

Impact of different doses of Biofertilizer; *Azotobacter*, *PSB* and their combination doses on available Nitrogen (N), Phosphorus (P) and Potassium (K) in soil after the harvest of *Aloe vera* crop.

Ajaz Ahmad Bandy*, Javaid Ahmad Khan

Dept. of Botany, Saifia Science P.G. College, Saifia Road, Bhopal - 462 001 (M.P.)

ABSTRACT

An experiment was conducted on *Aloe vera* L. to evaluate the inoculation impact of *Azotobacter*, PSB and their combination doses or treatments on the availability of nitrogen (N), phosphorus (P) and potassium (K) in the soil, after the crop harvest. The biofertilizers were applied in six different doses to *Aloe vera* plants in a planned manner viz T1 = 20 gm (*Azotobacter*), T2 = 40 gm (*Azotobacter*), T3 = 20 gm (PSB), T4 = 40 gm (PSB), T5 = 20 gm + 20 gm (*Azotobacter* + PSB), T6 = 40 gm + 40 gm (*Azotobacter* + PSB). Some plants were kept untreated and act as control population. The observation is recorded and mean value is presented as kilogram per hectare. The result indicated that maximum nitrogen availability in soil were found in the individual treatment (T2) of *Azotobacter*, which is recorded as 170 ± 6.7 kg/hac. The combination treatment (T6) of *Azotobacter* and PSB also shows maximum availability of N, which is recorded as 168 ± 7.21 Kg/hac., as compared to control 148 ± 8.12 kg/hac. The available phosphorous (P) is found to be maximum in the individual treatment (T4) of PSB which is recorded as 18.20 ± 1.6 kg/hac., followed by combination treatment (T6) where P content in soil records 18.12 ± 1.5 kg/hac. as against control population which is recorded as 12.24 ± 1.6 kg/hac. The highest quantity of potassium (K) in the soil is observed under the combination treatment (T6) of *Azotobacter* and PSB, which is recorded as 320 ± 1.6 kg/hac, followed by T5 (299 ± 1.6 kg/hac) and T2 (298 ± 1.6 kg/hac.). The dual inoculation of *Azotobacter* and PSB to *Aloe vera* shows better improvement in the availability of NPK in the soil after the harvest of crop. However the availability of N and P shows maximum value in individual treatments but over all results indicates that dual inoculation with higher doses are more better in terms of availability of NPK content in soil. Besides , bio-fertilizers remain eco-friendly with the soil environment.

Keywords: *Aloe vera*, *Azotobacter*, PSB, NPK content, harvest.

INTRODUCTION:-

The word Biofertilizers are itself self explanatory as they are ready to use the live formulates of such beneficial microorganisms which on application to seed, root or soil mobilize the availability of nutrients by their biological activity in particular, and help build up the micro flora and soil health in general. They are extremely beneficial in enriching the soil with those micro-organisms, which produce organic nutrients for the soil and help to combat diseases.

Azotobacter culture is one of the important biofertilizer which has been used world wide. It is free living, gram negative, rod shaped, nitrogen fixing bacterium. *Azotobacter* fixes 20-30 Kg N/hac and can be applied in various crops. Besides, It is capable of producing growth promoting substances such as gibberallins, cytokinins, Indole-acetic acid as well as antibiotics which suppress and control plant pathogens. Inoculation with *Azotobacter* sp. complements the symbiotic relationship between plant roots and fungi due to its nitrogen fixation, phytohormones - production and phosphate solubilization properties (Kumar et al., -2001, Narula et al., 1980). Three types of bacteria, i.e. symbiotic, non-symbiotic, free living are useful to reduce molecular form of atmospheric nitrogen into Ammonia (NH₄) which is assimilated by plants. The range of fixation is 2-15 mg N fixed / g of carbon source utilized. Phosphate solubilization is brought about by chelating effect and production of organic acids, Thus making increased quality of phosphorous available to plants. The PSB from subtropical soil has tri-calcium solubilising ability (Chen et al., 2005). The beneficial effects of dual inoculation have been reported by many workers (Mandhare et al., 1998, Sreeramula et al., 2000) for certain plant species.

