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RESEARCH ARTICLE

STIMULATIVE EFFECTS OF MORPHOLOGICAL, BIOCHEMICAL AND ENZYME CONTENTS OF NILA VEMBU (*ANDROGRAPHIS PANICULATA* NEES.)

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ABSTRACT

Biofertilizers are the substance that contains microorganism's living or latent cells. bio fertilizers increases the nutrients of host plants when applied to their seeds, plant surface or soil by colonizing the rhizosphere of the plant. The present research work was comprises to valuable medicinal plant of *Andrographispaniculata* for morphological, biochemical, and enzyme contents of various stages of its growth. The inoculants such as *Azotobacterchroococcum*, *Ppseudomonasfluorescens*, and vermicompost are singly and in combinations are allowed to grow in similar environmental conditions. the morphological parameters such as shoot length, root length, leaf length, leaf width, total leaf area, shoot girth, number of branches per plant, plant height were observed. The biochemical namely chlorophyll, "a" chlorophyll "b" and total chlorophyll, and carotewnoids were measured. The enzymes such as catalase, and paroxidase were observed in 30, 60, and 90 days after sowing in *Andrographispaniculata* plants. The maximum growth was measured in combined application of *Azotobacterchroococcum*, *Ppseudomonasfluorescens*, and Vermicompost alone. The minimum growth was measured in control.

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INTRODUCTION

Biofertilizers are living cells of different types of microorganism (bacteria, algae, fungi), which have an ability to mobilize nutritionally important elements from non-usable to usable form. They can be applied to seed, root or in order to soil. They improve the soil fertility and help plant growth and yield by increasing the number and biological activity of desired microorganisms in the root environment (Subba Rao *et al.* 1993). They have attracted greater attention particularly in developing countries like India. *Andrographispaniculata*is a plant that is native to South Asian countries such as India and Sri Lanka. It is frequently used for preventing and treating the common cold and flu (influenza) and is also used for a wide assortment of other conditions. It is used for digestive complaints including, constipation, intestinal gas, colic, and stomach pain for liverconditions including an enlarged liver, jaundice, and liver damage due to medications for infections pneumonia, tuberculosis, wounds, ulcers and itchiness. Some people use *Andrographis* for sore throat, coughs, woolen tonsils, bronchitis, and allergies.

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It is also used for "hardening of the arteries" (atherosclerosis), and prevention of heart disease and diabetes. Other uses include treatment of snake and insect bites, loss of appetite, kidneyproblems (pyelonephritis), hemorrhoids, and an inherited condition called familial Mediterranean fever. *Andrographispaniculata* is also used as astringent, bacteria killing agent, painkiller, fever reducer, and treatment for worms. It is an erect annual herb that grows 30 to 110 cm in height and is native to India, China, and Southeast Asia. It is widely cultivated in Asia.

The increase in crops yield has been reported to be 60 per centor more due to the use of chemical fertilizers. This undoubtedly boosted the food production but at the same time, indiscriminate and injudicious use of fertilizers has caused considerable damage to the environment through air, water and soil pollution (Yadav and Lourduraj, 2005a). It has also leaded to poor soil fertility and in turn affected the yield. Careful and scientific nutrient management can help in better crop yields, soil fertility and fertility economy (Jawahar and Suresh, 2007). All synthetic fertilizers must be replaced by organic fertilizerssuch as compost, farmyard manure, green manure and bio fertilizers. This organic farming improves soil formation, prevents soil erosion, it allows the soil to retain more nutrients and water and it encourages growth of soil organisms. It benefits water, as it does not pollute groundwater. Biofertilizers are cost effective, ecofriendly and renewable source of plant nutrients to supplement chemical fertilizers in sustainable agricultural system in India.

MATERIALS AND METHODS

The Present Investigation Was Carried To Find Out Effect Of *Andrographispaniculata* Nees. on Morphological Biochemical And Enzyme Studies Were Estimated.

