Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2018; SP1: 3199-3201

TR Barathkumar
Department of Horticulture, Faculty of Agriculture, Annamalai University, Annamalai nagar, Chidambaram, Tamil Nadu, India.

K Manivannan
Department of Horticulture, Faculty of Agriculture, Annamalai University, Annamalai nagar, Chidambaram, Tamil Nadu, India.

Correspondence
TR Barathkumar
Department of Horticulture, Faculty of Agriculture, Annamalai University, Annamalai nagar, Chidambaram, Tamil Nadu, India.

Studies on the effect of biofertilizers on growth, yield and alkaloid content of Ashwagandha roots (*Withania somnifera* Dunal.)

TR Barathkumar and K Manivannan

Abstract
Ashwagandha (*Withania somnifera* Dunal.) is one of the important medicinal plants attaining economic importance in recent days. The experiment was carried out at Department of Horticulture, Faculty of Agriculture, Annamalai University, Annamalai nagar.The treatments were conducted with the seedling dip of biofertilizers *viz.*, *Azospirillum* at the rate of 500 g per hectare, phosphobacteria at the rate of 500 g per hectare and combination of *Azospirillum* + phosphobacteria at the rate of 1 kg per hectare. The study revealed that maximum number of 5.69 roots per plant and root length of 38.16 cm were observed in plants treated with *Azospirillum* + phosphobacteria. Significantly higher root diameter of 1.14 cm and root yield of 6.03 q ha^{-1} was recorded in the application of *Azospirillum* + phosphobacteria. The maximum total alkaloid content and alkaloid yield of 1.379 per cent and 8.32 kg ha^{-1} respectively were registered when the plants treated with *Azospirillum* + phosphobacteria. Hence, the growth, yield and total alkaloid content of ashwagandha roots were increased favourably by the combined application of *Azospirillum* and phosphobacteria followed by phosphobacteria alone.

Keywords: biofertilizers, growth, yield and Ashwagandha roots

Introduction
Ashwagandha, which is an herb widely used in Ayurveda, is gaining importance in the modern medicine also. The root after drying yield the alkaloids such as withanine and somniferine, which are widely used in pharmaceutical preparation, by different drug, companies in south India. This crop is also mainly used as an ingredient of medicaments prescribed for curing disability and sexual weakness in the male. It is no wonder that ashwagandha has been equated to ginseng (*Panax ginseng* L.) and is popularly known as the "Indian ginseng". It is cheap and readily available hence it is draws the attention of the industrialists and farmers because of the increasing demand for its roots in pharmaceutical industry. But at present, the cultivation of this crop is not being done in a scientific manner to exploit the full potential. The economic potential of this medicinal plant could be realized by expanding its cultivation to better agricultural lands with efficient crop management.

In recent days, among the various production technologies, the efficiency of plants *viz*., improving the production and quality can be made by the addition of biofertilizers. The use of biofertilizers has resulted in increased yields and reduced requirements of chemical fertilizers (Subba Rao *et al.*, 1980). So far no work has been attempted on the use of biofertilizers in the production of ashwagandha.

Materials and Methods
The experiment was undertaken at Department of Horticulture, Faculty of Agriculture, Annamalai University, Annamalai nagar, Chidambaram, Tamil Nadu, India. The two raised seedbeds of 2m length, 1m width and 20 cm height were prepared. To each bed, 5kg of well decomposed farm yard manure were applied and mixed well. The seeds were treated with capton at the rate of 2 g per kg of seeds, mixed with sand and sown 2.0cm apart in 15 cm lines spaced. The beds were mulched with paddy straw and watering was done daily. The seedlings were transplanted 60 days after sowing. The treatments were conducted with the seedling root dip of biofertilizers *viz*., *Azospirillum* at the rate of 500 g per hectare, phosphobacteria at the rate of 500 g per hectare and combination of *Azospirillum*+phosphobacteria at the rate of 1 kg per hectare. The *Azospirillum* and phosphobacteria inoculant slurries were prepared individually by the mixing of *Azospirillum* inoculant and phosphobacteria inoculant respectively and the combination of *Azospirillum* and phosphobacteria inoculant slurry was also prepared in the same procedure. All the biofertilizers were inoculated by the root dipping

method before transplanting as per the treatments. The observations were recorded for the all growth and yield of ashwagandha roots. The total alkaloid content was estimated as per the procedure given by Srivastava and Iyer (1960). The population of *Azospirillum* and phosphobacteria were enumerated by MPN method (Dobereiner, 1980). The nitrogen, phosphorous and potassium content of plant tissues was estimated by micro kjeldhal method (Yoshda, 1976), vanadomolybdate phosphoric yellow colour method (Jackson, 1973) and flame photometer method (Jackson, 1973) respectively.