Nitrogen (N), phosphorus (P) and Potassium (K) are macronutrients and are needed to plant in macro quantity. Their presence in the soil is essential for normal growth and development of plant and any deficiency may cause several disturbance to normal plant growth and development. In the present experiment, Biofertilizers; *Azotobacter* and PSB were used for inoculation purpose. *Aloe vera* Linn. plants were cultivated in pots and in field and the suckers (pops) were inoculated with different dose quantity of biofertilizer both at individual level and in combination level. After the maturity of *Aloe vera* crop, the growth and yield parameters were observed and analysed at the maturity and data obtained were tabled. But, after the harvest of *Aloe vera* crop, the availability of nitrogen (N), phosphorus (P) and Potassium (K) were observed by soil analysis and the data obtained is compared with every treatment. So, in present investigation, the impact of *Azotobacter* and PSB (both at individual level and in combination level) on the availability of NPK in the soil after the harvest of *Aloe vera* crop was evaluated.

(2) MATERIAL AND METHODOLOGY:

2.a Experimental site and soil conditions:- The experiment was conducted at Saifia Science P.G. College, Bhopal. The climate at the experimental site is subtropical, which is mostly dry except monsoons. In summer season (March - middle June), temperature varies from the lowest 9.9 to 45.1 °C. although the plant has the ability to grow in any kind of soil but for better results, Sandy loamy soil is used.

2.b Biofertilizers inoculants and plant material:- Root suckers (pops) of *Aloe vera* of about 15-18 cm long were obtained from nursery of Bhopal. These root suckers were planted in pots and in field for culturing. Biofertilizers (*Azotobacter* sp. and PSB) were brought from M.P. Agro Industry Bhopal. The analysis of soil was done in Indian Institute of Soil science (IISS), Bhopal.

2.c Collection of soil samples:- The soil of the experiment site was collected for filling the pots and after and before harvesting of crop, soil samples were analysed for physico-chemical properties and for the availability of NPK. For the inoculation purpose, earthen pots of 20 inches were taken and then filled by soil. In the field culture, plant suckers were planted at a spacing of 70x60 cm accommodating 12 plants in 5.4 m² plot at four plants per row and 3 rows per plot.

2.d Experimental design:- The experiment was carried out in Randomised Block design (RBD), in which Aloe plant were inoculated with different dose of *Azotobacter*, PSB and their combination after planting of Pops of *Aloe vera* in the soil. The sukers were inoculated in a sequenced manner as treatment first (T1) and second (T2) were of *Azotobacter* with dose quantity of 20 gm and 40 gm respectively, T3 and T4 represents PSB with 20 gm and 40 gm dose quantity. Whereas in combination doses of T5 and T6, homogeneous mixture of *Azotobacter* and PSB were used for inoculation with dose quantity of 20 gm + 20 gm *Azotobacter* and PSB, and 40 gm +40 gm of *Azotobacter* and PSB. Some pops of *Aloe vera* were kept untreated both in pot and field culture which acts as control population.

After the harvest of crop, the soil was analysed by different methods as shown below for the detection of Available nitrogen, phosphorous and potassium quantity.

2.e Parameters recorded and statistical analysis:- The availability of Nitrogen (N) was determined by potassium permagnate method (Subbiash and Asija, 1956). Available phosphorus is evaluated by Olsen's method (1954) and available potassium was also analysed.

Available nitrogen (N):

Available nitrogen in soil was determined by potassium permanganate method (Subbiash and Asija, 1956). 20 g of soil sample was taken in one liter distillation flask, 20 ml distilled water was added to it followed by 100 ml of 0.32% potassium permanganate solution, then 100 ml of 2.5% NaOH was added and distilled. About 100 ml of distillate was collected in a 250 ml. conical flask

containing 20 ml boric acid mixed bromocresol green methyl red indicator and was titrated with standard 0.02 N H₂SO₄ solution, till the colour turned to light pink.

Available Phosphorus (P):

To extract and evaluate the available phosphorus from the soil the Olsen's method was used (Olsen et al., 1954). 2.5 g of soil was taken in 100 ml conical flask with 1 g Darco G-60 (Phosphorous free) and 50 ml 0.5 M NaHCO₃ solution was added to it. After shaking the flask for 30 minutes the solution was filtered through whitman No-40 filter paper. Then 5 ml filtrate was transferred into 25 ml volumetric flask and 5 ml ammonium molybdate solution was added. After proper shaking 1 ml of diluted stannous chloride (SnCl₂) solution was added and the volume was made upto 25 ml after shaking. The transmittance of the blue colour so developed was read after 10 minutes on spectronic - 20 at 660 nm wavelength and the phosphorus in gm/plot and gm/pot was calculated. Then average and mean value is calculated as Kg/hac. by statistical analysis.

