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The effects of organic fortification with pulse sprout extracts from horse gram and cowpea on the seedling quality characteristics of rice variety Co. 43

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Rice is the staple food for over half of the Indian population ways and means to fortify seeds organically for better seed vigour has become important and emphasized. Seed fortification is one of the important seed invigouration treatment. The study was conducted to determine the effects of seed fortification with pulse sprout extract on seedling quality characteristics of rice seeds. Laboratory experiment was conducted with the treatments which include fortification of rice seeds with water soaking, 1, 2, 3, 4 and 5% of horse gram sprout extract and cowpea sprout extract. Seed fortification was done in seeds with 12 ± 1% moisture content by soaking for 12 h. Later the soaked seeds were dried in shade for one day followed by sun drying to reach the original moisture content. The seedling quality characteristics were radical protrusion (%), days to maximum germination (days), speed of germination, germination (%), shoot length (cm), root length (cm), vigour index and dry matter production (g seedlings^{-10}). Among all the treatments used, 2% horse gram sprout extract enhanced comparatively better seedling quality characteristics. Hence, seed fortification with 2% horse gram sprout extract could be recommended for rice as a pre-sowing seed invigorative treatment.

Key words: Rice seeds, germination, vigour index, pulse sprout extract, horse gram, cowpea.

INTRODUCTION

Rice is the major staple food for more than two billion people in Asia and one third of the calorific intake of nearly one billion people of Africa and Latin America. The uninterrupted and disproportionate use of chemical fertilizers over a longer period of time has resulted in deterioration of soil quality and reduced yield. To maintain long – term food production there is a need for sustainable agricultural practices. This is one of the aims of organic farming and consumers are prepared to pay higher prices for certified organic products. In many developing countries agriculture is still largely based on low inputs, because farmers cannot afford the high costs of chemical fertilizers and pesticides. For such farmers, organic farming can provide a better economic alternative because the advantages are two fold; (i) the inputs are of lesser cost and (ii) the produces fetch higher price. Sprouting improves nutritional quality of seeds reported by Deshpande et al. (2002). Germination unfolds, and enzymes trigger elaborate biochemical changes. Proteins break into amino acids. Water-soluble vitamins such as B complex and vitamin C are created. Fats and carbohydrates are converted into simple sugars. Weight

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increases as the seed absorbs water and minerals (Vidal-Valverde et al., 2002). Kareem et al. (1989) reported thatseeds of rice Cv.IR 36 and IR 42 treated with 2.5% neem kernel extract gave more vigourous seedlings than untreated seeds. Joseph and Nair (1989) concluded that seed hardening in rice with ten percent cowdung solution registered its superiority in early germination, root and shoot growth and vigour index. Hence it was hypothesized that application of the nutrient extract from the sprouted pulses in the form of seed fortification will enable better crop growth and productivity of rice. Horse gram and cowpea are the low cost and easily available pulses in Tamil Nadu. Seed fortification treatment mainly supplies nutrient to the seed to germinate into vigourous seedlings. The objective of this research wok was to determine the effects of seed fortification with pulse sprout extract on seed germination and seedling quality characteristics in rice seeds.

MATERIALS AND METHODS

The study was conducted on rice variety CO 43. Seeds of rice obtained from Paddy Breeding Station, Tamil Nadu Agricultural University, Coimbatore was used for the study. The seeds were manually cleaned before conducting the studies. The Experiment was conducted at Department of Seed Science and Technology, Tamil Nadu Agricultural University, Coimbatore. Horse gram and cowpea pulse sprout extract were used for seed fortification. Horse gram and cowpea seeds (50 g each) were soaked overnight (16 h) at room temperature and incubated in a wet cloth for 12 h to enable sprouting. 100 g of sprouts were obtained from 50 g of dry seeds. Later, 100 g of sprouts of horse gram and cowpea were separately ground in a mixer-grinder by using 100 ml of ice water refrigerated at 5°C. The ground substance was squeezed through cloth bag and 100 ml extract of 100% concentration was obtained.

The rice seeds variety Co 43 were soaked in water and also varied concentrations such as 1, 2, 3, 4 and 5% of both horse gram and cowpea sprout extracts for 12 h. Later the soaked seeds were dried in shade for one day followed by sun drying to reach the original moisture content, 12 ± 1. Untreated seeds were maintained as control. Fortified seeds were subjected to germination test with four replicates of 100 seeds in between paper towels. The test conditions were 25 $\pm$ 2°C and 95 $\pm$ 5% RH, illuminated with fluorescent light (750 – 1250 Lux). The seeds were checked daily upto 14 days for protrusion of radicle. The number of normal seedlings were counted after 14 days and expressed as germination percentage. Observation were also made on radical protrusion (%), days to maximum germination (days) (Mauromicale and Cavallaro, 1995), speed of germination (Maguire, 1962), germination (%) (ISTA,1999), shoot length (cm) (the distance between collar region to the tip of the primary leaf), root length (cm) (the distance between collar region to the tip of the primary root), vigour index (Abdul-Baki and Anderson,1973) and dry matter production (g seedlings^{-10}), for which seedlings were dried in a hot air oven maintained at 85°C for 48 h and cooling in a desiccator for 30 min and afterwards weighing in an electronic digital balance.