Materials

Fertilizers

Biofertilizers such as *Azotobacterchroococcum*, *Pseudomonas fluorescens*, and vermicompost were collected from Government Recognized Agrocentre, Kumbakonam, Thanjavur District of Tamilnadu. The inorganic fertilizers such as urea as a nitrogen source (N), single super phosphate as a phosphorus source (P) and muriatic of potash as a potassium sources (K) have been purchased from Government Authorized Agrocentre, Kumbakonam, TamilnaduIndia.

Seed materials: The seeds of *Andrographispaniculata* was obtained from Tamil Nadu Rice Research Institute, Regional Research Station of Tamil Nadu Agricultural University, Aduthurai, Thanjavur District, Tamil Nadu, and India.

Name of the fertilizers	Treatment details	
Control	T1	Crop without fertilizer
Azotobacterchrococcum+ *Pseudomonasfluorescens*	T2	AZ+PF - 2+2 Kg/ha
Azotobacterchrococcum+ *P. fluorescens* + Vermicompost	T3	AZ+PF+VC- 2Kg+2Kg+2 t/ha
Azotobacterchrococcum+ Vermicompost+ Chemical fertilizers	T4	AZ+VC+CF - 2+2+ t/ha+110 Kg/ha
P. fluorescens + Vermicompost+ Chemical fertilizers	T5	AZ+PF+VC+CF - 2 + 2 + 2 t/ha+110 Kg/ha
Azotobacterchrococcum +*P. fluorescens*+ Vermicompost+Chemical fertilizer	T6	AZ+PF+VC+CF - 2 + 2 + 2 t/ha+110 Kg/ha

Application of fertilizers: The recommended doses of organic manures such as vermicompost (2 t/ha) were applied in single application. In addition, the combined application of inorganic fertilizers such as nitrogen (50 kg/ha), phosphorus (40 kg/ha) and potassium (30 kg/ha) were in single as well as in combination (110 kg/ha) were applied. The biofertilizerssuch as Nitrogen biofertilizers (*Azotobacterchrococcum* + *Pseudomonasfluorescens 2*+2 kg/ha), were applied in single as well as in combination (4kg/ha) chemical fertilizers such as N,P and K fertilizers in single and in combination alone and vermicompost is 2t/ha of all in the treatments.

Morphometric analysis: The morphological parameters such as shoot and root length, leaf length, leaf width, total leaf area, shoot girth, number of branches per plant, and plant height were measured in centimeter by plant parts were also estimated.

Biochemical analysis: Chlorophylland carotenoids by Arnon 1945

Enzymatic activity: The activities of catalase and peroxidase in nilavembu were estimated and recorded in the plants grownin pot culture experiments were analysed.

Enzyme assay

Catalase: Catalase activity was measured by the method of Machly and Chance (1967). One gram of leaf sample was homogenized in 10 ml of 0.1 M phosphate buffer (pH 7) and centrifuged at 4°C for 10 minutes at 10,000 rpm. An aliquot of 1 ml of the supernatant of the enzyme extract was added to the reaction mixture containing one ml of 0.01 M H_2O_2 and 3 ml of 0.1 M phosphate buffer. The reaction was stopped after incubation of 5 minutes at 20°C by adding 10 ml of one per cent H_2SO_4. The acidified medium without or with the enzyme extract was titrated against 0.005 N $KMnO_4$ and catalase activity was expressed as 'n' moles of H_2O_2 utilized (units min/mg/protein).

Peroxidase (Machly and Chance, 1967)

Donor: Hydrogen peroxidantoxireductase

One gram of fresh plant material was homogenized with 20 ml of ice-cold extraction medium containing 2 mM $MgCl_2$, 1mM EDTA, 10 mm-mercapto ethanol, 7 per cent PVP and 10 mm sodium meta-bisulphate. The homogenate was stained through two layers of cheesecloth and centrifuged at 10,000 rpm for 15 minutes. The supernatant was made upto 20 ml with the same buffer and it was used as the source of enzyme.

Assay: Assay mixture of peroxidayse contained 2 ml of 0.1 M phosphate buffer (pH 6.8), 1 ml of 0.001 M pyrogallol, 12 ml of 0.005 M hydrogen peroxidase and 0.5 ml of enzyme extract. The solution was incubated for 5 minutes at 25°C, after which the reaction was terminated by adding 1 ml of 2.5 N sulphuricacid.