Available Potassium (K):

Take 5 numbers of 1000 ml volumetric flask than N CH_3COONH_4 (pH 7.0) solution in one flask to fill it upto 1000 ml mark. It gives 0 ppm K_2O solution. With a graduated pipette take 5, 10, 15 and 20 ml of 1000 ppm K_2O solution in individual flasks. Dilute each to 1000 ml with N CH_3COONH_3 (pH 7.0) solution. Thus 5, 10, 15 and 20 ppm K_2O solution are prepared. Take few drops of butyl alcohol in each of the flask to improve spraying properties of the solution. Place the K filter in the flame photometer. Set up the photometer by atomizing 0 ppm K_2O solution and 20 ppm K_2O solution alternatively to 0 and 100 scale reading respectively. Atomise the solutions i.e 5, 10, 15 ppm K_2O solutions and record the meter readings. On a graph paper, plot the meter readings against the respective ppm K_2O , Join the points. The graph, Thus drawn, will be a straight line passing through the origin. This straight line is the standard curve.

RESULT AND DISCUSSION:-

Considerable variation in terms of quantitative attributes was observed after the analysis of available NPK (Nitrogen, Phosphorus and Potassium) quantity. The subsequent variation interms of NPK under the impact of *Azotobacter*, PSB (both at individual level and in combination level) were observed after the harvest of *Aloe vera* crop and data obtained were furnished in Table-1.

**Observation of Available "NPK" after the inoculation of biofertilizers
(Azotobacter, PSB and their combination doses)**

Treatments	Biofertilizers	Dose	Available nitrogen (N) kg/hac	Available phosphorus (P) kg/hac	Available potassium (K) kg/hac
T ₁	<i>Azotobacter</i>	20 gm	165 ± 5.9	14.12 ± 1.4	290 ± 1.4
T ₂	<i>Azotobacter</i>	40 gm	170 ± 6.7	15.20 ± 1.4	298 ± 1.6
T ₃	PSB	20 gm	149 ± 6.9	16.13 ± 1.3	289 ± 1.5

T ₄	PSB	40 gm	151 ± 6.7	18.20 ± 1.6	294 ± 1.7
T ₅	Azotobacter + PSB	20 gm + 20 gm	162 ± 7.12	17.12 ± 1.4	299 ± 1.6
T ₆	Azotobacter + PSB	40 gm + 40 gm	168 ± 7.21	18.12 ± 1.5	320 ± 1.6
T ₇	Untreated	Control	148 ± 8.12	12.24 ± 1.6	288 ± 1.7
	LSD _{0.05}		18.16	2.19	18.32
	CV (%)		7.31	9.72	6.10

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3.1 Effect of biofertilizers (Azotobacter and PSB) on available Nitrogen (N) in soil.

The availability of nitrogen (N) was found maximum in treatment second of 40 gm *Azotobacter*, which is recorded as T₂ = 170 ± 6.7 Kg N/hac followed by T₆ (combination treatment of 40 gm + 40 gm; *Azotobacter* and PSB) where it is recorded as 168 ± 7.21 Kg N/hac. The lowest value of available nitrogen was found in treatment three (T₃ = 149 ± 6.9 Kg N/hac) as compared to control (148 ± 8.12) as shown in Table-1 Fig.1. Similar findings have also been reported by Biswas et al., (2001) in mungbean cultivation. The high dose at individual level of inoculation of *Azotobacter* might have fixed and mobilized higher amount of nitrogen (N) and make it available for the plants. Nitrogen fixation bacteria not only increases the nitrogen for plants but also increases available nitrogen (N) in soil studied after post harvesting of crop (Singh & Singh 2006). A marked increase in soil Nitrogen (N) in post harvest soil were also reported by Govindan and Thirumurugan (2005).

