

### STUDIES ON THE ROLE OF AZOSPIRILLUM SP., PHOSPHOBACTERIA AND FLY ASH ON THE GROWTH AND YIELD OF BHENDI (ABELMOSCHUS ESCULENTUS L.)

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### Abstarct

A pot culture experiment was conducted at the Department of Microbiology, Faculty of Agriculture, Annamalai University to study the effect of biofertilizers and lignite Fly ash on the growth and yield of Bhendi. The biofertilizers viz., Azospirillum and Phosphobacteria were isolated from different locations of cuddalore district. The isolates were screened for their efficiency in nitrogen fixation, phosphate solubilization, IAA production, etc. Fly ash, a waste generated from coal mines (lignite) was obtained from Neyveli Lignite Corporation (NLC). The major role of Fly ash are it changes the soil <sub>pH</sub> thereby increases water holding capacity of soil and increases both uptake of native and applied nutrients by crop. The biofertilizers Azopirillum, Phosphobacteria and Fly ash were applied to Bhendi crop by framing various treatment schedules to study their performance. Among the different treatments studied, the treatment  $T_{12}(AZS+PB + LFA@40tha^{-1})$  showed maximum plant height (47.10), root length (21.63cm), number of flowers per plant (31.66), number of fruits per plant (29.66) and fruit yield was 92.43 gm per plant. The experiment revealed that application of biofertilizers and Fly ash increases soil fertility which in turn increases the fruit yield of Bhendi.

Key words: Azopirillum, Bhendi, Biofertilizers, Fly ash, Phosphobacteria

### Introduction

India is the second largest producer of vegetables in the world and accounts for about ten percent of world's production. Bhendi (Abelmoschus esculentus) is one major vegetable crop which is grown throughout the year in India. However, bhendi crop produces low yield due to several abiotic and biotic factors. Nowadays, the usage of biofertilizers is increasing among the vegetable crops by replacing the use of chemical fertilizers. The most important macronutrient required for the plant growth is Nitrogen. Among the biofertilizers, Azospirillum sp. is known to fix atmospheric nitrogen in crops and also involves in the production of plant growth promoting substances. Inoculating the Azospirillum sp. to the crops is known to increase the crop yield by 5-20 percent and also involves in saving chemical nitrogen usage upto 50 per cent from the recommended dose of fertilizer (Dart, 1986).

Next to nitrogen, phosphorus is the second essential macronutrient required for crop production which comprises around 0.2 percent of the plant dry weight.

The efficient phosphate solubilizers were used to solubilize fixed phosphorus from the soil their by make the availability of phosphorus to crop (Sharma and Bhalla, 1986). Fly ash is one of waste generated from coal mines (lignite)) and contains several elements which are used for the crop growth. Fly ash application at higher rates around 200 to 800 tons per hectare will improve the physical, chemical and biological properties of soil. In agriculture, Fly ash is used as a nutrient source and can also be used as a suitable liming agent in acidic agricultural soils (Adriano et al., 1980) and to correct nutrient deficiency or increase nutrient uptake of crops (Manoharan, 1996). A pot culture experiment was carried out in the Department of Microbiology, Faculty of Agriculture, Annamalai University to study the effect of biofertilizers Azospirillum and Phosphobacteria and Fly ash on the growth of Bhendi

### Materials and methods

#### Isolation of Azospirillum and Phosphobacteria

The bhendi rhizosphere soil samples were collected from different locations in cuddalore district and the collected soil samples were used for the isolation of *Azospirillum* and *Phosphobacteria*. NFB and Pikovskaya medium were used to isolate Azospirillum and PHOSPHOBACTERIA respectively. The isolates were studied for their characterization and efficiency. The isolates and lignite fly ash were inoculated to the crop with different treatments. The experimental design was CRD. The treatment details are:

