

Effect of Liquid Formulation of *Azotobacter* and PSB Inoculation on Growth and Yield of Lettuce

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ABSTRACT

The present investigation entitled “Effect of liquid formulation of *Azotobacter* and PSB inoculation on growth and yield of lettuce (*Lactuca sativa* L.)” was carried out at Department of Plant Pathology, College of Agriculture, Pune in view to study the population dynamics of *Azotobacter* and PSB at periodic interval, their effect on growth parameters and yield of lettuce crop. There were seven treatments, including seed treatment with liquid *Azotobacter* and liquid PSB @ 25 ml/kg of seed; Foliar spray of liquid *Azotobacter* and PSB @ 25ml/lit, in alone and in combination; respectively. Results indicates that seed treatment with liquid *Azotobacter* + foliar spray of liquid *Azotobacter*, was found superior amongst all the treatments conducted in respect of number of leaves (13.33), length of leaves (19.60 cm), breadth of leaves (8.70 cm), height of plant (84.23 cm), leaf area index (73.33 cm²), fresh weight 72 (24.06 g) and dry weight (7.42 g) of leaves, total dry matter weight (14.34 g), yield (24.06 g) over uninoculated control. Considering all these parameters, it could be concluded that liquid *Azotobacter* and PSB improve growth which may ultimately influence the yield of lettuce.

Key words: *Azotobacter*, PSB (Phosphate solubilizing bacteria), Growth parameters.

INTRODUCTION

Lettuce (*Lactuca sativa* L.) is an annual plant belong to family asteraceae. It is most often grown as a leafy vegetable, but sometimes for its stem and seeds. Lettuce is most often used in salads, also in other kinds of food, such as soups, sandwiches. Lettuce is a good source of vitamin A and potassium, as well as a minor source for several other vitamins and nutrients.

Liquid biofertilizer technology is an alternative solution to carrier based

biofertilizers. It comprises aids to preserving organism, to delivering them to their targets and improves their activities. These are special liquid formulation containing not only the desired microorganism and their nutrients but also special cell protectants or substances that encourage formation of resting spores or cyst for longer shelf life and tolerance to adverse condition.

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Unlike the lignite based biofertilizers, liquid biofertilizers have a longer shelf life (Rao, 2007). By applying an appropriate liquid biofertilizer, the overall cost of production will be much lower as compared to traditional chemical fertilizers (Chin, 2010).

Soil microorganisms have enormous potential in providing soil phosphates for plant growth. Phosphorus biofertilizers in the form of microorganisms can help in increasing the availability of accumulated phosphates for plant growth by solubilization (Goldstein, 1996; Gyaneshwar *et al.*, 2002). In addition, the microorganisms involved in P solubilization as well as better scavenging of soluble P can enhance plant growth by increasing the efficiency of biological nitrogen fixation, enhancing the availability of other trace elements and by production of plant growth promoting substances (Gyaneshwar *et al.*, 2002).

An application of biofertilizer to seeds improve its germination to a considerable extent and controls plant diseases infection due to antagonistic nature considering the adverse

effect of chemical fertilizers, the bio-fertilizers are economically cheaper and work as eco-friendly. In view of this background information, the present experiment was undertaken to study the population dynamics of *Azotobacter* and PSB at periodic interval, their effect on growth parameters and nitrogen uptake by lettuce crop.

MATERIALS AND METHODS

Seed inoculation

For inoculation of liquid *Azotobacter*, the seeds of lettuce were dipped in liquid *Azotobacter* suspension @ 25ml/kg of seeds and seed treatment with liquid PSB was given by dipping seeds in liquid PSB suspension @25 ml/kg.

Seed germination

The germination count was taken at 10th DAS and germination percentage was calculated for all the experimental pots by following formula:

$$\text{Germination Percentage} = \frac{\text{Total number of seeds germinated}}{\text{Total number of seeds sown}} \times 100$$

Number of leaves

Number of leaves per plant was recorded at 20 days interval by taking leaf count per lettuce plant.

Length and breadth of leaves

The length and breadth of leaves was recorded in centimetre by using meter scale at 20 days intervals.

Plant height

The height of plant in centimetre was recorded from collar region to the growing tip of the main stem at 20 days intervals.

Fresh and Dry weight of leaves

Fresh weight of leaf recorded immediately after the uprooting the plant sample. The leaves were detached from shoot and weigh on weighing balance and recorded leaf weight. Dry weight of leaves were recorded by air drying of leaves and it was wrapped in brown

paper bags followed by oven dried at 70± 20C till constant weight is obtained.