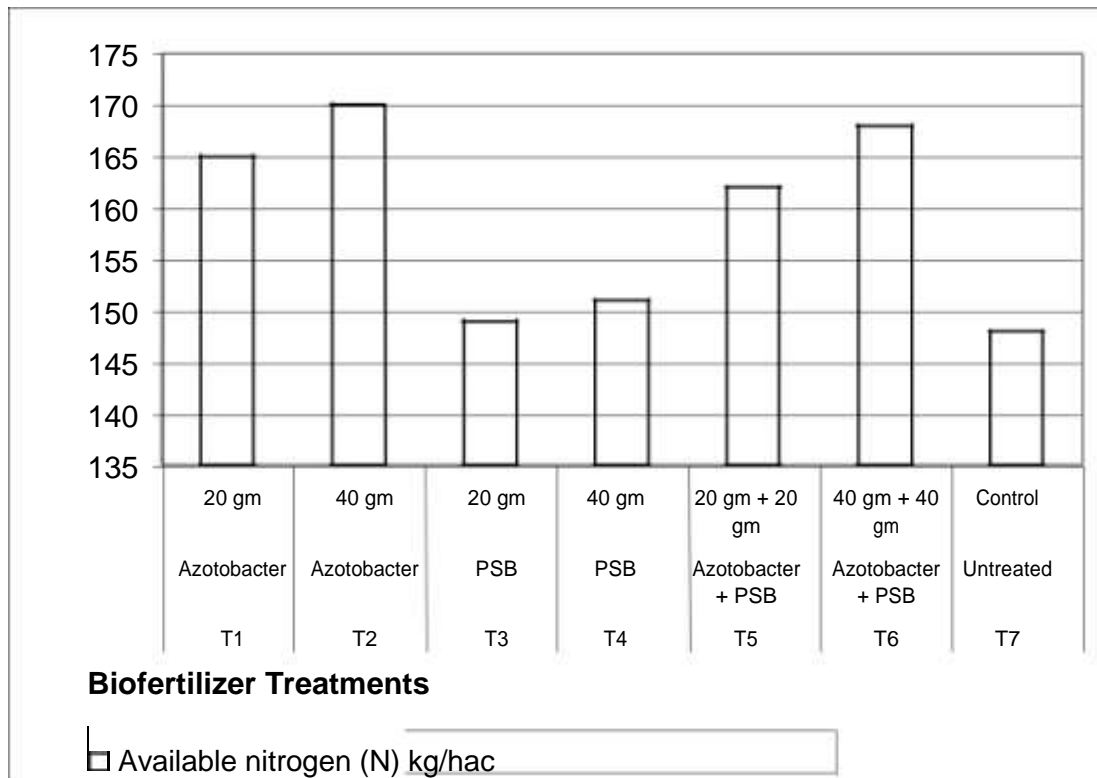


Fig.1 Post harvest Nitrogen (N) availability in soil after the harvest of *Aloe vera* crop.

3.2 Effect of Biofertilizer (*Azotobacter* and PSB) on available phosphorus (P) in soil:

Under the Individual treatment(T4) of PSB (40 gm), maximum phosphorus was found in the soil, which is recorded as 18.20 ± 1.7 kg/hac. followed by combination treatment (T6) of *Azotobacter* and PSB with dose quantity of 40 gm + 40 gm, which is recorded as 18.12 ± 1.5 kg/hac as shown in Table 1 and Fig. 2. The minimum availability of phosphorus (P) was found in T1 (*Azotobacter*) which is recorded as 14.12 ± 1.4 Kg/hac. as against the control population which is 12.24 ± 1.6 kg/hac. The increased availability of phosphorus in the soil might be due to the increased doses of PSB. About 98% of Indian soils are inadequate in available P. (Gaur 1987). Gaur and Rana (1990) also reported that phosphatases secreted by AM fungi or PSB must have hydrolysed organic P to inorganic P for utilization by plants. The present results are also in conformity of Chinussamy et al., (2006).

