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RESPONSE OF LINSEED (*LINUM USITATISSIMUM* L.) GENOTYPES TOWARDS PLANT GROWTH PROMOTING RHIZOBACTERIA (PGPR) AND PH STRESS

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ABSTRACT

Fourteen linseed genotypes were screened for their response towards germination after treating them with five PGPR (3AAB1, 3AAB7, 3BAB8, RBA6 and RBA8) strains. Out of fourteen genotypes like Guna local (1141), FR-11(970), CI-1538(456), LMH-62(433), A-71(111), Shekhar (C₁) and Parvati (C₂) performed best and selected for pH stress experiment. Inoculation of linseed genotypes which selected strains performed better at all pH stress levels. Overall, the highest germination was recorded in FR-11(970) with *Rhizobium* (RBA6). Five genotypes like Guna local (1141), FR-11(970), CI-1538(456), LMH-62(433) and A-71(111) were selected against check varieties (Shekhar and Parvati) depending on their performance like germination percentage. Among the PGPR culture, RBA6 recorded its best potentiality for increasing germination percentages at both acidic and alkaline level. These findings recorded the utility of use of PGPR as seed inoculation in linseed for the better improvement of their germination and performance in both acidic and alkaline condition.

INTRODUCTION

Linseed (*Linum usitatissimum* L.) (2n = 30) belongs to the family of *Linaceae* is next in importance to rapeseed and Mustard in area as well in production. The seeds contain lignans, a class of phytoestrogens considered to have antioxidant and cancer preventing properties Gill 1967. Soil pH is one of the most severe factors low germination, limiting nodulation, yield and physiological response in crops. Due to this increase in soil pH there is need of Plant Growth Promoting Rhizobacteria (PGPR) as seeds inoculants because they reduce the soil pH and environmental friendly. The use of PGPR is steadily increasing in agriculture and offers on attractive way to replace chemical fertilizers and pesticide. Evidently, PGPR holds enormous prospects in improved and sustainable plant production, including enhanced plant tolerance to stress, better plant nutrient uptake and reduced use of chemical inputs Glick *et al.*, 2007. The beneficial effect of these bacteria has been attributed to their ability to produce various compounds including phytohormones, organic acids and siderophores, fixation of atmospheric nitrogen, phosphate solubilization, antibiotics that suppress deleterious rhizobacteria or some other unidentified mechanisms. The capacity of microorganisms to stimulate germination and improve development of plants has been adapted for *in vitro* and *in vivo* conditions of some agricultural and ornamental plants Ayyadurai *et al.*, 2006, Biswas *et al.*, 2000; Noel *et al.*, 1996 and Tsavkelova *et al.*, 2007). The both abiotic and biotic factors affect the agricultural crops and its yield under stress conditions PGPR's containing ACC deaminase activity are present in various soils and assures improvement of plant growth and development under stress conditions such as heavy metal stress, phyto pathogens, flooding, drought and high salt concentration. Keeping this in view an experiment was conducted to study the response of Linseed (*Linum usitatissimum* L.) genotypes towards plant growth promoting rhizobacteria (PGPR) and pH stress.

MATERIALS AND METHODS

The present study was conducted during 2013-2014 at Biochemistry and Microbiology (PGPR) Laboratory, Department of Biological Sciences, Sam Higginbottom Institute of Agriculture, Technology & Science (Deemed-to-be-University), Allahabad (U.P). Firstly, 14 Linseed genotypes (Fawn colour: FR-11(970), Bharati (304), G-S-40(990), Yellow colour: CI-1538(456), A-71 (111), Bengal -22 (373), A- 76(115), Brown colour: LMH-62 (433), Guna Local (1141) and Check varieties : Shekhar, Padmini, Neelum, Swetha, Parvati} were screened all together to observe the impact of Azotobacter (3AAB1, 3AAB7, 3BAB8) and Rhizobium (RBA6 ,RBA8) bacterial strains inoculation. And out of fourteen genotypes, best selected Linseed genotypes and bacterial strains were further again conducted for pH stress. The experiment was carried out with Completely Randomized Design

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(CRD) one way ANOVA. Secondly, Association of Official Agriculture Chemists (1920), the nutrient agar were prepared by adding 5 gm of NaCl (sodium chloride), 5 gm of peptone, 3 gm of Beef extract, 15 gm of Agar powder, distilled water and maintained pH upto 7.2 - 7.4. The prepared mixture were mixed thoroughly in the conical flasks and autoclaved at 121°C for 15 minutes. The conical flasks containing nutrient agar were cooled down to 45-50 °C and poured into Petri plates. Allow to solidify for at least 30 minutes. After solidification of nutrient agar, take the culture with the help of loop and streak the culture and were incubated at 37°C for 24-48 hours. Thirdly, general screening of 14 Linseed genotypes with PGPR bacterial strains has done by germination test (ISTA, 1999) in the laboratory condition. Germination tests were carried out to determine the effect of inoculation with PGPR bacterial strains on germination percentage. The seeds were inoculated with PGPR bacterial strains by dipping in 9 ml of distilled water + 1 loop culture in the test tubes. Shake it vigorously and leave for 30 minutes. The inoculated seeds were transferred to the containers containing germination paper. Uninoculated controls were used for comparison. The laboratory observations were taken according to the day intervals 3rd day, 5th day, 7th day and 9th day. And fourthly, the water pH was adjusted to different pH levels (pH 5, pH 6, pH7, pH8, pH9) by adding 1Normal HCl or 1 Molar NaOH in conical flasks and test tubes. The conical flasks and test tubes were autoclaved at 121°C for 15 minutes. After cooling down of test tube, the seeds were inoculated with bacterial strains by dipping in 9 ml of distilled water at different pH levels pH5, pH6, pH 7, pH 8 and pH 9 + 1 loop culture in the test tubes. The inoculated seeds were transferred to the containers containing germination paper for germination test to observe the response of different PGPR bacterial strains with linseed genotypes under pH stress. The