Leaf area index

The leaf area index was measured by paper graph method. Fresh leaf collected from plant and placed on graph paper, draw an outline around the margins of leaf by using pencil and calculate the squares.

Total dry matter weight of plant

Plants uprooted at maturity stage and giving gentle wash to roots for remove adhering soil on root surface, air dried and the root and shoots 23 were wrapped in brown paper bags separately and oven dried at 70± 20C till constant weight is obtained and recorded as a dry weight of shoot and root which ultimately gives the total dry matter weight of plant.

Yield of Lettuce per plant

Harvested fresh lettuce leaves were taken and weight was recorded immediately as a yield.

RESULTS AND DISCUSSION

Effect of liquid *Azotobacter* and PSB inoculation on growth of lettuce

Effect on seed germination

The results on seed germination shown statistically non-significant over uninoculated control. However, marginal increase in seed germination in seed treatment with liquid *Azotobacter* @ 25ml/kg + seed treatment with liquid PSB @ 25ml/kg (84.44%) than uninoculated control (75.55%). The increase in germination percentage due to inoculation of liquid bioinoculants may be due to ability to suppress the growth of antagonists present in soil and on seed coat and release of plant growth promoting substances around seed rhizosphere. Similar observations were also noted by Jadhav and Patil (1985) reported that *Azotobacter* as biofertilizer performed better than inorganic fertilizers in relation to seed germination of paddy plant and Sajindranath *et al.* (2002) in okra due to biofertilizers like *Azotobacter* and PSB. Sharma *et al.* (2007) found that increase in seed germination due to inoculation with PSB in *Cicer arietinum* L. The bioinoculants *Azotobacter* and PSB found the significant increase in seed germination percentage was reported by Pathak *et al.* (2013). Similar findings by Mahato *et al.* (2009) with *Azotobacter* as a biofertilizer.

Effect on number of leaves per plant

It was revealed that inoculation with liquid *Azotobacter* and PSB significantly increased the number of leaves per plant over the uninoculated control. The results obtained were statistically significant at 45 DAS and subsequently at 65 DAS due to application of seed treatment with liquid *Azotobacter* @ 25 ml/kg + foliar spray of liquid *Azotobacter* @ 25 ml/lit (13.33) over uninoculated control (11.33). The results are in conformity with the observation recorded by Jackson *et al.* (1964) found that inoculation with *Azotobacter* accelerate the stem and leaf growth of tomato. Similar findings were reported by

Chandrikapure *et al.* (1999) that number of leaves was maximum in marigold with treatment of 100 per cent N + *Azotobacter* + PSB. Ingle *et al.* (2008) noted the significantly increased growth characters like height of plant, number of leaves by application of liquid biofertilizers such as PSB and *Azotobacter* in okra.

Effect on length and breadth of leaves

The data on length and breadth of leaves were recorded at 25 DAS shown statistically non-significant that indicates there was no any remarkable difference either in increasing or decreasing the length and breadth of leaves. However, there was marginal increase in length and breadth of leaves ranged from 11.57 to 14.37 cm and 2.97 cm to 4.30 cm respectively, over uninoculated control. The results found statistically significant at 45 DAS which ranged from 12.80 cm to 15.73 cm length of leaves and 4.07 cm to 5.63 cm breadth of leaves. The significantly maximum length of leaves observed at 65 DAS which found with application of seed treatment with liquid *Azotobacter*@ 25 ml/kg + foliar spray of liquid *Azotobacter* @ 25 ml/lit (19.60 cm) over uninoculated control (14.37 cm) and breadth of leaves 57 found significant with same treatment (8.70 cm) over rest of treatments and uninoculated control (6.80 cm), respectively. The results were similar to that of Narayan *et al.* (2007) showed treatment with *Azotobacter*, PSB along with application of chemical fertilizers in tomato and Singaravel *et al.* (2008) in okra, Dastager *et al.* (2010) in cowpea and also found by Radhakrishnan and Mallikarjunaiah (1983) in vegetable crop. Bouthaina *et al.* (2010) indicated that the plant height (cm), root length, leaf area and leave per plant increased significantly with bio-fertilizer treatments.