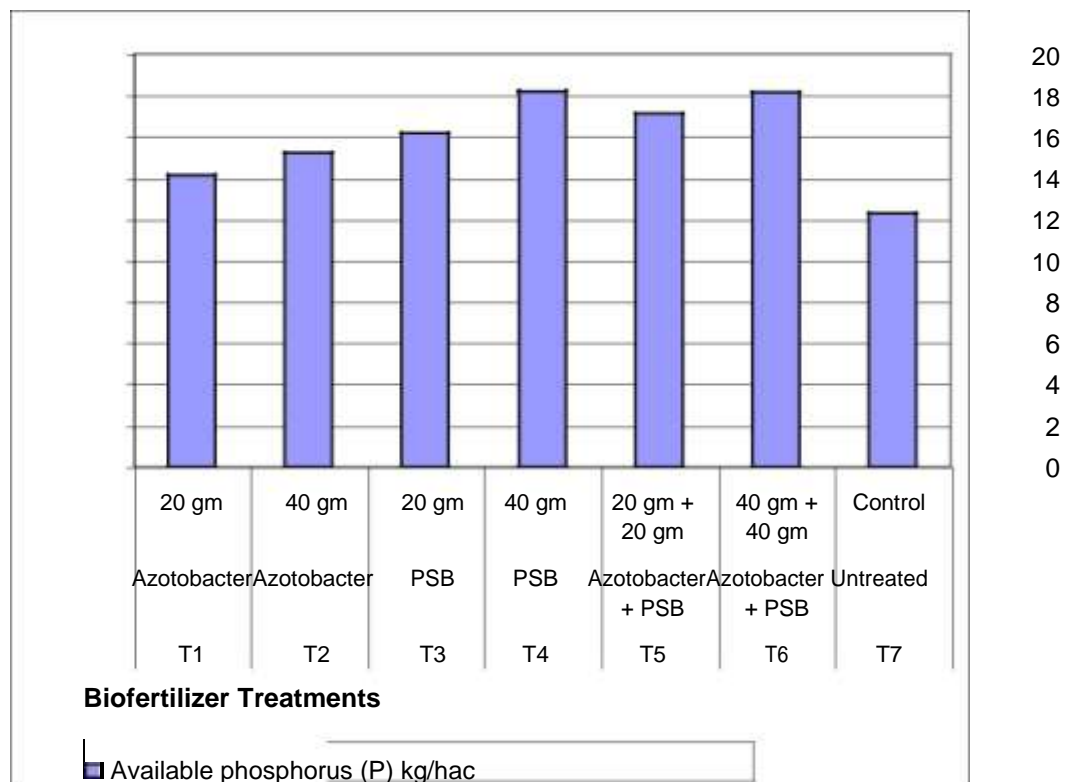


Fig.2 Variation of available phosphorus (P) in soil after the harvest of *Aloe vera* crop.

3.3 Effect of Biofertilizers (Azotobacter and PSB) on available potassium (K) in soil.

The potassium (K) availability after the harvest of *Aloe vera* crop also shows subsequent variation under the impact of *Azotobacter* and PSB. The potassium was found maximum in treatment six (T6 = 320 ± 1.6 kg/hac.) as against the control, which is recorded as 288 ± 1.7 kg/hac. Among the individual treatments. The T2 (40 gm *Azotobacter*) treatment records maximum value which is recorded as 298 ± 1.6 kg/hac. as shown in Table-1 and Fig.3. The higher combination doses of *Azotobacter* and PSB might have increased impact on the solubilization and mobilization of nutrients in the soil. The dual inoculation have a great impact on the availability of potassium in the soil after the harvest of *Aloe vera* crop. The dual inoculation has been reported by many workers. (Mandhare et al., 1998 and Vasiler et al., 2001). The results are also in agreement with the findings of Sharma and Namdeo (1990).