All the data collected were subjected to analysis of variance (ANOVA) using the Factorial Completely Randomized Design and means were separated using the least significant differences method (LSD) at 5% level of significance (Panse and Sukatme, 1985) only when a significant "F" test was obtained. Wherever necessary, the percent values were transformed to angular (Arc-sine) values before analysis.

RESULTS AND DISCUSSION

Highly significant variations were observed in the evaluated seedling quality parameters obtained from the experiments (Table 1). The results showed that horse gram 2% pulse sprout extract effected the maximum radical protrusion of 71% whereas the control was 60%. The seeds fortified with horse gram 2% pulse sprout extract required 4 days for maximum germination, while the control seeds required 5.3 days. With respect to the speed of germination, the seed treated with horse gram 2 and 3% pulse sprout extract and 4 and 5% cowpea pulse sprout extract resulted in the maximum speed of germination of 5.8, whereas the control was 5.2. As shown in Table 1, the germination percentage of 80 was the highest and this was with the seeds fortified with 2% horse gram extract, whereas that of the control seeds was 60%. In Table 2, the minimum shoot length of the control seeds was 9.2 cm and this was followed by horse gram 1% extract of 9.3 cm. The horsegram 2% pulse sprout extract induced the maximum shoot length of 11.4 cm. Among the treatments 5% horse gram extract induced the minimum root length 16.9 cm and the control brought about a value of 17.8 cm. The maximum root length obtained with the use of horsegram 2% extract was 19.8 cm. The control seeds yielded the minimum dry matter production of 0.12 g seedlings^{-10} whereas the maximum dry matter production obtained by the use of 2 and 3% horsegram pulse sprout extract was 0.19 g seedlings^{-10}. Among the evaluated seedling vigour characters and computed vigour index values of all the concentrations, the seeds fortified with 2% horse gram pulse sprout extract yielded the highest value of 2496 while the control seeds brought about the lowest vigour index of 1620.

In this experiment it was found that the seed fortified with horse gram extract recorded higher values for all the parameters compared to the same concentrations of cowpea extract. This might be because bioactive substances present in horse gram extracts are more optimum for rice seeds than cow pea extracts. Kadam and Salunkhe (1985) reported that horse gram is an excellent source of iron and molybdenum. Pre-sowing seed treatments with nutrients produce physiological effects on seed and thereby improve its emergence and productivity (Natarajan, 2003). Sprouting has been identified as an inexpensive and effective technology for improving the quality of legumes, by enhancing their digestability, increasing the content of amino acids.(Chang and Harrold,1988) and reducing the levels of antinutrients (Vidal valvarde et al., 2002).The results revealed that irrespective of the pulse sprout all the treatments recorded better performance compared to control. This may be due to nutritional quality increased in pulse sprouts which is used for seed fortification. Augustin and Klein (1989) reported that the content of phosphorous, potassium, zinc and copper increased

Table 1. Effect of squeezed extract of sprouted horsegram and cowpea at lower concentration on seed germination and seedling vigour of rice seeds.

Treatment and concentration	Radicle protrusion (%)	Days to maximum germination (Days)	Speed of germination	Germination (%)
Control	60(52.53)[c]	5.3	5.2	60(50.76)[d]
Water	63(50.76)[bc]	4.6	5.5	62(51.94)[cd]
Horse gram 1%	65(53.73)[b]	4.3	5.7	76(60.66)[ab]
Horse gram 2%	71(57.41)[a]	4.0	5.8	80(63.43)[a]
Horse gram 3%	68(55.55)[ab]	4.0	5.8	72(58.05)[b]
Horse gram 4%	65(53.73)[b]	4.0	5.6	68(55.55)[bc]
Horse gram 5%	55(47.87)[d]	4.3	5.2	64(53.13)[c]
Cowpea 1%	60(50.76)[c]	5.3	5.7	62(51.9)[cd]
Cowpea 2%	60(50.76)[c]	5.3	5.7	64(53.13)[c]
Cowpea 3%	62(51.94)[bc]	4.6	5.7	64(53.13)[c]
Cowpea 4%	62(51.94)[bc]	4.3	5.8	72(58.05)[b]
Cowpea 5%	62(51.94)[bc]	4.6	5.8	68(55.55)[bc]
Mean	52(43.56)	4.46	5.62	55 (47.87)
SEd	4.158	0.503	0.217	6.198
C.D.(0.05)				
T	0.64	0.083	0.096	0.69
C	0.72	0.09	0.19	0.77
T × C	1.44	0.18	1.44	1.55

Figures in parenthesis are arcsine values.