Effect on height of plant

The results on plant height revealed that an application of liquid biofertilizers and its seed inoculation significantly increased the respective growth parameters over uninoculated control. The results show gradually increase in plant height from 25 DAS to 65 DAS and maximum height of plant

observed at 65 DAS, which ranged from 53.40 cm to 84.23 cm. The significantly higher plant height (84.23 cm) was recorded due to application of seed treatment with liquid *Azotobacter* @ 25 ml/kg + foliar spray of liquid *Azotobacter* @ 25 ml/lit over rest of the other treatments and uninoculated control. The similar findings were obtained by Reddy and Lakhdive (1982) in sorghum when seed was inoculated with *Azotobacter* and Radhakrishnan and Mallikarjunaiah (1983) in vegetable crop. Ingle *et. al.* (2008) found that significantly increase growth parameters like plant height of okra by application of liquid biofertilizers such as *Azotobacter* and PSB and also by Singaravel *et. al.* (2008).

Effect on leaf area index

The results on leaf area index were found statistically significant over uninoculated control and it was ranged from 34.13 cm² to 73.33 cm² at harvest stage. The significantly maximum values noticed in seed 58 treatment with liquid *Azotobacter*@ 25 ml/kg + foliar spray of liquid *Azotobacter*@ 25 ml/lit (73.33 cm²), over rest of all other treatments and uninoculated control. The similar results found by Shivappa *et. al.* (1976) and Khullar *et. al.* (1978), they observed that significant increase in leaf surface area of chilli due to *Azotobacter* inoculation. Bauthaina *et. al.* (2010) indicates that plant height, leaves per plant and leaf area increased significantly with biofertilizers treatment.

Effect on fresh and dry weight of leaves

The data on fresh and dry weight of leaves found statistically significant and maximum at 65 DAS which ranged from 9.99 g to 24.06 g and 2.77 g to 7.42 g, respectively. The significantly maximum fresh weight of leaves observed in seed treatment with liquid *Azotobacter*@ 25 ml/kg + foliar spray of liquid *Azotobacter*@ 25 ml/lit (24.06 g) over uninoculated control and maximum dry weight of leaves noticed in same treatment (7.42 g) over uninoculated control. The similar findings noticed by Debnath (1997) reported significant improvement in fresh and dry weight of plant in gladiolus which inoculated with *Azotobacter*, over uninoculated control. Bum

Ki Park *et al.* (2005) reported that enhancement of lettuce growth resulted in increase of lettuce fresh weight by 23 per cent due to application of phosphate biofertilizers and also found by Bhadoria *et al.* (2005) in tomato due to treatment with *Azotobacter* culture. Bouthaina *et al.* (2010) indicated that the root length, shoot and root fresh and dry weights, leaf area and leave per plant increased significantly with biofertilizer treatments.

Effect on total dry matter weight

All the above maintained growth promoting character helped significant increasing the shoot and root biomass leads to increase in total dry matter weight of plant. The data on total dry matter weight revealed 59 that an application liquid bioinoculants which increased the dry matter weight over uninoculated control. All inoculants recorded different dry matter weight of shoot ranged from 7.93g to 13.35g, dry weight of root ranging from 0.99 g to 0.17 g and total dry matter weight ranged from 8.10g to 14.34 g. The significantly maximum dry matter weight of plant was observed in seed treatment with liquid *Azotobacter* @ 25 ml/kg + foliar spray of liquid *Azotobacter* @ 25 ml/lit (14.34 g) it was superior over all other treatments and uninoculated control. The seed inoculation with liquid biofertilizers fixes nitrogen and solubilize insoluble phosphate in soil and make available to plants due to which plant grows vigorously producing more shoot and root biomass. Similar results were found by Reddy and Lakhdive (1982) in hybrid shorghum, Dibut *et. al.* (1993) in onion due to inoculation with liquid *Azotobacter* and Debnath (1997) in case of gladiolus. Deokar and Sawant (2001) observed that biofertilizer significantly increased the dry matter yield of chilli. Singaravel *et al.* (2008) reported that liquid biofertilizers significantly increased the growth character and dry matter weight of okra.