Resuluts and Discussion

The influence of biofertilizers on the number of roots, root length, root diameter, root yield, total alkaloid content and alkaloid yield were recorded (Table-1).

Azospirillum+phosphobacteria root dip of seedlings yielded significantly more number of 5.69 roots followed by phosphobacteria alone (4.01) and *Azospirillum* alone (3.73) than the control (2.40). The maximum root length of 38.16 cm was observed with in the plants treated with *Azospirillum* + phosphobacteria followed by phosphobacteria alone (32.12 cm) over the untreated plants (24.60 cm). This may be due to the production of enzymatic root exudate produced when phosphobacteria associated with the crop roots. This was in agreement with the findings of Chezhiyan *et al*. (2003) in *Phyllanthus amarus.* Application of both *Azospirillum* + phosphobacteria significantly increased root thickness of 1.14 cm followed by phosphobacteria alone (1.01 cm) over the control (0.86 cm).

The combination of *Azospirillum* + phosphobacteria produced highest root yield of 6.03 q ha^{-1} followed by phosphobacteria alone (3.98 q ha^{-1}) than that of untreated plants (2.28 q ha^{-1}). This may be due to the synthesis of growth substances and uptake of more nutrients. These results were in agreement with the reports of Sathiyaraj *et al*. (2006) in *Coleus forskohlii*.

The maximum total alkaloid content (1.379 per cent) and alkaloid yield (8.32 kg ha^{-1}) were registered when the plants were treated with the combined application of *Azospirillum* and phosphobacteria. This may be due to the enhanced foliage growth and assimilation rate by nitrogen and growth substances supplied by *Azospirillum* and more uptake of phosphorous due to increased availability of phosphorous in absorbable form by phosphobacteria favourable interaction of biofertilizers with the production of phytohormones and carbohydrates.

There was a profound positive influence on the microbial population when both the *Azospirillum* and phosphobacteria were inoculated. The same trend was seen when either one of the biofertilizers were inoculated on the specific microbial population. (Table-2)

The uptake of major nutrients of nitrogen, phosphorus and potassium which serves as a diagnostic tool for understanding the cell physiology of the plant at various nutrient levels were estimated. Among the biofertilizers, the combined application of *Azospirillum* and phosphobacteria significantly increased the nitrogen uptake with giving the maximum effect of 473.0mg $plant^{-1}$ (Table-2). The favourable interaction between *Azospirillum* and phosphobacteria resulted in higher nitrogen uptake. The next best in order was the application of *Azospirillum* alone treated plant (458.33 mg $plant^{-1}$) because it uses pectin as the sole carbon source. The pectinolytic activity might be involved in a hydrolysis of the middle lamellae of *Azospirillum* colonized cortical cell without causing cell collapse, which may accelerated water and nutrient uptake as stated by Gopal and Paramaguru (2006) in senna and Rajeswari and Arumugam Shakila (2015) in Ambrette.

The combined application of *Azospirillum* and phosphobacteria favorably showed the maximum amount of 21.31 mg $plant^{-1}$ of phosphorous uptake this may be due to the synergetic interaction between *Azospirillum* and phosphobacteria in keeping a balanced nutrient ratio in the plant cells. The next best in order was the application of phosphobacteria alone (19.96mg $plant^{-1}$) treated plants. Application of phosphobacteria significantly increased the uptake of phosphorous at all stages. Phosphate solubilization was brought about by the production of organic acids, some of which have been identified as malic, glyoxalic, succinic fumaric, citric and alpha ketoglutaric acids. Other acids like lactic and 2-ketogluconic acids acted as powerful chelator of calcium while humic acid, fulvic acids from stable complexes with iron and aluminium phosphate and thus increased the quality of phosphorus due to phosphobacteria in the present study was similar to the earlier findings of Tiwari and Kulmi (2005) in isabgol and Senthilkumar and Sivagurunathan (2012) in green gram and cow pea.

Application of *Azospirillum* and phosphobacteria significantly recorded the potassium uptake with the maximum effect of 446.39 mg $plant^{-1}$. The next best in order was *Azospirillum* alone (437.57 mg $plant^{-1}$) applied plants. This may be due to the higher production of dry matter and higher content of potassium in the plants, which was in accordance to the reports Gopal and Paramaguru (2006) in senna and Rajeswari and Arumugam Shakila (2015) in Ambrette.

Though the effect of biofertilizers were not significant themselves. Whereas, all the biofertilizers applications were either together or individual giving significant differences when compared to control plants.

