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# BIO SEED TREATMENT IN SUSTAINABLE AGRICULTURE BY INDUCING MOISTURE STRESS TOLERANCE IN RICE SEED GERMINATION

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## KEYWORDS

Biopriming  
Moisture stress  
*Pseudomonas fluorescens*

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## ABSTRACT

A laboratory experiment was conducted in the Department of Seed Science and Technology, TNAU, Coimbatore during 2013-2014 to analyse the impact of bio priming seed treatment on drought resistance. In this experiment, addition of beneficial microorganism in rice seed priming treatment, induced the drought tolerance in seed germination and established the vigorous seedlings. Seeds of rice hybrid CORH 4 and its parental lines viz., COMS 23A (male sterile), CB 174R (male) were bioprimered with 4 % *Pseudomonas fluorescens*, 20% liquid *Azospirillum*, 15% liquid phosphobacteria and 20 % liquid Azophos, for 12 h duration, separately. Hydroprimed and nonprimed seeds act as control. Among the different bioagents, 4% *Pseudomonas fluorescens* for 12h was found to be the best seedbioprimer treatment which substantially improved the speed of germination, germinationpercentage, seedling length, dry matter production, vigour index over control, under moisture stress condition. This treatment improved 11% of CORH 4 seed germination under extreme moisture stress (20% Water Holding Capacity) condition over unprimed control.

## INTRODUCTION

Indian agriculture mainly depends on monsoon rainfall. Generally usage of beneficial microorganism like bio control agents and bio fertilizers effectively reduces the chemical fertilizers and pesticide in rain fed agriculture (Shaguntala *et al.*, 2012). Moisture stress is a critical environmental factor that restricts seed germination. Moisture stress during the earlier phase of seed germination affects the field emergence, seedling establishment ultimately the yield of the crop (Ceasay, 2004).

Seed priming is an efficient method for increasing seed vigour and synchronization of germination, as well as the growth of seedlings of many crops under stressful conditions and it play a positive role in the tolerance to abiotic stresses. Priming is seed invigoration treatments, which consist of a controlled imbibitions of the seeds followed by dehydration back to their initial water content (Bradford, 1986). This treatment could enable the crop to establish under initial moisture stress condition and established vigorous seedling also give higher yield under terminal moisture stress condition (Musa *et al.*, 2001).

In addition to that many beneficial bioagents also involved in sustainable agriculture under abiotic stress condition. So this experiment was conducted for analyse the combined effect of priming and bioagent as bioprimer seed treatment and to identify the perfect bioprimer treatment which enhance the germination of CORH 4 rice hybrid under moisture stress condition.

## MATERIALS AND METHODS

### Bio priming

The bioagents viz., 4 % *Pseudomonas fluorescens*, 20% liquid *Azospirillum*, 15% liquid phosphobacteria and 20 % liquid Azophos were prepared and the seeds were soaked in equal volume of solutions for 12h durations. After bioprimering, the seeds were removed from the solutions and shade dried at room temperature till bring back the seed moisture to its original level.

### Moisture Stress

A known quantity (weight basis) of sand was taken in an aluminium tray. Then, known quantity of water was added till the media reached saturation and weighed again. The difference in weight and the total quantity of water added to reach saturation was noted. This ratio of sand and water was taken as 100 % moisture holding capacity and accordingly, 80, 60, 40 and 20 % of water was added to sand media to create 80, 60, 40 and 20 % water holding capacities, respectively. Four replicates of 100 seeds from each treatment were sown in each of the above water holding capacity and kept in the germination room and at the end of 14 days, the following observations were made.

### Germination

The germination test was conducted by following the procedure outlined in ISTA (2011) using paper medium by following between paper method. Four replicates

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of 100 seeds each were germinated in a germination room maintained at  $25 \pm 2$  p C temperature and  $90 \pm 3$  % RH. At the end of fourteenth day of sowing, the number of normal seedlings in each replication was counted and the germination was calculated and expressed in percentage.

