13.46	5.49	0.197
	Interactions	0.44	N.S.	N.S.	N.S.	0.194	0.17	N.S.	19.03	N.S.	0.278

(A) = 100% chemical N (B) = 50 % chemical + Bio - Azospirillum (C) = 50 % chemical + Bio - Azotobacter

growth regulating substance such as IAA, GA and/or cytokines which is produced by *Azospirillum* know to promote better growth (Tien *et al.*, 1979; Tiwary *et al.*, 1998; Panward and Elanchezian, 1999; Rethati *et al.*, 2000). However, when the total N requirements for okra plant were added as 100 % of ammonium sulphate, the less plant growth values were recorded.

The two factors of treatments, i.e. cultivars and nitrogen treatments as interaction had a significant effect on the most of okra plant growth criteria's, these more evidence in 2nd season. Generally, plants of Balady okra cultivars which supplied by nitrogen fertilizer as ammonium sulphate plus treating by *Azospirillum* resulted the most vigor plant growth characters. These results were completely similar in the two experiments.

B. Total Pods Yield and its Some Physical Properties:

The presented data in Table (4) shows that the two okra cultivars, i.e. Balady and/or Eskandarin affected no significantly the total pods yield of okra, i.e. total pods weight as tons/fed., total pods as No./ plants and average pods weight g/plant as well as the pods physical properties, i.e. length and diameter of pod. These findings are in good similar in both two seasons, except that of total pods weight as tons/fed. in the 2nd season which Balady cultivar recorded a significance increase over than the Eskandarni one.

Addition of the total requirements of nitrogen fertilize which needed for okra plant as 100 % of ammonium sulphate gained less total pods yield as tons/fed, g. /plant and/or numbers/plant. Moreover, the poorest physical properties also gained with addition total quantity of nitrogen which needed as ammonium sulphate. On the contrary the heaviest pods weight were associated with that plants which supplied 50 % of total nitrogen requirements as ammonium sulphate plus the treating with bio-fertilizer as *Azospirillum*. The significant effect of bio-fertilizers may be due to the effect of different strains group such as nitrogen fixers, nutrient mobilizing microorganisms, which help in availability of metal and their forms in the composted materials, and increased the levels of extractable NPK (Macro or micro-nutrients).

It is evident that, the enhancement in plant growth attributes by the application of nitrogen fertilizer as 50 % of the needed requirements in the form of ammonium sulphate plus the inoculation by bio-fertilizer (*Azospirillum*), reflected on the total pods yield and its physical quality. Similar results were reported earlier by Jha and Mathur, 1993, Hegde *et al.*, 1999 and Selvakumari *et al.*, 2000.

Regarding to the pods quality, the obtained data reveals that, the highest values of pod dimension were recorded with that plants which applied by 50 % of chemical nitrogen fertilizer plus the inoculation by *Azotobacter* in both experiments of 2005 and 2006.

The interaction between two cultivar of okra and 3 nitrogen treatments had a significant effect on pods yield as tons/fed., and length of pod in both seasons. However, the significant response of average pods number (1st season) and average pods weight per plant (2nd season), also were noticed. Generally in spite of the no significant effect of the interactions on some parameters of pods yield, but it could be reported that Balady cultivar of okra plant when applied half of its requirement of nitrogen fertilizer as ammonium sulphate plus the inoculation by *Azospirillum* gained the heaviest tonnage of pods yield.

Table 5: Effect of the integrated use of Bio- inoculants and chemical nitrogen fertilizers on nutritional values as percentages of okra pod yield during the season of 2005.