The amount of purpurogallin formed was determined by reading the absorbance at 420 nm against a blank prepared by adding the extract after the addition of 2.5 N sulphuric acids. The activity was expressed in unit = 0.1 absorbance mg/protein/min.

RESULTS AND DISCUSSION

Nature can be considered as the ultimate chemist. About 80% of the world's inhabitants still depend on natural products that have inspired chemists and physicians for years because of their rich structural diversity and complexity (Divya*et al.,* 2011). *Andrographispaniculata* is a plant that has been effectively used in traditional aian medicines for countries. *Andrographispaniculata* family Acanthaceae, it is perceived "blood purifying" property results in its use in diseases where bload "abnormalities" are considered causes of disease, such as skin eruptions, boils, scabis, and chronic undetermined fevers. The aerial part of the plant, used medicinally, contains a large number of chemical constituents, mainly lactones, deterpenoidsglycocides. (Yang *et al.,* 2009).*Andrographispaniculata* having antibacterial, antifungal, antiviral, choleretic, hypoglycemic, hypocholesterolemic, and adaptogenic effects (Bhatnager *et al.,* 1961).

Plate I. Effect of combined treatment on growth and development of A.paniculata 30th day

Plate II: Effect of combined treatment on growth and development of *A.paniculata* 60th day

Table 1. Effect of combined application of various fertilizers on morphometric analysis of *Andrographispaniculata* on 30th Days

Treatments	Shoot length (cm)	Root length (cm)	Leaf length (cm)	Leaf width (cm)	Total leaf area(cm)	No of leaves	Shoot girth (cm)	No. of branches	Plant height (cm)
T_1	4.7±0.414	4.4±0.132	3.5±0.105	1.1±0.033	1.9± 0.057	6±0.180	0.1±0.003	-	9.1± 0.273
T_2	8.2±0.246	6.9±0.207	4.2±0.126	2.3±0.069	4.8±0.144	15±0.450	0.1±0.003	-	15.1±0.456
T_3	10.3±0.349	9.6±0.288	5.1±0.153	3.0±0.090	7.7±0.231	49±1.470	0.1±0.003	1±0.003	19.9±0.597
T_4	9.1±0.273	7.3±0.219	4.8±0.144	2.9±0.087	7.0±0.222	44±1.320	0.2±0.006	-	16.4±0.492
T_5	8.3±0.249	7.8±0.234	4.3±0.129	2.6±0.078	5.6±0.168	41±1.230	0.1±0.003	-	16.1±0.483
T_6	12.3±0.369	9.6±0.456	6.2±0.186	4.1±0.123	12.7±0.369	53±1.590	0.3±0.009	1±0.003	21.9±0.657

± Standard Deviation T_1- Crop without fertilizer, T_2- *Azotobacterchroococcum+Pseudomonasfluorescens* - 2+2 Kg/ha, T_3- *Azotobacterchroococcum+ Pseudomonasfluorescens* +Vermicompost - 2Kg+2Kg+2 t/ha, T_4- *Azotobacterchroococcum*+ Vermicompost +Chemical fertilizers - 2+2+ t/ha+110Kg/ha, T_5.*Pseudomonas fluorescens* + Vermicompost +Chemical fertilizers - 2 + 2 t/ha+110Kg/ha and T_6- *Azotobacterchroococcum+Pseudomonasfluorescens* +Vermicompost +Chemical fertilizers - 2 + 2 + 2 t/ha+110Kg/ha

Table 2. Effect of combined application of various fertilizers on morphometric analysis of *Andrographispaniculata* on 60th Days