Treatment	Treatment details
T <sub>1</sub>	Control
T <sub>2</sub>	AZS
T <sub>3</sub>	PB
T <sub>4</sub>	AZS + PB
T <sub>5</sub>	LFA @ 20 t ha-1
T <sub>6</sub>	LFA @ 40 t ha <sup>-1</sup>
T <sub>7</sub>	AZS + LFA @ 20 t ha-1
T <sub>8</sub>	$AZS + LFA @ 40 t ha^{-1}$
T <sub>9</sub>	$PB + LFA @ 20 t ha^{-1}$
T <sub>10</sub>	$PB + LFA @ 40 t ha^{-1}$
T <sub>11</sub>	$AZS + PB + LFA @ 20 t ha^{-1}$
T <sub>12</sub>	$AZS + PB + LFA @ 40 t ha^{-1}$

AZS - Azospirillum, PB - Phosphobacteria, LFA- Lignite Fly Ash

Nitrogen, phosphorus and potassium were applied @ 40: 50: 30kgha<sup>-1</sup> for all the treatments including control (absolute).

#### **Biometric Observations**

The plant growth parameters such as plant height, root length, number of flowers per plant, number of fruits per plant, fruit yield, dry matter production of the crop and also the nutrient uptake such as NPK and NPK content were observed from these treatments at 30, 60 and 90 DAS.

#### Results

## Effect of *Azospirillum*, *Phosphobacteria* and Fly ash on plant height and root length of Bhendi

In the experimental study, among the all treatments, the maximum plant height and root length was recorded maximum on 90<sup>th</sup> day in the treatment  $T_{12}$  (47.10cm) (21.63cm) followed by the treatment  $T_{11}$  (45.36cm) (19.66cm). The minimum plant height and root length was recorded in the treatment  $T_1$  control (33.20 cm) (15.93cm). The data pertaining to plant height and root length were presented on Table 1.

# Effect of *Azospirillum, Phosphobacteria* and Fly ash on number of flowers plant<sup>-1</sup>, number of fruits plant<sup>-1</sup> and yield of Bhendi.

In the experimental study, among the treatments, the maximum number of flowers per plant, number of

fruits per plant and were recorded on the treatment  $T_{12}$  (31.66) (29.66) and (92.43gm/pt) followed by the treatment  $T_{11}$  (29.33) (27.33) and (89.70gm/pt). The minimum number of flowers plant<sup>-1</sup>, number of fruits plant<sup>-1</sup> and yield on bhendi recorded in the  $T_1$  control (17.33) (14.66) and (80.33gm/pt). These data were presented on the Table 2.

### Effect of *Azospirillum*, *Phosphobacteria* and Fly ash on dry matter production plant<sup>-1</sup> of Bhendi.

In the experimental study, among the treatments, the dry matter production of bhendi was recorded maximum on 90<sup>th</sup> day for the treatment  $T_{12}$  (19.00 gm plant<sup>-1</sup>) followed by the treatment  $T_{11}$  (18.79 gm plant<sup>-1</sup>). The minimum number of flowers plant<sup>-1</sup>, number of fruits plant<sup>-1</sup> and yield on bhendi recorded in the T<sub>1</sub> control (10.75 gm plant<sup>-1</sup>). These data were presented on the Table 3.

## Effect of *Azospirillum, Phosphobacteria* and Fly ash on N, P and K uptake of Bhendi.

The nitrogen, phosphorus and potassium uptake by bhendi are presented in Table 4.

The uptake of nitrogen has been ranged from 39.24 to 47.00 kg ha<sup>-1</sup>. Maximum uptake of N (46.22 kg ha<sup>-1</sup>) was recorded in the treatment  $T_{12}$ -AZS + PB + LFA@40 t ha<sup>-1</sup> which was followed by  $T_{11}$  -AZS + PB + LFA @