### Root length

At the time of germination count, ten normal seedlings were selected at random from each replication and used for measuring the root length of seedlings. Root length was measured from the point of attachment of seed to the tip of primary root. The mean values were calculated and expressed in centimetre.

### Shoot length

The seedlings used for measuring root length were also used for measuring shoot length. The shoot length was measured from the point of attachment of seed to tip of the terminal leaf and the mean values were expressed in centimetre.

### Vigour index

Vigour index was computed using the following formula and the mean values were expressed in whole number (Abdul-Baki and Anderson, 1973).

$$\text{Vigour index} = \text{Germination (\%)} \times \text{Seedling length (cm)}$$

## RESULTS AND DISCUSSION

The performance of bioprimered seed under different water holding capacities namely 20, 40, 60 and 80 per cent was compared with nonprimed seeds. The results revealed that the seeds bioprimered with 4 per cent *Pseudomonas fluorescens* for 12 h enhanced the germination under moisture stress conditions than other bioprimering treatments (figure 1). The increase in germination over nonprimed seeds was 11, 10, 7 and 6 per cent in COMS 23A, 10, 8, 8 and 7 per cent in CB 174R and 11, 13, 7 and 7 per cent in CORH 4 under 20, 40, 60 and 80 per cent water holding capacities, respectively. All bioprimered seeds exhibited improved germination rate (figure 1), seedling length (table 1 & 2) and vigour index (plate 1 & figure 2) over nonprimed and hydroprimed seeds under different moisture stress condition.

This result agrees with Chavanet *al.* (2014) who reported that the primed soybean seed produced taller seedling than nonprimed seed. Seed priming has improved seed germination and seedling establishment under extreme drought conditions.

**Table 1: Effect of seed bioprimering on root length (cm) under moisture stress conditions in rice hybrid CORH 4 and its parental lines COMS 23A and CB 174R.**

WHC Bioprimering treatments (T)	COMS 23A					CB 174R					CORH 4							
	20 %	40%	60%	80%	Mean	20 %	40%	60%	80%	Mean	20 %	40%	60%	80%	Mean			
T <sub>0</sub>	16.2	17.4	18.0	18.3	17.5	15.0	16.0	16.5	17.0	16.1	16.4	17.0	17.8	18.2	17.4			
T <sub>1</sub>	17.0	17.8	18.2	18.5	17.9	16.4	16.5	17.2	17.4	16.9	16.6	17.2	18.0	18.5	17.6			
T <sub>2</sub>	18.5	19.0	19.5	19.8	19.2	18.0	18.2	18.5	18.5	18.3	18.4	18.4	18.8	19.0	18.7			
T <sub>3</sub>	17.5	18.0	18.6	19.0	18.3	16.4	17.2	17.5	17.9	17.3	17.2	17.4	18.3	18.5	17.9			
T <sub>4</sub>	17.6	18.3	18.8	19.0	18.4	17.1	17.4	18.0	18.2	17.7	17.3	17.6	18.5	18.8	18.0			
T <sub>5</sub>	17.8	18.0	18.3	19.3	18.4	16.8	17.5	17.9	18.0	17.6	17.0	17.5	18.4	18.6	17.9			
Mean	17.4	18.1	18.6	19.0	18.3	16.6	17.1	17.6	17.8	17.3	17.2	17.5	18.3	18.6	17.9			
	T		W		T x W		T		W		T x W		T		W		T x W	
SEd	0.113		0.093		0.227		0.119		0.097		0.238		0.125		0.102		0.250	
CD (P=0.05)	0.226		0.185		NS		0.237		0.193		NS		0.249		0.203		NS	

Treatment (T) details:

