Effect of rhizobacterial inoculation on withaferin - A content of ashwagandha (var. Jawahar 20) roots

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Plant growth promoting rhizobacteria *viz.*, *Azospirillum*, *Azotobacter*, *Bacillus* and *Pseudomonas* were isolated from rhizosphere soil and roots of ashwagandha plants collected from various locations in Tamil Nadu. The isolated strains were characterized by morphological, physiological and biochemical tests and were examined for nitrogen fixation, phosphate solubilization, phytohormone production, siderophore production and antagonistic activity. A pot culture experiment was conducted at the dept of Agricultural Microbiology, Tamil Nadu Agricultural University, Coimbatore. The results revealed that combined inoculation of *A. lipoferum* - AAs-11, *Azotobacter* - AAz-3, *Bacillus* - APb-1 and *Pseudomonas fluorescens* - APs-1 enhanced the biochemical constituents of ashwagandha such as chlorophyll, protein and total alkaloid contents, especially Withaferin-A.

Key words : Rhizobacteria, Aswagandha, Withaferin-A

INTRODUCTION

The indigenous systems of medicine namely Siddha, Ayurveda and Unani have been in existence for several centuries. The WHO has estimated that over 80 per cent of the world population meets their primary health care needs through traditional medicine (Lambert, 1997). Ashwagandha is used as a tonic in geriatrics, being efficacious in relieving hand and limb tremors of people at old age (Atal et al., 1975). It has been equated to ginseng (Panax ginseng) of China and is popularly known as the "Indian Ginseng". The most important pharmacological use of ashwagandha is as adaptogen with antistress antioxidant, antitumor, anti-inflammatory, mind boosting and has rejuvenating properties (Singh et al., 1990). The biofertilizers are ecofriendly and low cost technology and their application may play a major role in soil fertility, nutrient transformation, crop sanitation and sustainability. The rhizobiocoenosis is an important biological process that plays a major role in satisfying the nutritional requirement of these crops. Studies on the rhizobacterial population in the rhizosphere region and testing the suitability of the isolated rhizobacteria as seed and soil inoculant will be highly useful in improving the productivity and quality of this commercially important medicinal plant.

MATERIALS AND METHODS

A pot culture experiment was conducted during the year 2004 at the Department of Agricultural Microbiology, Tamil Nadu Agricultural University, Coimbatore (T.N.) to study the effect of combined inoculation of rhizobacteria on growth, yield and quality of ashwagandha (var. Jawahar 20). The rhizobacterial isolates *viz.*, *Azospirillum lipoferum*-AAs-11, *Azotobacter*-AAz-3, *Bacillus*-APb-1 and *Pseudomonas fluorescens*-APs-1 were prepared as carrier based inoculants used for this study. The pots were filled with potting mixture (soil + sand + FYM) and the rhizobacteria treated seeds were sown at 25 seeds per pot and finally 5 seedlings were maintained. The experiment was conducted in completely randomized block design with three replications. The analysis of the alcoholic extracts of the root samples were carried out by the method suggested by Velde *et al.* (1983) by using the high performance thin layer chromatography (HPTLC) available at J.S.S. College of Pharmacy, Ooty.

Sample preparation :

Ten gram of root samples were dissolved in 100 ml of methanol and filtered through Whatman No. 42 filter paper. The methanol extracts of root samples were used for direct application on the HPTLC plate.

Preparation of the standard solution :

With a ferin-A at the rate of 5.4 mg was dissolved in 5 ml of methanol so as to get a concentration of $1.08 \text{ mg} \text{ ml}^{-1}$.

Selection of HPTLC plates :

Pre-coated silica gel GF_{254} plates with alumina support in size of 20 x 20 cm were used for the present study.