	Plant height (cm)			Root length (cm)		
Treatments	Days after sowing			Days after sowing		
	30	60	90	30	60	90
T <sub>1</sub> -Control	12.20	22.63	33.20	9.16	12.36	15.93
T <sub>2</sub> - AZS	13.83	28.30	38.23	10.53	13.23	17.43
$T_3 - PB$	13.43	27.33	37.26	10.16	13.06	17.03
$T_4 - AZS + PB$	14.13	29.26	39.30	11.06	13.53	17.70
$T_5 - LFA@20t ha^{-1}$	12.83	24.66	35.50	9.36	12.56	16.66
$T_6^{-}$ - LFA @ 40 t ha <sup>-1</sup>	13.03	26.53	36.13	9.73	12.83	16.89
$T_7 - AZS + LFA$ @20 t ha <sup>-1</sup>	15.16	32.76	43.26	12.03	14.26	18.33
$T_{8} - AZS + LFA$ @ 40 t ha <sup>-1</sup>	15.60	33.63	44.13	12.43	14.70	18.80
$T_9 - PB + LFA$ @ 20 t ha <sup>-1</sup>	14.33	30.23	40.46	11.16	13.70	17.96
$T_{10} - PB + LFA$ @ 40 t ha <sup>-1</sup>	14.66	31.53	41.33	11.66	14.00	18.00
$T_{11} - AZS + PB + LFA @ 20 ha^{-1}$	16.43	34.26	45.36	12.90	15.26	19.66
$T_{12} - AZS + PB + LFA @ 40 t ha^{-1}$	17.33	36.30	47.10	14.40	17.26	21.63
SED	0.15	0.16	0.18	0.16	0.10	0.15
CD (p=0.05)	0.30	0.32	0.36	0.32	0.21	0.31

Table 1: Plant height and root length

Treatments	No. of flowers plant <sup>-1</sup>	No. of fruits plant <sup>-1</sup>	Yield (gm/pt)
	30 DAS	60 DAS	90 DAS
T <sub>1</sub> – Control	17.00	14.33	80.33
$T_2 - AZS$	22.66	19.66	85.66
T <sub>3</sub> -PB	21.66	18.66	85.78
$T_4$ -AZS + PB	24.00	18.00	86.14
$T_5 - LFA @ 20 t ha^{-1}$	19.33	16.33	83.60
$T_6 - LFA @ 40 t ha^{-1}$	21.00	18.00	83.65
$T_7 - AZS + LFA @ 20 t ha^{-1}$	27.00	25.33	87.06
$T_8 - AZS + LFA @40 t ha^{-1}$	28.66	26.00	87.70
$T_9 - PB + LFA @ 20 t ha^{-1}$	25.66	23.00	86.48
$T_{10} - PB + LFA @40 t ha^{-1}$	26.33	24.33	86.15
$T_{11} - AZS + PB + LFA @ 20 t ha^{-1}$	29.33	27.33	89.70
$T_{12}$ - AZS +PB+ LFA @40 t ha <sup>-1</sup>	32.33	30.66	92.17
SED	0.28	0.29	0.13
CD (p=0.05)	0.56	0.58	0.26

**Table 2:** Azospirillum, Phosphobacteria and Fly ash on number of flowers plant, number of fruits plant and yield of Bhendi.

**Table 3:** Effect of Azospirillum, Phosphobacteria and Fly ash on dry matter production plant<sup>-1</sup>of Bhendi.

Treatments	Dry matter production (gm/pt) Days after sowing			
	30	60	90	
T <sub>1</sub> - Control	1.24	3.16	10.75	
$T_2$ - AZS	1.86	4.15	13.43	
$T_3 - PB$	1.60	3.95	12.54	
$T_4$ -AZS+PB	2.07	4.55	15.38	
$T_5$ - LFA@20t ha <sup>-1</sup>	1.36	3.49	10.32	
$T_6^{-}$ - LFA @ 40 t ha <sup>-1</sup>	1.43	3.72	11.26	
$T_{7}^{-}$ -AZS + LFA@ 20 t ha <sup>-1</sup>	3.20	5.73	17.03	
$T_{8}$ - AZS + LFA @ 40 t ha <sup>-1</sup>	3.80	6.12	17.90	
$T_{9}^{0}$ - PB + LFA @ 20 t ha <sup>-1</sup>	2.50	4.97	16.58	
$T_{10} - PB + LFA @ 40 t ha^{-1}$	2.85	5.42	16.68	
$T_{11}^{10}$ -AZS + PB + LFA	4.62	6.51	18.79	
$@20 ha^{-1}$				
$T_{12}$ -AZS + PB + LFA	5.37	7.84	19.00	
$(20^{12}40 \text{ t ha}^{-1})$				
SED	0.11	0.17	0.10	
CD (p=0.05)	0.23	0.34	0.32	

20 ha<sup>-1</sup> (45.68 kg ha<sup>-1</sup>). Minimum uptake of 'N' was recorded in  $T_1$  control (39.24 kg ha<sup>-1</sup>).