Effect on yield

There was significant increase in yield of lettuce due to application of liquid *Azotobacter* and liquid PSB and it was varied between 9.99 g plant-1 to 24.06 g plant-1. The maximum

yield of lettuce was recorded with application of seed treatment with liquid *Azotobacter* @ 25 ml/kg + foliar spray of liquid *Azotobacter* @ 25 ml/lit (24.06g) over uninoculated control. The results show conformity with Shivappa et al. (1976) reported that maximum response to *Azotobacter* inoculation was observed in vegetable crops than in cereals. They also observed that yield of chilli was increased from 12 to 39 per cent due to *Azotobacter* inoculation. Tomar et al. (1996) found the significant effect of phosphate solubilizing 60 bacteria along with phosphorus on growth parameters and yield of gram. Kedar et al. (2002) concluded that there was 18 per cent increase in grain yield due to using *Azotobacter* inoculants only over the control. Scherer (2006) evaluated the effect of foliar application of biofertilizers on growth and yield of common bean. Tesfaye et al. (2007) the finding demonstrated that, saving of 50 kg N ha⁻¹ and an average increase of 13.5 per cent marketable yield of onion due to *Azotobacter* inoculation in the presence of 75 kg N ha⁻¹. Gaikwad et al. (2008) carried out field experiment with liquid *Azotobacter chroococcum* along with foliar spray which significantly increase for weight, grain and dry matter yield of groundnut. Hernando et al. (2011) confirmed that the weight, diameter and yield of lettuce and cabbage favoured with the biofertilizer applications.

SUMMARY AND CONCLUSION

Application of liquid *Azotobacter* and liquid phosphorus solubilizing bacteria increase growth parameters and yield of lettuce crop significantly over uninoculated control.

Effect of inoculation with liquid *Azotobacter* and liquid PSB inoculants on the growth of lettuce plants revealed that seed treatment with liquid *Azotobacter* + foliar spray of liquid *Azotobacter*, was found superior amongst all the treatments conducted in respect of number of leaves (13.33), length of leaves (19.60 cm), breadth of leaves (8.70 cm), height of plant (84.23 cm), leaf area index (73.33 cm²), fresh weight 72 (24.06 g) and dry weight (7.42 g) of leaves, total dry

matter weight (14.34 g), yield (24.06 g) over uninoculated control.

Thus, the above studies indicate that the liquid *Azotobacter* and liquid PSB were shows positive effect in respect of growth and yield of lettuce as well as biochemical properties of soil.

REFERANCES

- Bhadoria, S. K. S., Dwivedi, Y. C. and Kushwah S. S. 2005. Effect of *Azotobacter* inoculation with nitrogen level on quality character of tomato. Veg. Sci. 32 (1) : 94-95.
- Bouthaina, A. E. G., Rhawhia A. M., Tomader E. R., Mona Morsy E. S. 2010. Effect of some soil microorganisms on soil properties and wheat production under north Sinai conditions. J. of Appl. Sci. Res. 4(5): 559-579.
- Bum Ki Park, J. H. 2005. Effect of phosphate biofertilizer produced by *Enterobacter intermedium* on rhizosphere soil properties and lettuce growth. Korean J. Soil Sci. Fert. 38 (1) : 15-24.
- Chandrikapure, K. R., Sadawarte, K. T., Panchbhai, D. M. and Shelke, B. D. 1999. Effect of bioinoculants and graded doses of nitrogen on growth and flower yield of marigold (*Tagetes erecta* L.). Orissa J. Horti. 27 (2) : 31-34
- Chin, M. W. 2010. The best organic liquid biofertilizer on earth replacing chemical fertilizers. EzineArticle.com <http://ezinearticles.com/>. The best organic liquid biofertilizer on earth replacing chemical fertilizer.
- Dastager, S. G., Deepa, C. K. and Pandey, A. 2010. Isolation and characterization of novel plant growth promoting *Micrococcus* spp. NII-0909 and its interaction with cowpea. Plant Physiol. Biochem. 48 (12) : 987-992.
- Debnath, S. 1997. Studies on *Azotobacter* with special reference to gladiolus. M. Sc. (Agri). Thesis submitted to MPKV, Rahuri (M.S.), India.