Treatments	Shoot length (cm)	Root length (cm)	Leaf length (cm)	Leaf width (cm)	Total leaf area(cm)	No of leaves	Shoot girth(cm)	No. Of branches	Plant height (cm)
T_1	5.1±0.150	3.2±0.096	4.5±0.135	1.7±0.051	3.8±0.114	9±0.270	0.4±0.012	-	8.3±0.249
T_2	9.6±0.280	7.3±0.219	5.0±0.150	2.4±0.007	6.0±0.180	48±1.440	0.2±0.006	2±0.060	17.1±0.513
T_3	12.2±0.366	9.4±0.282	4.4±0.132	2.5±0.075	5.5±0.165	54±1.620	0.2±0.006	3±0.090	21.6±0.648
T_4	11.4±0.324	9.2±0.276	4.8±0.144	2.4±0.00)	5.8±0.174	43±1.290	0.2±0.006	2±0.060	20.6±0.618
T_5	10.9±0.327	8.3±0.249	4.3±0.129	2.1±0.063	4.5±0.135	45±1.350	0.2±0.006	2±0.060	19.2±0.576
T_6	16.8±0.540	10.3±0.309	5.2±0.156	2.5±0.075	6.5±0.195	62±1.860	0.3±0.009	4±0.120	27.1±0.813

± Standard Deviation T_1- Crop without fertilizer, T_2- *Azotobacterchroococcum+Pseudomonasfluorescens* - 2+2 Kg/ha, T_3- *Azotobacterchroococcum+ Pseudomonasfluorescens* +Vermicompost - 2Kg+2Kg+2 t/ha, T_4- *Azotobacterchroococcum*+ Vermicompost +Chemical fertilizers - 2+2+ t/ha+110Kg/ha, T_5.*Pseudomonas fluorescens* + Vermicompost +Chemical fertilizers - 2 + 2 t/ha+110Kg/ha and T_6- *Azotobacterchroococcum+Pseudomonasfluorescens* +Vermicompost +Chemical fertilizers - 2 + 2 + 2 t/ha+110K

Plate III: Effect of combined treatment on growth and development of *A.paniculata* 90th day

Table 3. Effect of combined application of various fertilizers on morphometric analysis of *Andrographispaniculata* on 90th Days

Treatments	Shoot length (cm)	Root length (cm)	Leaf length (cm)	Leaf width (cm)	Total leaf area (cm)	No of leaves	Shoot girth (cm)	No. of branches	Plant height (cm)
T_1	7.3±0.219	6.3±0.189	4.7±0.141	1.9±0.057	4.5±0.135	35±0.003	0.1±0.003	2±0.060	13.6±0.408
T_2	11.5±0.345	7.8±0.234	5.1±0.153	2.4±0.072	6.1±0.183	53±1.590	0.25±0.007	4±0.120	21.3±0.639
T_3	14.0±0.420	11.7±0.351	5.1±0.153	2.69±0.78	6.6±0.198	71±2.130	0.31±0.093	5±0.150	25.7±0.771
T_4	13.4±0.402	10.0±1.030	5.1±0.153	2.7±0.081	6.9±0.207	62±1.860	0.26±0.007	4±0.120	23.4±0.702
T_5	13.8±0.141	10.1±0.303	5.0±0.150	2.3±0.174	5.8±0.174	65±1.950	0.27±0.008	4±0.120	23.9±0.717
T_6	20.0±0.618	13.2±0.369	5.3±0.159	2.6±0.078)	6.9±0.207	81±2.430)	0.35(0.015	7±0210	33.8±1.014

± Standard Deviation T_1- Crop without fertilizer, T_2- *Azotobacterchroococcum+Pseudomonasfluorescens* - 2+2 Kg/ha, T_3- *Azotobacterchroococcum+ Pseudomonasfluorescens* +Vermicompost - 2Kg+2Kg+2 t/ha, T_4- *Azotobacterchroococcum+* Vermicompost +Chemical fertilizers - 2+2+ t/ha+110Kg/ha, T_5-*Pseudomonas fluorescens* + Vermicompost +Chemical fertilizers - 2 + 2 t/ha+110Kg/ha and T_6- *Azotobacterchroococcum+Pseudomonasfluorescens* +Vermicompost +Chemical fertilizers - 2 + 2 + 2 t/ha+110Kg/ha.