The uptake of phosphorus has been ranged from 9.37 to 15.78 kg ha<sup>-1</sup>. Maximum uptake of 'P' was recorded in the treatment  $T_1$  -AZS + PB + LFA@40 t ha<sup>-1</sup> (14.78 kg ha<sup>-1</sup>) which was followed by  $T_{11}$  -AZS + PB + LFA @ 20 ha<sup>-1</sup> (13.83 kg ha<sup>-1</sup>). Minimum uptake of 'P' was recorded in  $T_1$  control (9.36 kg ha<sup>-1</sup>).

The uptake of potassium has been ranged from 42.15 to 52.63 kg ha<sup>-1</sup>. Maximum uptake of 'K' was recorded in the treatment  $T_{12}$  AZS + PB + LFA@40 t ha<sup>-1</sup> (50.63

 Table 4: Effect of Azospirillum, Phosphobacteria and Fly ash on N, P and K uptake of Bhendi.

Treatments	N uptake (kg ha¹)	P uptake (kg ha <sup>-1</sup> )	K uptake (kg ha¹)
	30	60	90
$T_1$ – Control	39.24	9.36	42.15
$T_2 - AZS$	44.12	10.89	48.85
$T_3 - PB$	42.49	11.99	47.83
$T_4 - AZS + PB$	42.92	11.54	48.37
$T_5 - LFA @ 20 t ha^{-1}$	41.14	10.71	46.84
$T_6 - LFA @ 40 t ha^{-1}$	42.01	10.78	47.17
$T_7 - AZS + LFA @ 20 t ha^{-1}$	44.41	11.00	50.05
$T_{8}^{-}$ - AZS + LFA @40 t ha <sup>-1</sup>	44.70	11.24	50.80
$T_9 - PB + LFA @ 20 t ha^{-1}$	43.31	12.19	49.26
$T_{10} - PB + LFA @ 40 t ha^{-1}$	43.84	12.87	49.54
T <sub>11</sub> - AZS +PB+ LFA	44.62	12.44	40.33
@20 t ha <sup>-1</sup>			
T <sub>12</sub> - AZS +PB+ LFA	46.22	14.78	50.72
@40 t ha <sup>-1</sup>			
SED	0.23	0.23	0.23
CD (p=0.05)	0.47	0.47	0.46

**Table 5:** Effect of *Azospirillum, Phosphobacteria* and Fly ash on N, P and K content of Bhendi.

Treatments	Available N (kg ha <sup>.</sup> 1)	Available P (kg ha <sup>.</sup> 1)	Available K (kg ha <sup>.</sup> 1)
	30	60	90
$T_1 - Control$	92.11	10.62	137.61
$T_2 - AZS$	97.95	12.14	141.52
$\overline{T_3} - PB$	94.20	12.24	146.09
$T_4 - AZS + PB$	96.89	12.78	141.91
$T_5 - LFA @ 20 t ha^{-1}$	93.48	11.96	139.75
$T_6 - LFA @ 40 t ha^{-1}$	93.68	12.03	140.61
$T_{7} - AZS + LFA @ 20 t ha^{-1}$	98.99	12.25	143.01
$T_{8}^{-}$ - AZS + LFA @40 t ha <sup>-1</sup>	100.79	12.48	143.30
$T_9 - PB + LFA @ 20 t ha^{-1}$	94.91	14.21	142.44
$T_{10} - PB + LFA @40 t ha^{-1}$	95.91	14.73	142.72
$T_{11} - AZS + PB + LFA$	101.26	14.86	143.28
@20 t ha <sup>-1</sup>			
$T_{12}$ - AZS +PB+ LFA	103.50	18.80	147.60
@40 t ha <sup>-1</sup>			
SED	0.23	0.05	0.03
CD(p=0.05)	0.47	0.10	0.07

kg ha<sup>-1</sup>) which was followed by  $T_{11}$  –AZS + PB + LFA @ 20 ha<sup>-1</sup> (51.07 kg ha<sup>-1</sup>). Minimum uptake of 'K' was recorded in  $T_1$  control (42.15 kg ha<sup>-1</sup>).