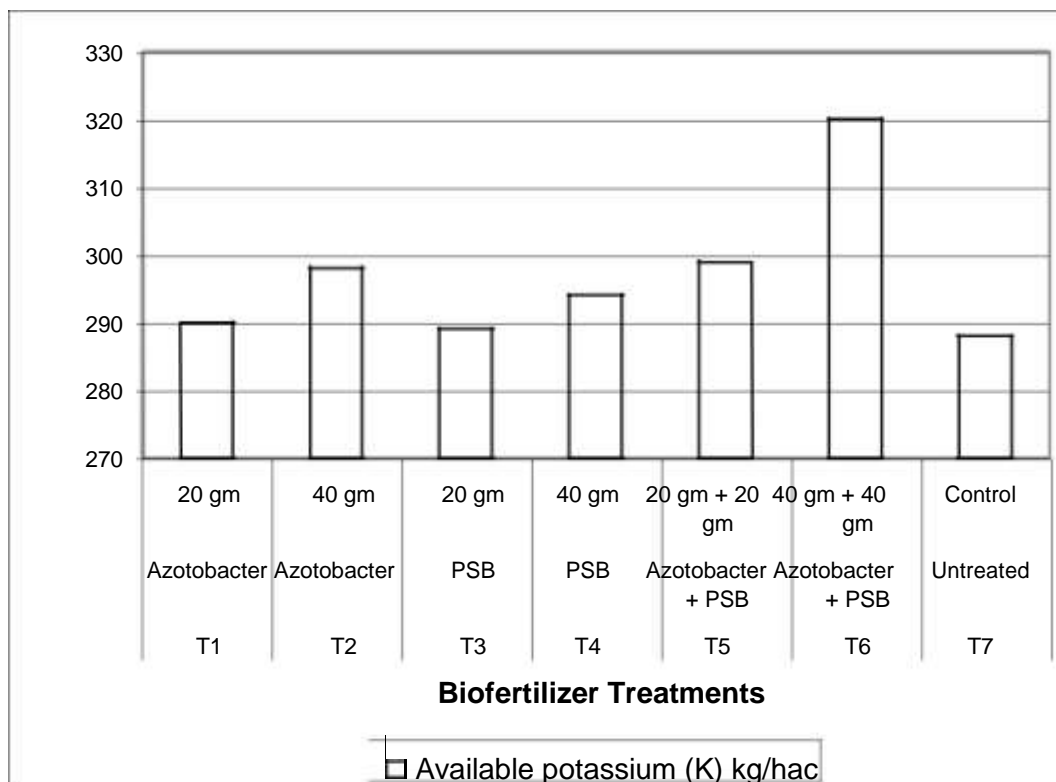


Fig.3 Effect of Biofertilizers (Azotobacter and PSB) on available potassium (K) in soil.

After the analysis of soil, It was concluded that combination doses of biofertilizer inoculation shows vigorous and individual doses shows semi vigorous impact on the availability of nitrogen, phosphorous and potassium, in the soil after the harvest of *Aloe vera* crop. Besides, higher dose quantity inoculation of biofertilizers have also an impact on greater solubilization and mobilization of nutrients and make them available for plants. similar kind of results were also found in the work done by various research scholars and scientists so far and also the results are in conformity with the results of impact of biofertilizers on oil yield crops.

ACKNOWLEDGEMENT:

I am thankful and record my sincere gratitude to my supervisor, Dr. Zia-ul-Hassan, who guided this work. I am also thankful to Rayees Ahmad Dar (Research Scholar, Biotechnology) for providing me valuable information about Biofertilizer mode of action.

REFERENCES:

1. Biswas, B.C.; Das and Kalwe, P.S. (2001) crop response to fertilizers. *Fert. News*, 46 (2): 15-18.
2. Chen, Y.P.; Rekha, P.D.; Arun, A.B.; Shen, F.T.; Lai, W.A. and Young, C.C. (2005). Phosphate solubilizing bacteria from subtropical soil and their Tricalcium Phosphate ability. *Elsevier B.V.*
3. Chinnusamy, M.; Kanshik, B.D. and Prasanna, R. (2006). Growth, Nutritional and yield parameters of wet land rice as influenced by microbial consortia under controlled condition. *J. of plant nutri.* 29 (50); 857-871.
4. Gaur, A.C. (1987). Organic manures and Biofertilizers, Division of Microbiology, IARI, *New Delhi*, 46.
5. Gaur, A.C. and Rana (1990), Role of Mycorrhiza, PSB and their interaction on growth and uptake of nutrients by wheat crop. In: current trends in Mycorrhizal

Research. Proceedings of the National Conference on mycorrhiza held at HAU, Hisar pp. 105.

6. Govindan, K. and Thirumurrigan, V. (2005). Synergistic association of *Rhizobium* with PSB under different sources of nutrient supply on productivity and soil fertility in soyabean (*Glycine max.*) *Ind. J. Agron.* 50(3) 214-217.
7. Kumar, V.; Behl, R.K. and Narula, N. (2001). Establishment of phosphate solubilizing strains of *Azotobacter Chroococum* in the rhizosphere and their impact on wheat cultivars under green house conditions, *Microbiological research*, 156: 87-94.
8. Madhare, V.K.; Patil, P.L. and Gadekar, D.A. (1998). Phosphorous uptake of onion as influenced by *Glomus fasciculatum*, *Azotobacter* and phosphorous levels, *Agric. Sci. Digest*, 18(4): 228-230.
9. Narula N.; Lakshminarayana, K.D. and Tauro, (1980). Ammonia excretion by *Azotobacter Chroococum*. *Biotechnol Sci.* 23: 467-470.
10. Sharma, K.N. and Namdeo, K.N. (1999). Effect of biofertilizer and phosphorous on growth and yield of soyabean (*Glycine max. L. Crop Res. Hisar.* 17: 160 - 163.
11. Singh, M.S. and Singh, I. (2006). Studies on influence of Biofertilizers and bio regulators on flowering, yield and fruit quality of strawberry Cv. Sweet Charic. *Ann. Agric. Res. New Series.* 27(3): 261 - 264.
12. Vasiler, N. ; Vassileva, M.; Azeon, R. and Medina, A. (2001). Interactions of an arbuscular Mycorrhizal fungus with free or co-encapsulated cells of *Rhizobium trifolia* and *Yarrowia lipolytica* inoculated into a soil - plant system *Biotechnol. Lett.* 23: 149-151.