Table 4. Photosynthetic pigments of *Andrographispaniculata* (mg/g fr. wt.) on 30th Days

Treatment detail	Chlorophyll "a"	Chlorophyll "b"	Total chlorophyll	Carotenoid
T_1	0.480 ±0.014	0.292 ±0.008	0.772 ±0.023	0.210 ±0.006
T_2	0.549 ±0.016	0.311 ±0.009	0.860 ±0.025	0.298 ±0.008
T_3	0.614 ±0.018	0.341 ±0.010	0.954 ±0.028	0.300 ±0.009
T_4	0.523 ±0.015	0.309 ±0.009	0.810 ±0.024	0.280 ±0.008
T_5	0.549 ±0.016	0.311 ±0.009	0.860 ±0.025	0.273 ±0.021
T_6	0.643 ±0.019	0.438 ±0.013	1.081 ± 0.032	0.321 ±0.009

± Standard deviation T_1- Crop without fertilizer, T_2- *Azotobacterchroococcum+Pseudomonasfluorescens* - 2+2 Kg/ha, T_3- *Azotobacterchroococcum+Pseudomonasfluorescens* +Vermicompost - 2Kg+2Kg+2 t/ha, T_4- *Azotobacterchroococcum+* Vermicompost + Chemical fertilizers - 2+2+ t/ha+110Kg/ha, T_5-*Pseudomonas fluorescens+* Vermicompost +Chemical fertilizers - 2 + 2 t/ha+110Kg/ha and T_6- *Azotobacterchroococcum+Pseudomonasfluorescens* +Vermicompost +Chemical fertilizers - 2 + 2 + 2 t/ha+110Kg/ha

Table 5. Photosynthetic pigments of *Andrographispaniculata* (mg/g fr. wt.) on 60th Days

Treatment detail	Chlorophyll "a"	Chlorophyll "b"	Total chlorophyll	Carotenoid
T_1	0.532 ±0.015	0.329 ±0.009	0.861 ±0.025	2.321 ±0.069
T_2	0.650 ±0.019	0.428 ±0.012	1.078 ±0.032	0.350 ±0.010
T_3	0.662 ±0.019	0.438 ±0.013	1.100 ±0.033	0.360 ±0.010
T_4	0.581 ±0.017	0.411 ±0.012	0.992 ±0.029	0.360 ±0.010
T_5	0.017 ±0.578	0.400 ±0.012	0.978 ±0.029	0.332 ±0.009
T_6	0.683 ±0.020	0.493 ±0.014	1.117 ±0.033	0.382 ±0.011

± Standard deviation

T_1- Crop without fertilizer, T_2- *Azotobacterchroococcum+Pseudomonasfluorescens* - 2+2 Kg/ha, T_3- *Azotobacterchroococcum+Pseudomonasfluorescens* +Vermicompost - 2Kg+2Kg+2 t/ha, T_4- *Azotobacterchroococcum+* Vermicompost +Chemical fertilizers - 2+2+ t/ha+110Kg/ha, T_5-*Pseudomonas fluorescens* + Vermicompost +Chemical fertilizers - 2 + 2 t/ha+110Kg/ha and T_6-*Azotobacterchroococcum+Pseudomonasfluorescens* +Vermicompost +Chemical fertilizers - 2 + 2 + 2 t/ha+110Kg/ha

Table.6 Photosynthetic pigments of *Andrographispaniculata* (mg/g fr. wt.) on90thDays

Treatment detail	Chlorophyll "a"	Chlorophyll "b"	Total chlorophyll	Carotenoid
T_1	0.673 ±0.020	0.458 ±0.013	1.131 ±0.033	0.410 ±0.012
T_2	0.709 ±0.021	0.471 ±0.014	1.180 ±0.035	0.455 ±0.013
T_3	0.711 ±0.021	0.488 ±0.014	1.199 ±0.035	0.462 ±0.013
T4	0.689 ±0.020	0.473 ±0.014	1.162 ±0.034	0.436 ±0.013
T_5	0.687 ±0.020	0.470 ±0.014	1.157 ±0.034	0.425 ±0.012
T_6	0.758 ±0.022	0.599 ±0.017	1.357 ±0.040	0.490 ±0.014

± Standard deviation T_1- Crop without fertilizer, T_2- *Azotobacterchroococcum+Pseudomonasfluorescens* - 2+2 Kg/ha, T_3- *Azotobacterchroococcum+Pseudomonasfluorescens* +Vermicompost - 2Kg+2Kg+2 t/ha, T_4- *Azotobacterchroococcum*+ Vermicompost +Chemical fertilizers - 2+2+ t/ha+110Kg/ha, T_5-*Pseudomonas fluorescens* + Vermicompost +Chemical fertilizers - 2 + 2 t/ha+110Kg/ha and T_6- *Azotobacterchroococcum+Pseudomonasfluorescens* +Vermicompost +Chemical fertilizers - 2 + 2 + 2 t/ha+110Kg/ha.