### Effect of *Azospirillum*, *Phosphobacteria* and Fly ash on N, P and K content of Bhendi.

The available nitrogen, phosphorus and potassium in the rhizosphere soil of bhendi are presented in Table 5.

The maximum availability of nitrogen was recorded in the treatment  $T_{12}$  AZS + PB + LFA@40 t ha<sup>-1</sup> (105.50 kg ha<sup>-1</sup>), which was followed by  $T_{11}$  –AZS + PB + LFA @ 20 ha<sup>-1</sup> (103.26 kg ha<sup>-1</sup>) and the minimum availability was recorded in  $T_1$  control (93.11 kg ha<sup>-1</sup>).

The highest available soil phosphorus was recorded in the treatment  $T_1 AZS + PB + LFA@40$  t ha<sup>-1</sup> (17.80 kg ha<sup>-1</sup>) which was followed by  $T_{11} - AZS + PB + LFA$ @ 20 ha<sup>-1</sup> (15.86 kg ha<sup>-1</sup>). The least available soil phosphorus was recorded in  $T_1$  control (11.62 kg ha<sup>-1</sup>).

The maximum availability of soil potassium was recorded in the treatment  $T_{12}$  AZS + PB + LFA@40 t ha<sup>-1</sup> (145.60 kg ha<sup>-1</sup>) which was followed by  $T_{11}$  -AZS + PB + LFA @ 20 ha<sup>-1</sup> (144.28 kg ha<sup>-1</sup>). Minimum uptake of 'K' was recorded in  $T_1$  control (139.61 kg ha<sup>-1</sup>).

### Discussion

In the present study, interesting results were obtained when an attempt was made to study the effect of Azospirillum, phosphobacteria and Fly ash for the growth and development on Bhendi. Twelve treatments were fixed to study the growth parameters of Bhendi. In the present investigation, plant height and root length were maximum in the treatment  $T_{12}$  (AZS + PB+ LFA @40 t ha<sup>-1</sup>). Growth parameters increased due to Azospirillum, phosphobacterial inoculation as they fix atmospheric nitrogen, solubilize phosphorus and production of growth promoting substances which increased the growth parameters. Fly ash proves to be a better soil conditioner and can also be used as a cheapest source of micronutrient for crops and thereby increase the plant growth. The present observation was supported by various workers (Anajali Deshmukh et al., 1999; Amirthaling am, 1988; Deka et al., 1992; Parvathamet al., 1989; Subbiah, 1991; Bashan and Hvanony, 1990 and Naveret al., 1985).

The inoculants of *Azospirillum*, phosphobacteria were used invariably for almost all the crops to boost the yield. In this investigations, higher number of floweres plant<sup>-1</sup>, higher number of fruit plant<sup>-1</sup>, and yield plant<sup>-1</sup> were recorded in the pot treatment  $T_{12}$  (AZS + PB+ LFA @ 40t ha<sup>-1</sup>). It is well established fact that the enhanced microbial activity with the biofertilizer application and LFA would have contributed to increase the nutrient availability, which resulted in increased yield of crop (Matte and Kene, 1995 and Savitha, 1998).

In the present study, interesting results were obtained in the dry matter production of Bhendi. Maximum amounts of dry matter production was observed in the treatment AZS + PB+ LFA @ 40t ha<sup>-1</sup>. The increased dry matter production was due to the application of *Azospirillum*. It is well established fact that *Azospirillum* gives nitrogen, solubilizing the insoluble phosphates, thereby plants absorb higher amount of N,P,K and adds possible amounts of micro and macro nutrients which results in the higher dry matter production.

In the present study, the uptake of N, P and K were maximum in the treatment AZS + PB+ LFA@ 40t ha<sup>-1</sup>.

In the pot culture experiment, the maximum available soil, N, P and K were observed in the treatment receiving AZS + PB+ LFA@ 40t ha<sup>-1</sup>. The available N, P and K contents were increased with fly ash application (Anjali Deshmukh *et al.*, 2000, Savitha, 1998).

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