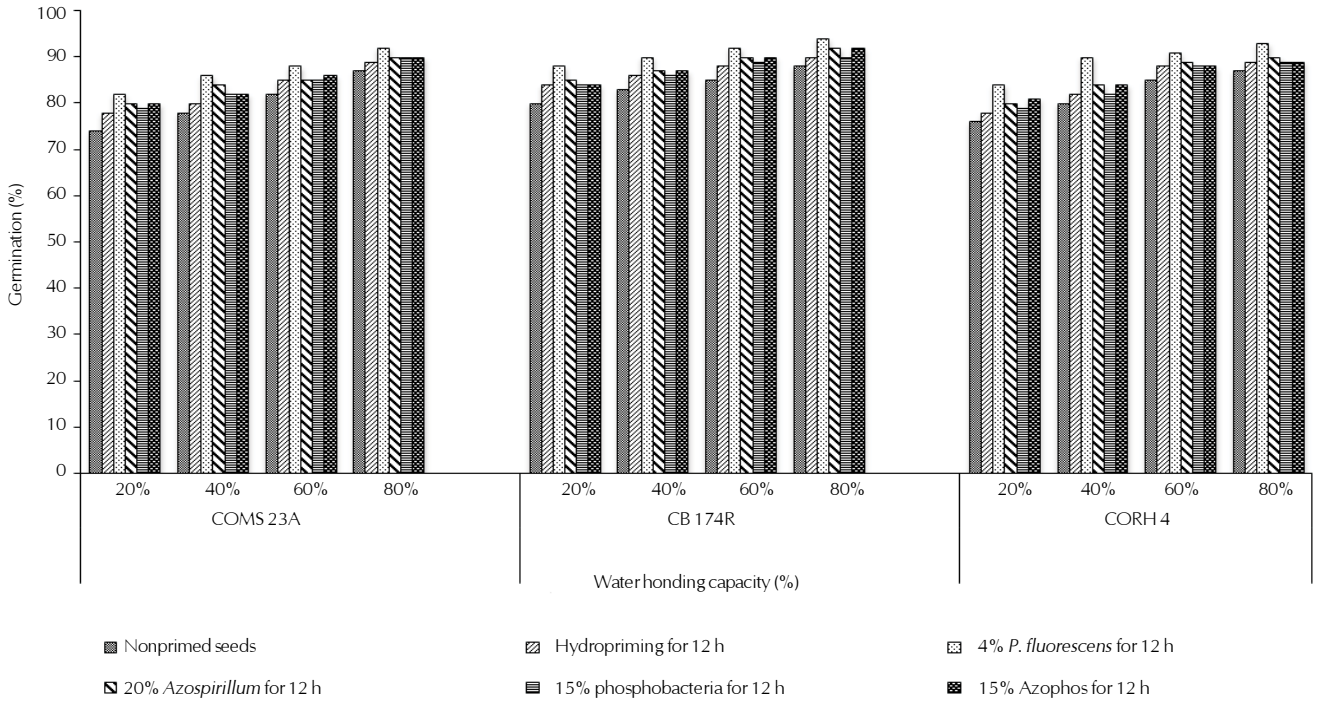
T<sub>0</sub>-Nonprimed seed; T<sub>1</sub>-Hydropriming for 12 h; T<sub>2</sub>-Bioprimering with 4 % *P. fluorescens* for 12 h; T<sub>3</sub>-Bioprimering with 20 % *Azospirillum* for 12 h; T<sub>4</sub>-Bioprimering with 15 % phosphobacteria for 12 h; T<sub>5</sub>-Bioprimering with 15 % Azophos for 12 h

**Table 2: Effect of seed bioprimering on shoot length (cm) under moisture stress conditions in rice hybrid CORH 4 and its parental lines COMS 23A and CB 174R**