Table 7: Effect of application of organic fertilizers, inorganic fertilizers and biofertilizers on catalase activity (min/mg/protein) of *Andrographispaniculata* on 30, 60 and 90th days

Treatments	30 Days	60Days	90Days
T_1	5.493 ±0.164	6.982 ±0.209	7.322 ±0.219
T_2	7.652 ±0.229	7.848 ±0.235	8.100 ±0.243
T_3	7.958 ±0.238	8.123 ±0.243	8.200 ±0.246
T_4	7.844 ±0.235	7.685 ±0.230	8.000 ±0.240
T_5	7.628 ±0.228	7.428 ±0.222	7.658 ±0.229
T_6	8.100 ±0.243	8.420 ±0.252	8.500 ±0.255

± Standard deviation T_1- Crop without fertilizer, T_2- *Azotobacterchroococcum+Pseudomonasfluorescens* - 2+2 Kg/ha, T_3- *Azotobacterchroococcum+Pseudomonasfluorescens* +Vermicompost - 2Kg+2Kg+2 t/ha, T_4- *Azotobacterchroococcum*+ Vermicompost +Chemical fertilizers - 2+2+ t/ha+110Kg/ha, T_5-*Pseudomonas fluorescens* + Vermicompost +Chemical fertilizers - 2 + 2 t/ha+110Kg/ha and T_6-*Azotobacterchroococcum+Pseudomonasfluorescens* +Vermicompost +Chemical fertilizers - 2 + 2 + 2 t/ha+110Kg/ha

Table8: Effect of application of organic fertilizers, inorganic fertilizers and biofertilizers on peroxidase activity (min/mg/protein) of *Andrographispaniculata* on 30,60, and 90th Days

Treatments.	30 Days	60 Days	90 Days
T_1	12.742 ±0.382	14.280 ±0.428	15.531 ±0.465
T_2	13.995 ±0.419	15.228 ±0.456	16.541 ±0.496
T_3	14.550 ±0.436	16.665 ±0.499	16.739 ±0.502
T_4	13.752 ±0.412	15.338 ±0.460	15.987 ±0.479
T_5	13.654 ±0.409	15.110 ±0.453	15.781 ±0.473
T_6	14.685 ±0.440	15.722 ±0.471	17.382 ±0.521

± Standard deviation T_1- Crop without fertilizer, T_2- *Azotobacterchroococcum+Pseudomonasfluorescens* - 2+2 Kg/ha, T_3- *Azotobacterchroococcum+Pseudomonasfluorescens* +Vermicompost - 2Kg+2Kg+2 t/ha, T_4- *Azotobacterchroococcum*+ Vermicompost +Chemical fertilizers - 2+2+ t/ha+110Kg/ha, T_5-*Pseudomonas fluorescens* + Vermicompost +Chemical fertilizers - 2 + 2 t/ha+110Kg/ha and T_6- *Azotobacterchroococcum+Pseudomonasfluorescens* +Vermicompost +Chemical fertilizers - 2 + 2 + 2 t/ha+110Kg/ha