Characters		%				ppm			
Treatments		Protein	N	P	K	Fe	Mn	Zn	Cu
Cultivars	N treatments								
Balady	(A)	5.00	0.80	0.103	0.340	5.76	0.11	0.189	0.152
	(B)	5.92	0.95	0.145	0.416	7.33	0.140	0.213	0.207
	(C)	5.75	0.92	0.138	0.395	7.08	0.145	0.202	0.246
	Mean	5.56	0.89	0.129	0.384	6.72	0.133	0.201	0.201
Esxandarani	(A)	4.63	0.74	0.072	0.346	6.24	0.119	0.194	0.151
	(B)	6.17	0.99	0.159	0.382	7.43	0.128	0.206	0.225
	(C)	5.81	0.93	0.126	0.356	6.85	0.126	0.201	0.216
	Mean	5.53	0.89	0.119	0.361	6.84	0.124	0.200	0.197
Average	(A)	4.81	0.77	0.088	0.343	6.00	0.116	0.192	0.151
	(B)	6.04	0.97	0.152	0.399	7.38	0.134	0.210	0.216
	(C)	5.78	0.93	0.132	0.375	6.97	0.136	0.201	0.231
L.S.D. at 5%	Cultivars	N.S.	N.S.	N.S.	N.S.	N.S.	0.003	N.S.	N.S.
	N-treatments	0.33	0.05	0.013	0.009	0.27	0.004	0.005	0.028
	Interactions	N.S.	N.S.	N.S.	0.013	N.S.	0.006	N.S.	N.S.

(A) = 100% chemical N (B) = 50 % chemical + Bio – Azospirillum (C) = 50 % chemical + Bio - Azotobacter

Table 6: Effect of the integrated use of Bio- inoculants and chemical nitrogen fertilizers on nutritional values as percentages of okra pod yield during the season of 2006.

Characters		%				ppm			
Treatment		Protein	N	P	K	Fe	Mn	Zn	Cu
Cultivars	N treatments								
Balady	(A)	4.00	0.64	0.094	0.365	5.56	0.119	0.200	0.157
	(B)	7.98	1.28	0.134	0.381	7.40	0.133	0.214	0.206
	(C)	8.23	1.32	0.182	0.416	6.75	0.158	0.215	0.211
	Mean	6.74	1.08	0.137	0.387	6.57	0.137	0.209	0.192
Esxandarani	(A)	5.04	0.81	0.083	0.396	6.03	0.120	0.193	0.170
	(B)	7.08	1.13	0.154	0.428	7.14	0.137	0.222	0.209
	(C)	6.42	1.03	0.144	0.425	6.78	0.153	0.230	0.236
	Mean	6.18	0.99	0.127	0.416	6.65	0.137	0.215	0.205
Average	(A)	4.52	0.72	0.088	0.380	5.80	0.120	0.196	0.164
	(B)	7.32	1.21	0.144	0.405	7.27	0.135	0.218	0.208
	(C)	7.32	1.17	0.163	0.421	6.77	0.155	0.222	0.224
L.S.D. at 5%	Cultivars	N.S.	N.S.	N.S.	0.009	N.S.	N.S.	N.S.	N.S.
	N-treatments	0.75	0.12	0.009	0.012	0.30	0.009	0.006	0.013
	Interactions	1.06	0.17	0.012	N.S.	N.S.	N.S.	N.S.	N.S.

(A) = 100% chemical N (B) = 50 % chemical + Bio – Azospirillum (C) = 50 % chemical + Bio - Azotobacter

C. Nutritional Values of Okra Yield:

Tables (5 and 6) show clearly that, the nutritional values of okra pods are responded by the treatments of two cultivars and nitrogen fertilizers as well as its interactions in the two experimental seasons. However, protein, N, P, K, Fe, Mn, Zn and Cu content in pods tissues responded no significantly within the two cultivars. These were true in both seasons, except Mn (1st season) and K (2nd season) which recorded a significant difference at 5 % level. In spite of the no significant response, generally the obtained data reveals that, the better nutritional values were recorded with Balady cultivar.

Regarding the relationship between nitrogen treatments and the nutritional values of okra pods, the presented data indicates that plants which received half of its nitrogen fertilizer requirement as ammonium sulphate plus the inoculation by *Azospirillum* as nitrogen fixation gained the best nutritional values. These are in good accordance for all determined elemental values and protein content in 1st season.

In the second season some changes occurred, where protein, N and Fe content followed the same pattern of change like that above mentioned, but other nutritional values recorded their peaks with that okra plants which supplied ammonium sulphate as half of its nitrogen needs plus the *Azotobacter* inoculations.

The plants treated with bacterial inoculants showed better results for mineral content which are important for vital body function such as acid base and water balance.