The findings revealed that the maximum values of number of roots, root length, root diameter, root yield, total alkaloid content and alkaloid yield were recorded when the *Azospirillum*+phosphobacteria followed by phosphobacteria alone. Thus, from the above information, it can be stated that the combined application of *Azospirillum*+phosphobacteria followed by phosphobacteria alone favourably influenced the growth, yield and alkaloid content of ashwagandha roots.

Table 1: Effect of biofertilizers on root parameters, root yield and alkaloid yield of ashwagandha roots

Treatment	No. roots $plant^{-1}$	Root length (cm)	Root diameter(cm)	Root yield (q ha^{-1})	Total alkaloid Content (%)	Alkaloid yield (kg ha^{-1})
Control	2.40	24.60	0.86	2.28	0.452	1.03
Azospirillum	3.73	29.96	0.95	3.54	0.458	1.62
Phosphobacteria	4.01	32.12	1.01	3.98	0.703	2.80
Azospirillum + Phosphobacteria	5.69	38.16	1.14	6.03	1.379	8.32
S.E±.	0.64	2.61	0.04	0.60	0.324	0.39
CD at 5%	1.29	5.23	0.08	1.22	0.651	0.79

Table 2: Influence of biofertilizers on NPK uptake and population of *Azospirillum* and phosphobacteria

Treatment	*Azospirillum* ×10^5 g of soil^{-1}	Phosphobacteria ×10^5 g of soil^{-1}	Nitrogen (mg plant^{-1})	Phosphorus (mg plant^{-1})	Potassium (mg plant^{-1})
Control	6.90	8.10	435.93	12.15	422.27
Azospirillum	18.00	13.00	458.33	16.35	437.57
Phosphobacteria	14.10	16.90	450.51	19.96	431.24
Azospirillum + Phosphobacteria	20.90	18.98	473.00	21.31	446.39
S.E. ±	0.97	0.50	7.21	1.96	3.92
CD at 5%	1.97	1.01	14.45	4.01	8.18

References

1. Dobereiner J. Forage grasses and grain crops. In: Methods for evaluating biological nitrogen fixation. Bergersen F.J. (ed.). John wiley and Sons, New York, 1980, 535-555.
2. Jackson ML. Soil chemical analysis. Prentice Hall of India Pvt. Ltd., New Delhi, 1973.
3. Srivastava S, Iyer K. An easy method for the estimation of total alkaloids in Ashwagandha. *Indian J Pharm.* 1960; 22:97-98
4. Subba Rao NS, Tilak KVBR, Lakshmikumari M, Singh CS. *Azospirillum* a new bacterial fertilizer. *Indian Fmg.* 1980; 30:3-5.
5. Yoshida S, Forno DA, Cook JH, Gomez KA. Laboratory manual for physiological studies of rice. III ed., International Rice Research Institute, Manila, Philippines, 1976, 7-76.
6. Chezhyian N, Saraswathy S, Vasumathi R. Studies on the effect of organic manures, biofertilizers and plant density on growth, yield and alkaloid content of bhumyamalaki (*Pyllanthus amarus* Schum and Thohn). South Indian Hort. 2003; 51(1-6):96-101.
7. Gopal NO, Paramaguru P. Synergistic interaction of Azospirilum and Phosphobateria solublizing bacteria for enhancing the growth, yield and rhizosphere microbial dynamics of Senna (*Cassia angustifolia* Vahl.) In: International conference on globalization of traditional, complementary and alternative systems of medicine, HC&RI, TNAU, Coimbatore, Tamil Nadu, India. 2006, 38.
8. Rajeswari R, Arumugam Shakila. Effect of integrated nutrient management practices on yield characters of Ambrette (*Abelmosches moschatus* Medic.) Plant Archives. 2015; 15(1):537-540.
9. Senthilkumar PK, Sivagurunathan P. Comparative effect on growth and yield of green gram (*Phaseolus radiate* L.) and cow pea (*Vigna sinensis* Edhil.) Int. J Curr. Microbiol. App. Sci 2012; 1(1):34-39.
10. Sathiyaraj M, Thirumurugan V, Balasubramanian R. Effect of organic, inorganic fertilizers and plant growth substances on productivity of *Coleus forskohlii*. In: International conference on globalization of traditional, complementary and alternative systems of medicine, HC&RI, TNAU, Coimbatore, Tamil Nadu, India. 2006, 76.
11. Tiwari PN, Kulmi GS. Effect of biofertilizers, organic, inorganic attrition on growth and yield of Isabgol (*Plantago ovata* Forsk) Research on crops 2005; 6(3):568-571.