The chemical fertilizer is the major supplier of nutrients besides organic manures. The continuous and excess use of chemical fertilizers over a longer period of time has resulted in deterioration of soil health and causes less productivity (Yadav and Lourduraj, 2005a). In this context, the role of organic manures and biofertilizers in sustainable agriculture assumes special significance particularly in the present context of very high cost of chemical fertilizers. Organic farming is becoming a major tool for sustaining the soil quality degraded by intensive use of synthetic chemicals for increasing crop production. Therefore, the use of bio-agents as biofertilizers or biopesticides is an integral part of organic farming. The Vermicompost contain plant growth regulating substances including plant growth hormones and humic acids which are probably responsible for increase in germination, growth and yield of plants (Atiyeh *et al.,* 2002; Arancon *et al.,* 2006). Biofertilizers are cost effective, ecofriendly and renewable source of plant nutrients to supplement chemical fertilizers and organic manures in sustainable agricultural system in India. They are microbial inoculants which enhance crop production through improving the nutrient supplies and their availability (Wani and Lee, 2002). The use of biofertilizers undoubtedly boosted not only the food production but also, it shows the positive effects on physico-chemical properties of soil, nitrogen transformation, macro and micronutrient uptake and nutritional composition (Mahesh and Hosmani, 2004). Among biofertilizers, *Azotobactor* and *phosphobacterium* is recommended for grain legumes and other crop plants to improve productivity and argument the soil nitrogen status. A "good" strain of *Rhizobium Azotobactor* and *phosphobacterium* iscapable of forming effective nitrogen fixing nodules in the legumes. These *rhizobia* should be superior in their ability to survive in the soil and should have the ability to fix nitrogen symbiotically under diverse agroclimaticconditions (Brahmaprakash and Hudge, 2002).

Pot culture experiment – morphometrical parameters: Pot culture experiment was conducted to find out the effect of organic fertilizers, inorganic fertilizers and biofertilizers on growth, biochemical and enzymatic activities of *Andrographispaniculata* plants. The growth of *Andrographispaniculata* grown under various fertilizers application.

Morphometric analysis of *Andrographispaniculata:* The results on the effect of organic fertilizers, inorganic fertilizers and biofertilizers, in combined inoculation on the shoot length, root length, leaf length, Total leaf area, number of leaver per plant, shoot girth, number of branches per plant and plant height of *Andrographispaniculata* plants at various stages of its growth (30, 60 and 90 days) are shown in Table 1,2,3 and Plate I.The highest shoot lengths, root length, leaf length, Total leaf area, number of leaver per plant, shoot girth, number of branches per plant and plant height (12.3±0.369, 9.6±0.459, 6.2±0.186, 4.1±0.123, 12.7±0.369, 53±1.590, 0.3±0.009, 1±0.003 and 21.9±0.657; 16.8±0.540, 10.3±0.309, 5.2±0.156, 2.5±0.075, 6.5±0.195, 62±1.86, 0.3±0.009, 4±0.12, and 27.1±0.813; 20.0±0.618, 13.2±0.369, 5.3±0.159, 2.6±0.078, 6.9±0.207, 81±2.43, 0.35±0.015, 7±0.21, and 33.8±1.014 cm) were recorded in *Andrographispaniculata* plant grown in single status of biofertilizer application. The lowest shoot length, root length, leaf length, Total leaf area, number of leaver per plant, shoot girth, number of branches perplant andplant height (4.7±0.414,4.4, ±0.132, 3.5±0.105, 1.1±0.033, 1.9±0.057, 6,0±0.180, 0.1±0.003, 9.1±0.273; 5.1±0.150, 3.2±0.096, 4.5±0.135, 1.7±0.051, 3.8, ±0.114, 9±0.27, 0.4±0.012, 8.3±0.249; and 7.3±0.219, 6.3±0.189, 4.7±0.141, 1.9±0.057, 4.5±0.135, 35±0.003, 0.1±0.003, 2±0.060, 13.6±0.408 cm) was recorded at various stages of its growth (30, 60 and 90 days) in the plants grown without any fertilizer application. (Table 1,2,&3). Similar results were recorded earlier in various crops such as potato (Ghosh and Das, 1998), wheat (Mohiuddin *et al.,* 2000) and soybean (Pandher *et al.,* 2003; Padmaja and Lavanya, 2005). Increase in root length and shoot length by biofertilizer inoculation was reported in mint (Kothari *et al.,* 1999) and *Albizialabbek* (Kumudha and Gomathinayagam, 2007). It may be due to the correction of internal hormonal imbalance by exogenous application of suitable biofertilizers for improving better growth (Samra and Dillon, 1993).