Generally, the use of bio-N fertilizer was highly affective for increasing vegetative growth, pods yield and enhancing nutritive values. It could be summarized that bio-N fertilization could be used to reduce the total cost of the chemical N requirements to 50 % or less, consequently which will help in health and money saving as well as reduce the pollution.

It could be concluded that, the application of bio-fertilizer as bacterial microorganisms with chemical nitrogen fertilizer for okra plantation resulted the better nutritional values in pods tissue if compared with that plants which received its all nitrogen fertilizer requirements as chemical.

The beneficial effects of bacterial inoculation on increased protein content and elemental nutrition might have been due to the supply of high available amount of nitrogen to the growing tissue and organs supplied by N₂-Fixing *Azospirillum* and *Azotobacter* Rukmani, 1990).

The statistical analysis of the collected data reveals that the differences within different N treatments were significantly at 5 % level. These were true for all nutritional values in both seasons.

The interaction treatments between the two okra cultivars and 3 nitrogen application treatments gained no statistical significant effect on the nutritional values. These held good in the two experiments except the content of K and Mn (1st season) as well as protein, N and P (2nd season).

Generally, the no significant response of most nutritional values in okra pods tissue may be attributed to the independent effect of the interaction treatments and each factor of the interaction act independently.

REFERENCES

- Arangarasan, V., K.M. Darshinamoorthy, G. Ramesh, and S.P. Palaniappan, 2000. Micro nutrient mixture. A unique blend of organisms. Science Express: 3.
- Browen, J.D. and O. Lillaland, 1946. Rapid determination of potassium and sodium in plant material and soil extracts by flame photometry. Amer. Soc. Hort. Sci., 38: 341-364.
- Champman, H.D. and P.F. Pralt, 1978. Methods of analysis for soils .plants andwaters Univ .California, Div. Agric. Sci. Priced pud., 3034.
- Gomez, K.A. and A.A. Gomez, 1984. Statistical procedures for Agriculture Research Second Ed. Willey Inter Science., Publ. 357-423.
- Hedge, D.M., B.S. Dwivedi, S.N. Bahu and Sudhakara, 1999. Bio-fertilizers for cereal production in India. Indian Journal of agriculture Science, 69: 73-83.
- Jha, d. and R.S. Mathur, 1993. Combined effect of nitrogenous fertilizers and *Azospirillum* brasilense on the yield of nitrogen uptake by pearl millet. International Journal of Tropical Agriculture, 11: 31-35.
- Panward, J.D. and R. Elanchezian, 1999. Effect of 2,4-D and *Azospirillum* brasilense on growth and yield in the nodule induced transplanted wheat. Indian J. of plant Physiology, 3: 143-146.
- Rethati-Beatrix, Dallmann-Klara, Simon-Ibolya, K. Balint-Andor, 2000. Characterization of Hungarian rice cultivars in nitrogen fixing associations with bacteria. Cereal Res. Communication, 28: 9-16.
- Rukmani, R., 1990. Physical chemical and biological regulation of fruit characters and yield in Okra (*Abelmoschus esculents* L.). Department of Floriculture, College of Horticulture, Vellanikara Kerala Agri. University, India.
- Selvakumari, G., M. Basker, D. Jayanthi and K. Mathan, 2000. Effect of integration of fly ash with fertilizers and organic manures on nutrient availability, yield and nutrient uptake of rice. J. of Soil Sci. the Soc., 48: 268-278.
- Subba rao, N.S., K.V.B.R. Tilak, C.S. Singh and M. Lakshmikumari, 1979. Response of few economic species of graminiae plants to inoculation with *Azospillum* brasilense. Current Science, 52: 439-440.
- Tien, T.N., N.H. Gaskins and D.H. Hubn\bbell, 1979. Plant growth substances produced by *Azospillum* brasilense and their effect of growth of pearl Millet. Applied and Environmental Microbiology, 37: 1016-1024.
- Tiwary, D.K. D.M. Abuhasan and Chattopadhyay, 1998. Studies on the effect of inoculation with *Azotobacter* and *Azospirillum* on growth yield and quality of Banana. Indian Journal of Agriculture, 42: 235-240.
- Troug, E. and A.H. Mayer, 1939. Improvement in the denies calorimetric method for phosphorus and arsenic. Indian engineering chemical anwal Ed., 1: 136-139.