- Deokar, K. P. and Sawant, D. M. 2001. Inhibition of cucumber mosaic virus in chilli by biofertilizers. J. of Maharashtra Agric. Univ. 26 (3) : 276-279.
- Dibut, B. R., Martinez, R. and Ganzalez, R. 1993. Stimulation of growth and yield of onions by bacterization in red ferralitic soils. Edn. By Villegas INICA (1) : 223-225.
- Gaikwad, A. L., Deokar, C. D., Shete, M. H. and Pawar, N. B. 2008. Studies on effect of phyllosphere diazotrophs on growth and yield of groundnut. J. Plant Disease Sci. 3 (2).
- Goldstein, A. H. 1996. Bacterial solubilization of mineral phosphates : historical perspective and future prospects. Am. J. Altern. Agric. 1 : 51-57.
- Gyaneshwar, P., Kumar, G. N., Parekh, L. J. and Poole, P. S. 2002. Role of soil microorganisms in improving phosphorus nutrition of plants. Plant and soil. 245 : 83-93.
- Hernando, C., Tulio, L., Edwin, P. and Ruth, P. 2011. The effect of three liquid biofertilizers in the production of lettuce (*Lactuca sativa* L.) 77 and cabbage (*Brassica oleracea* L. var *capitata*). Agronomia Colombiana 29 (3) : 415-421.
- Ingle, V. G., Tatar, P. G., and Raut, U. A. 2008. Effect of biofertilizer with reduced doses of nitrogen on growth of okra. Annuals Plant Physio. 22 (2) : 255-258.
- Jackson, R.M., Brown, M.E. and Burlingham, S.K. 1964. Effect of *Azotobacter chroococcum* and gibberellins on germination, growth and yield of tomato. Nature, London. 203: 851-852.
- Jadhav, S.W. and Patil, P.L. 1985. Effect of *Azotobacter* on growth and yield of paddy. Indian J. Microbiol. 21 (3): 212-213.
- Kader, M. A., Mian, M. H. and Hoque, M. S. 2002. Effects of *Azotobacter* inoculant on the yield and nitrogen uptake by wheat. Online J. Biol. Sci., 2: 259-261.
- Khullar, S.S., Chahal, V.P.S. and Kaur, P.P. 1978. Effect of *Azotobacter* inoculation on the chlorophyll content and other characters of carrot, radish, bringal and chilli. Indian J. Microbiol. 18 (2): 138-140.
- Mahato, P., Badoni, A. and Chauhan, J. S. 2009. Effect of *Azotobacter* and nitrogen on seed germination and early seedling growth in tomato. Researcher. 2009;1(4):62-66
- Pathak, D. V., Singh, S. and Saini, R. S. 2013. Impact of bio-inoculants on seed germination and plant growth of Gauva (*Psidium guajava*) J. of Hort. and forest. Vol. 5(10), pp. 183-185.
- Radhakrishnan, D. and Mallikarjuniah R. R. 1983. Effect of bacterial inoculants on growth of palak and amaranthus. Vith Southern Reg. Conf. On Microbial Inoculants, held at U.A.S Bangalore on Sep. 22-23 (Abst.) pp. 16.
- Rao, D. L. N. 2004. Edited paper on "Biofertilizer Research Progress work 2004-2007". All India Network Project on Biofert., Indian Inst. of Soil Sci., ICAR, Bhopal.
- Rao, D. L. N. 2007. Edited paper on "Biofertilizer Research Progress work 2004-2007". All India Network Project on Biofert., Indian Inst. of Soil Sci., ICAR, Bhopal.
- Reddy, G.V. S. and Lakhdive, B.A. 1982. Effect of seed sorghum (CSH5). M.Sc. (Agri.). Thesis submitted to P.D.K.V. Akola (M.S.), India.
- Sajindranath, A. H, Narwadkar, P. R., Prabhu, T. and Rathod, N. G. 2002. Effect of biofertilizers and growth regulators on germination in okra. South-India Hort. 50 (4-6) : 538-542.
- Scherer, E. E. 2006. Common bean response to liquid biofertilizer application. Agrappecuoria- Catarinense. 19 (1): 85-88.
- Sharma, K., Dak G., Agrawal, A., Bhatnagar, M. and Sharma, R. (2007). Effect of

- phosphate solubilizing bacteria on the germination of *Cicerarietinum* seeds and seedling growth. J. Herb. Med. and Toxicol. 1:61-63.
- Shivappa, K.S., Khan, A.N. A., Subramanyan, M.B. and patil, R.B. 1976. Studies on the Azotobacter inoculation of crop plants. Madras agric. J. 63 (11-12): 606-610.
- Singaravel, R., Suhatiaa, K., Vembu G. and Kamraj, S. 2008. Effect of liquid biofertilizer on the nutrient content and uptake of okra. Asian J. Soil. Sci. 3 (2) : 217-219.
- Tesfaye B., Pal, N. and Saxena A. K. 2007. Response of onion (*Allium cepa* L.) to combined application of biological and chemical nitrogenous fertilizers. Acta. Agriculturae Slovenica, 89 (1) : 107- 114.
- Tomar, R.K.S., Namdeo, K. N. and Ranghu J. S. 1996. Efficacy of phosphate solubilizing bacteria biofertilizers with phosphorus on growth and yield of gram (*Cicer arietinum*). Ind. J. Agron. 41 : 412- 415.