Biochemical studies

Photosynthetic pigments of *Andrographispaniculata:* The result on the effect of various fertilizers (organic fertilizers, inorganic fertilizers and biofertilizers) on photosynthetic pigments content of *Andrographispaniculata* at various stages of its growth (30, 60 and 90 days) are shown in Table 4, 5 & 6. The highest chlorophyll a' chlorophyll b, total chlorophyll and carotenoid content (0.758±0.022, 0.599±0.017, 1.357±0.040 and 0.490±0.014 mg/g fr. wt. basis) were recorded in 90 days old crop plants grown with combined application of bio fertilizers. The lowest chlorophyll a, chlorophyll b, total chlorophyll and carotenoid contents (0.480±0.014, 0.292±0.008, 0.772±0.023 and 0.210±0.006 mg/g fr.wt.basis) were recorded in 30 days plant grown without fertilizer application. Similar findings of increased chlorophyll content may be due to fertilizer application was recorded in various plants such as soybean (Hesketh*et al.,* 1985), mulberry (Susheelamma *et al.,* 2002 and 2007), cucumber (Karuppaiah and Kathiravan, 2006), soybean (Thiyageswariand Selvi, 2006) and *Albizialebbek* (Kumudha and Gomathinayagam, 2007) and maize (Tejeda *et al.,* 2008). Carotenoid is an accessory pigment in photosynthetic assimilation of plants. The highest carotenoid content was registered in the crop grown in combined of application biofertilizers followed by inorganic fertilizers and organic fertilizers. The lowest content was recorded in the crop grown without fertilizer. Similar findings of increase in carotenoid content were reported in *Acanthus illicifolius* (Ravikumar*et al.,* 2004) and*Cucumissativus* (Karuppaiah and Kathiravan, 2006). The application of organic manures and inorganic fertilizers applied maize plants showed the highest increase in the carotenoid contents (Tejeda*et al.,* 2008).

Enzymatic activities

Catalase activity of *Andrographispaniculata:* The result on the effect of application of various fertilizers (organic fertilizers, inorganic fertilizers and bio fertilizers) on catalase activity in *Andrographispaniculata* plant at various stages of its growth is shown in table 7. The highest catalase activities (8.100±0.243, 8.420±0.252 and 8.500±0.255 min/mg/protein at 30, 60 and 90 days) were recorded in *Andrographispaniculata* grown with combined application of biofertilizers. The lowest catalase activities (5.493±0.164, 6.982±0.209 and 7.322±0.219 min/mg/protein at 30, 60 and 90 days) were recorded in*Andrographispaniculata* plant grown without any fertilizer application.

Peroxidase activity of *Androgrphispaniculata:* The, result on the effect of application of various fertilizers (organic fertilizers, inorganic fertilizers and bio fertilizers) on peroxidase activity in *Andrographispaniculata* plant at various stages of its growth is shown in table 8.The highest peroxidase activities (14.685±0.440, 15.722±0.071 and 17.382±0.521 min/mg/protein at 30, 60 and 90 days) were recorded in *Androgrphispaniculata* grown with combined application of bio fertilizers. The lowest peroxidayse activities (12.742±0.382, 14.280±0.428 and 15.531±0.465 min/mg/protein at 30, 60 and 90 days) were recorded in*Andrographispaniculata*plant grown without any fertilizer application.

Conclusion

Andrographispaniculata and *Phyllanthusamarus* has been used in Ayurvedha, unani, and Siddha system of medicines from all over the world. The plants were given either in the form of powder extrects are in its isolated compounds are fortified has been used in national and international markets for various diseases. These are safe nontoxic and strong natural antioxidant potential and well known for its medicinal properties and widely used by oriental countries. Biofertilizers are natural and nontoxic beneficial microbes in soil and environments. It is ecofriendly and improves plant growth and productivity of medicinal plants and benefit for our nation. Biofertilizers are the substance that contains microorganism's living or latent cells. Biofertilizers increases the nutrients of host plants when applied to their seeds, plant surface or soil by colonizing the rhizosphere of the plant. Biofertilizers are more cost-effective as compared to chemical fertilizers.

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