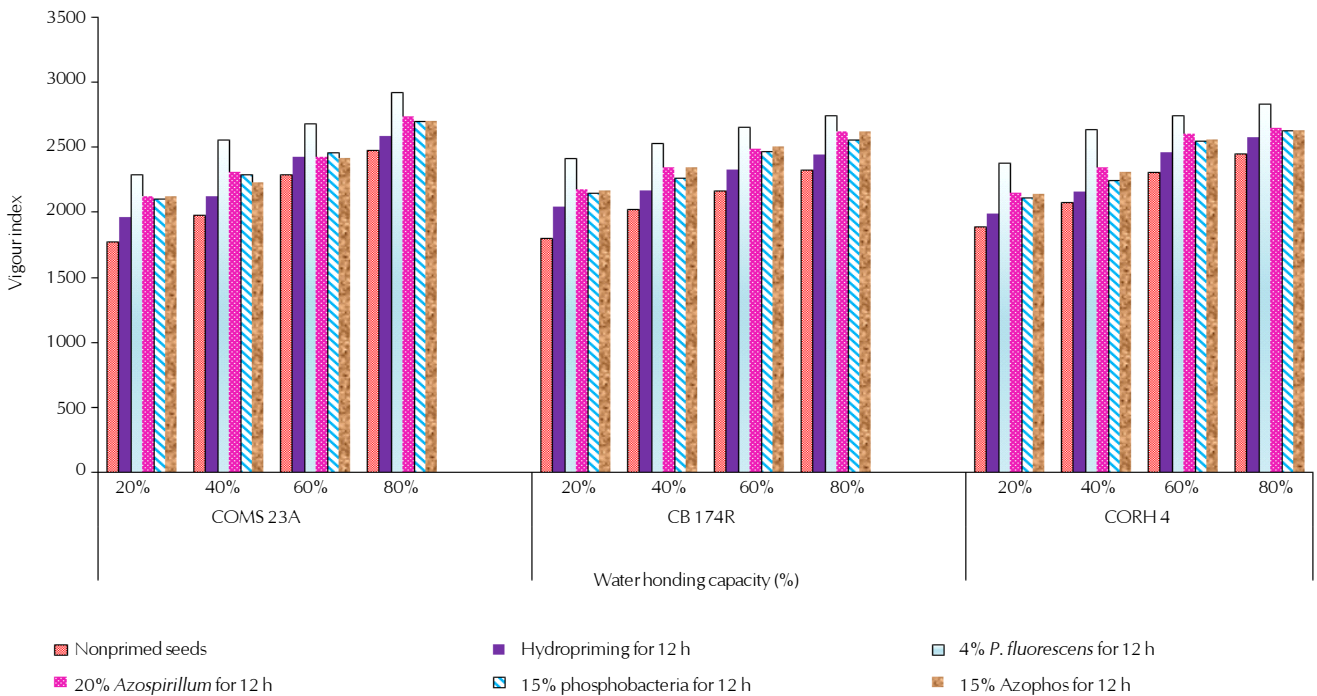
WHC Bioprimering treatments (T)	COMS 23A					CB 174R					CORH 4							
	20 %	40%	60%	80%	Mean	20 %	40%	60%	80%	Mean	20 %	40%	60%	80%	Mean			
T <sub>0</sub>	7.8	8.0	10.0	10.2	9.0	7.5	8.4	9.0	9.5	8.6	8.5	9.0	9.4	10.0	9.2			
T <sub>1</sub>	8.2	8.8	10.4	10.6	9.5	8.0	8.8	9.3	9.8	9.0	9.0	9.2	10.0	10.5	9.7			
T <sub>2</sub>	9.5	10.8	11.0	12.0	10.8	9.5	10	10.4	10.8	10.2	10.0	11.0	11.4	11.5	11.0			
T <sub>3</sub>	9.1	9.6	10.0	11.5	10.0	9.3	9.8	10.2	10.7	10.0	9.7	10.6	11.0	11.0	10.6			
T <sub>4</sub>	9.0	9.7	10.2	11.0	10.0	8.5	9.0	9.8	10.2	9.4	9.5	9.8	10.5	10.8	10.1			
T <sub>5</sub>	8.8	9.2	9.8	10.8	9.6	9.0	9.5	10.0	10.5	9.8	9.5	10.0	10.7	11.0	10.3			
Mean	8.7	9.4	10.2	11.0	9.8	8.6	9.3	9.8	10.3	9.5	9.4	9.9	10.5	10.8	10.1			
	T		W		T x W		T		W		T x W		T		W		T x W	
SEd	0.065		0.053		0.130		0.074		0.061		0.149		0.076		0.062		0.152	
CD (P=0.05)	0.130		0.106		0.259		0.148		0.121		NS		0.152		0.124		0.304	