Table 2. Effect of squeezed extract of sprouted horsegram and cowpea at lower concentration on seed germination and seedling vigour of rice seeds**.**

Treatments concentrations	Shoot length (cm)	Root length (cm)	Dry matter production (g seedlings^{-10})	Vigour index
Control	9.2	17.8	0.12	1620
Water	10.7	17.3	0.14	1736
Horse gram 1%	9.3	18.5	0.16	2113
Horse gram 2%	11.4	19.8	0.19	2496
Horse gram 3%	11.1	19.0	0.19	2167
Horse gram 4%	10.2	18.0	0.17	1918
Horse gram 5%	9.6	16.9	0.14	1696
Cowpea 1%	9.7	17.1	0.14	1686
Cowpea 2%	10.7	17.5	0.14	1779
Cowpea 3%	10.7	17.1	0.15	1736
Cowpea 4%	11.1	18.0	0.15	1804
Cowpea 5%	10.9	17.6	0.15	1710
Mean	**10.38**	**17.9**	**0.153**	**1918**
SEd	0.757	0.857	0.021	259.752
C.D.(0.05)				
T	0.17	0.31	0.002	31.80
C	0.19	0.34	0.002	35.55
T×C	0.39	0.69	0.005	71.11

significantly as result of germination in various legumes. He also observed that germination of a wide array of legumes significantly improved the thiamine content between 7.2 and 147.7%. Similar results was reported by Martin-Cabrejas et al. (2007) that the remarkable increase in sodium content increases the nutritional qualities of sprouted legumes. Seed fortification with micronutrients such as, manganese sulphate, ferrous sulphate, zinc sulphate, ammonium chloride, ammonium molybdate, potassium chloride, potassium dihydrogen phosphate, magnesium sulphate, borax and ammonium sulphate in three concentrations of0.5, 0.75 and 1.0 percent have been proved to be better than control in terms of speed of germination, germination percent, root length (cm), shoot length (cm), dry matter production (g seedlings^{-10}) as well as vigour index (Marimuthu, 2007). Grzywnowicz-Gazda (1982) reported that soaking of spring barley seeds inborax, manganese sulphate, Ammonium molybdate, zinc sulphate, Ferrous sulphate and magnesium sulphate either individually or in mixture for 24 h increased the trace element concentration in the grain. The seed treatment was also found to increase the germination capacity, growth and dry matter production of barley seedlings when compared with untreated control. Nitrogen containing compound might have stimulated the germination by increasing the seed cytokinin content, occurring naturally in seeds, which interacted with growth inhibitors and enhance the metabolic process, leading to higher germination (Khan, 1980). FaShui et al. (1996) noticed an increase in germination rate, seed vigour, seedling fresh weight, seedling height and root length when maize seeds were soaked in solution of 0.3% $CaCl_2$, 0.1% $ZnSO_4$ either individually or in combination.

Seed fortification with molybdenum as a Sodium molybdate, Zinc sulphate and Manganese sulphate at 0.1, 0.2 and 0.2% respectively, brought maximum seed physiological quality attributes in laboratory (Natesan, 2006). The fortification with zinc sulphate was found to increase the level of vitamins, biotins and thiamins and its coenzymes (Srimathi and Malarkodi, 2000). The overall performance of the treatments underscore that manganese sulphate (1.0%), ferrous sulphate (0.75%) and ammonium molybdate (0.75%) can well serve as seed fortification agent to increase the seed vigour of rice seeds. During seed fortification, the first phase of germination ends with completion of imbibition process and hence the time taken from sowing to emergence is much reduced (Hegarty, 1970). Fortification of seed increased the field germination of corn by promoting embryo growth (Zubenko, 1959). The improvement in field emergence due to fortification could also be ascribed to activation of cells, which results in the enhancement of mitochondrial activity leading to the formation of more high energy compounds and vital biomolecules, which are made available during the early phase of germination observed by (Dharmalingam et al., 1988). These initial changes culminate in enlargement of the latent embryo. The probable reason for higher germination in fortified treatment could be due to greater hydration of colloids, higher viscosity and elasticity of protoplasm, increase in bound water content, lower water deficit, more efficient root system (May et al., 1962) and increased metabolic activity (Joseph and Nair, 1989). It might also be due to enhanced metabolic activity resulted in early germination as stated by Kamalam and Nair (1991). Thus it is obvious that the presence of bioacitve substances in sprouted horse gram and cowpea extracts such as, amino acid, vitamins and minerals could have resulted in fortification of rice seeds as corroborated by earlier reports. The seed vigour extended due to fortification of rice seeds with pulse sprout extracts had resulted in better seedling growth as reflected in germination percentage, root length and shoot length. The increase in dry matter production due to the treatments in the present study is in accordance with the report of Periyathambi and Palaniyappan (1981), Selvaraju (1992) in sorghum and Rangasamy et al. (1993) in agricultural crops.

The seeds fortified with horsegram 2% extract recorded the maximum increase over the control to the tune of 9.2, 11, 33, 58 and 54% for radical protrusion percentage, root length (cm), germination percentage, dry matter production (g seedling^{-10}) and vigour index, respectively. It was followed by horsegram 3% extract. With respect to cowpea pulse sprout, the maximum increment of 3, 1, 13, 25, and 11, respectively was recorded in cowpea 4% extract which was followed by 5% extract. Thus the study on seed fortification revealed that with respect to horse gram, 2% horse gram sprout extract was comparatively the most appropriate concentration followed by 3 %.Similarly, among the cowpea extract concentrations, 4% concentration was also the most appropriate followed by 5% concentration.

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