containers were kept at room temperature 20°C to 22°C for 24 hours. The observations were taken according to the day intervals 3rd day, 5th day, 7th day and 9th day. Uninoculated controls were used for comparison. Simultaneously, inoculated treatments and uninoculated controls under different pH stress (pH5, pH6, pH8, and pH9) were also used for the comparison towards the neutral pH7. The best performance of Linseed genotypes with bacterial culture strain that perform well under different pH stress were observed and identified.

RESULTS AND DISCUSSION

Effect of PGPR on seed germination of linseed genotypes under acidic and alkaline stress

The seed germination percentages under pH5, pH6, pH8 and pH9 ranged from 72 to 96 %, 40 to 96 % , 60 to 96 % and 32 to 96 % with mean value of 90.66, 88.44, 88.88 and 68, respectively. FR-11 (970) with RBA6 was founded 96 % highest seed germination at pH5, pH8 and pH9 as compared to over neutral pH7 (88%). Shekhar (check) with RBA8 was founded 96 % highest seed germination at pH6 as compared to over neutral pH7 (92%). The use of PGPR strains significantly affected the seed germination. However, the rate of increased in seed germination varied with PGPR strains. The effect of PGPR on germination percentage of linseeds genotypes under pH stress was statistically significant. PGPR inoculation with linseed genotypes exhibited the potential to pH stress. The bacterial strain RBA6 and 3AAB1 showed good performance and exhibited maximum increased in germination percentage under acidic stress (pH5, pH6) and alkaline stress (pH8, pH9). According to some reports PGPR were found to produce better results. The improvement in seed germination by PGPR was also found in work with wheat and sunflower Shaikat *et al.*, 2006, mungbean, Das and Singh 2015. Saikia and

Table1: Mean performance for the effect of PGPR on seed germination (%) of linseed genotypes under pH stress.

Treatments	Culture	Seed germination (%)				
		pH 5	pH 6	pH 7	pH 8	pH 9
Guna Local (1141)	Control	80	84	92	60	52
Guna Local (1141)	3AAB7	92	96	96	96	84
FR-11 (970)	Control	72	80	76	88	76
FR-11 (970)	RBA6	96	92	88	96	96
FR-11 (970)	RBA8	92	96	96	96	80
Cl- 1538 (456)	Control	88	84	92	84	48
Cl- 1538 (456)	3AAB7	92	96	96	96	96
Cl- 1538 (456)	RBA6	92	92	96	92	84
LMH- 62 (433)	Control	92	80	44	88	72
LMH- 62 (433)	3AAB7	96	96	96	92	44
LMH- 62 (433)	RBA6	96	96	96	96	96
A-71 (111)	Control	88	92	88	72	32
A-71 (111)	RBA8	96	96	96	96	44
Shekhar (C ₁)	Control	88	40	56	84	32
Shekhar (C ₁)	RBA8	92	96	92	92	40
Parvati (C ₂)	Control	88	88	64	84	60
Parvati(C ₂)	3AAB1	96	96	76	96	96
Parvati (C ₂)	3BAB8	96	92	80	92	92
Mean value		90.66	88.44	84.44	88.88	68
Range Maximum		96	96	96	96	96
Minimum		72	40	44	60	32
Coefficient of variation (C.V.)		1.11	1.14	1.18	1.13	1.47
Critical Difference(C.D.) (5 %)		1.66	1.66	1.66	1.66	1.66
Mean value of three replications						

Bezbaruah 1995 reported increased seed germination of *cicer arietinum*, *Phaseolus mungo*, *vigna catjung* and *zea mays*. The possible way to increased in this parameters due to the positive effect of *Azotobacter* and their mechanism such as synthesis of growth regulators such as auxin, increased in enzymes activity such as phosphatase and peroxidase and tolerant to pH stress. It has been observed in this experiment that culture strains which consistently increased germination percentage had uniformity effect on germination percentage of some genotypes and bacterial culture strains. Similar improvement of seed germination parameters by rhizobacteria has been reported in other cereals such as sorghum Raju *et al.*, 1999, Rice Pradhan and Mishra and 2015 and pearl millet Niranjana *et al.*, 2004. These findings may be due to the increased synthesis of hormones like gibberellins, which would have triggered the activity of specific enzymes that promoted early germination, such as amylase, which have brought an increase in availability of starch assimilation. Many PGPR destroy 1-aminocyclopropane-1- carboxylate (ACC), a precursor of the ethylene, via production of the enzyme ACC deaminase, which in turn facilitates plant growth and development by decreasing plant ethylene levels. In addition, several forms of stress are relieved by ACC deaminase producers, such as effects of phytopathogenic bacteria, and resistance to stress from polyaromatic hydrocarbons, from Ca²⁺ and Ni²⁺, and from salt and drought Glick *et al.*, 2007.

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