Treatment (T) details:

T<sub>0</sub>-Nonprimed seed; T<sub>1</sub>-Hydropriming for 12 h; T<sub>2</sub>-Bioprimering with 4 % *P. fluorescens* for 12 h; T<sub>3</sub>-Bioprimering with 20 % *Azospirillum* for 12 h; T<sub>4</sub>-Bioprimering with 15 % phosphobacteria for 12 h; T<sub>5</sub>-Bioprimering with 15 % Azophos for 12 h



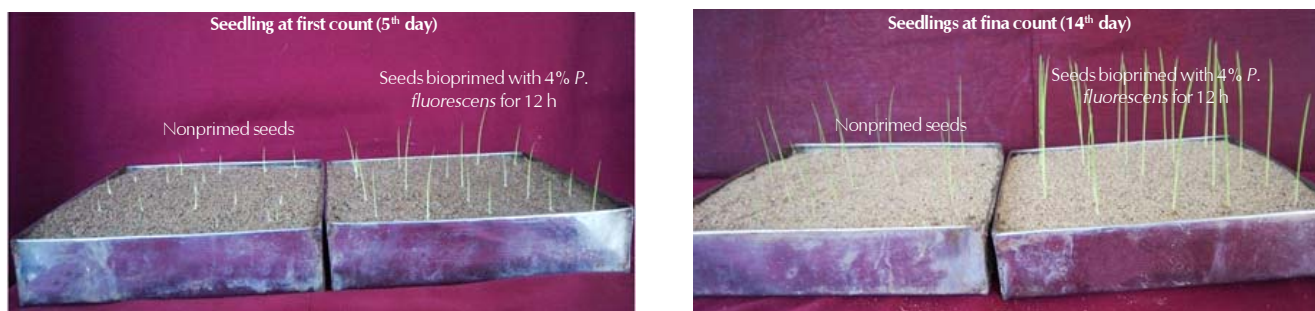
**Figure 1: Germination (%) under moisture stress conditions in bioprime seeds of rice hybrid CORH 4 and its parental lines COMS 23A and CB 174R**



**Figure 2: Vigour index under moisture stress conditions in bioprime seeds of rice hybrid CORH 4 and its parental lines COMS 23A and CB 174R**

The positive role of priming on abiotic stress tolerance might be due to the improved antioxidant production (Mittler, 2002). Antioxidants are natural defensive elements in seed that scavenge excessive reactive oxygen species (ROS). In priming

the excessive ROS were scavenged during early imbibition process and it play an essential role in ensuring successful germination, especially under stress conditions (Bailliet *al.*, 2008).



**Plate 1: Seedling growth of bioprimered CORH 4 rice seed under water stress of 20% water holding capacity**

With regards to PGPR strains, application of PGPR can enhance phytohormones content of seed under moisture stress condition (Ansari *et al.*, 2012). Phytohormones play a critical role in regulating plant growth and its response to stress. *Pseudomonas fluorescens* treatment had improved phytohormonal characters under water deficit condition. The *P. fluorescens* protect plants from drought stress (Loon, 1998) and significantly promote the seedling growth under stress condition. Application of *Pseudomonas sp.* under water stress improved the antioxidant (Heidari and Golpayegani, 2012). This experiment concluded that rice seed bioprimered with 4% *Pseudomonas fluorescens* for 12h could be able to withstand extreme moisture stress (20 % WHC) condition. The tolerance nature of bioprimered seeds might be due to enhanced antioxidant and phytohormone accumulation during bioprimering.

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