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Full Length Research Paper

Rhizobia – As ameliorant to soil reaction

Y. P. Dubey

Department of Organic Agriculture, CSK HPKV Palampur H.P. India/Department of Statistics and Mathematics, CSK HPKV Palampur H.P. India. E-mail: ypdubey@rediffmail.com. Tel: 01894-230382.

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Soil reaction (pH) is important property soil for crop productions. Many chemicals as ameliorates are required to make soil productive. In organic agriculture, bio-ameliorates are prerequisite. Rhizobia are acid producers and Bradyrhizobia are alkali producers. Rhizobia and Bradyrhizobia are used as ameliorate to study their impact on soil reaction (pH) in laboratory conditions for 21 days. It was observed that Rhizobia decreased soil pH and Bradyrhizobia increased soil pH during incubation.

Key words: Acid producers, alkali producers, brardyrhizobia, soil pH, rhizobia.

INTRODUCTION

Soil reaction (pH) is a significant problem facing agriculture production and limits legume production (Bordeleau and Prevost, 1994). Leguminous plants depend on symbiotic nitrogen fixation for their growth and nitrogen requirement (Brockwell et al., 1991). Out of total nitrogen requirements about 75-80% of total nitrogen requirement of leguminous crop can be met through symbiotic nitrogen fixation (Verma, 1993). Legumes and their rhizobia exhibits varied response to soil reaction. It limits rhizobium survival and persistence in soil and reduces nodulation. Rhizobia have been classified into two categories, that is, Rhizobia (fast growers) and Bradyrhizobia (slow growers). Rhizobia (fast growers) are acid producers and Bradyrhizobia (slow growers) are alkali producers (Tilak, 1993). But the basis for differences in pH tolerance among strains of rhizobia and Bradyrhizobia is still not clear (Correa and Barneix, 1997). Although, several workers have shown that cytoplasm pH of acid tolerance strains is less strongly affected by external acidity. Tolerance of pH either acidic or alkaline by Rhizobia or Bradyrhizobia is due to their inherent character, that is, acid production or alkali production. This characteristic of rhizobia to produce acid or alkali in medium is well established by several workers, but no work is reported in soils. Rhizobia- plants associations have immense significance in soil amelioration programmes for diverse habitats (Sharma et al., 2005).

In a present era of agriculture, organic agriculture is gaining momentum owing to consciousness of people towards safe food and the use of agricultural chemical in

crop production and its consequence on health of human beings, animals and soil. People want food for them as well as for animals, which should be free from agricultural chemicals and have a synergistic effect with environment. Many more ameliorants are available in market to improve soil physical and chemical conditions, but under organic conditions of farming, alternative use of agricultural chemicals is the need of hours. Nitrogen fixation by has emerged as an important tool to meet out the nitrogen requirement of the crops under organic farming. Rhizobium fixed more than the 50% of total biological nitrogen through symbiotic process. So that the contribution rhizobia under organic management can not be ignored as source to meet out the nitrogen requirement of legume crops and their other beneficial properties may be exploited for other purposes of crop production under organic management. Keeping these things in mind, the present investigation was taken into the study, to find out the effect of Rhizobia (fast grower) and Bradyrhizobia (slow grower) on different soils.

MATERIALS AND METHODS

This experiment was conducted in Soil Microbiology laboratory in the department of Soil Science, with soils having pH 6.0 to 8.90. Soils having pH (6.0- 6.18) were collected from Palampur and Soils having pH more than 7.0 were obtained from Soil Testing Laboratory of division of Soil Science and Agricultural chemistry, IARI Pusa, New Delhi with two types of rhizobia - one is fast growing and another is slow growing rhizobia. All the seven culture are indigenous and isolated from different crops. After authentications these strains were used in this study. Out of seven



Figure 1. Rhizobium leguminosarum phaseoli (French bean).

cultures, six are fast growers known as Rhizobia and one is slow grower known as Bradyrhizobia. These strains are designated an H.P.F.-1, H.P.F.-2, H.P.F.-3 and H.P.F.-4 H.P.P.-1 H.P.L.-1 (Rhizobia fast grower) H.P.M.-1 (Bradyrhizobia slow grower). Since, fast growers are acid producer and slow grower is alkali producers. Observations in the Laboratory experiment was conducted up to 21 days because there was no any impact either increase or on the decrease of soil pH after 21 days so the data was reported only upto 21 days.

Measurement of soil pH

Soil pH was measured in 1:2.5 soil and distilled water ration soil water suspension with the help of potentiometer graduated in milli volt. Inoculated and uninoculated studies were conducted in triplicate for each soil. These samples were shacked on shaker for 30 min. The pH of suspension was recorded and presented in table as initial soil pH. Out of these two sets, one set was inoculated with 2 ml of inoculums having optimum population with Rhizobia (fast grower) and Bradyrhizobia (slow grower), whereas, another set of experiment was having three samples each were kept as control. The optimum population was maintained on the basis of turbidity method. Rhizobia (Fast grower) were used to inoculate in six sets of soil samples having soil pH 6.10 to 8.90 and Bradyrhizobia (slow grower) was used to inoculate only in one set of samples having soil pH 6.10. These set of experiments were conducted up to three weeks (21 days). The pH of suspension of each bottle was recorded at three days interval. Average values of soil pH, after 3 days intervals were plotted and are presented as Figure 1.

RESULTS

Soils pH of inoculated and un-inoculated sample was recorded and data are presented in Table 1 and average value of soil pH is presented as Figures. It is evident from table that inoculation by acid producing (fast growing) Rhizobia decreased soil pH of acidic as well as in alkaline soils as compared to no-inoculated soil. In acidic soils, soils pH, increased by inoculation and in alkaline soils,

soil pH, decreased by inoculation of acid producing Rhizobia (fast grower). Rhizobia act as buffer after establishment in the soil. So they try to optimize the soil pH range. Thus the fast growers (Rhizobia) are acid producer and reduce the soil pH both under acid and alkali conditions, whereas slow growers (Bradyrhizobia) are alkali producers, so the increased soil pH, which is depicted in table. Out of seven set of experiments, six strains of rhizobia were fast grower (acid producers) and one strain was slow grower (alkali producers). Soil having initial pH acidic (6.10), inoculated by slow grower (Bradyrhizobium) increased soils pH (6.10 to 6.38) as compared to un-inoculated (6.10 to 6.15). Whereas, inoculation by fast grower's decreased soils pH as compared to no-inoculation of acidic soils (Table 1). Soils pH increased by inoculation of fast growing Rhizobia from initial soils pH, but, this increase was less in inocu-lated set of experiments as compared to no-inoculation (Figures 5, 6 and 7). Since the soil was submerged during study periods so the soil pH increased during submergence because H ions concentration increased at the top layer during submergence so during this period the soil pH of uninoculated treatment also increased. The maximum variation was reported between 15 to 18 days; therefore maximum time for study was taken for 21 days. It is describable from table-1 that decrease in the soils pH by inoculation was recorded (7.80 to 7.40, 8.20 to 8.06 and 8.90 to 8.40) whereas this reduction in non inoculated sets was from (7.80 to 7.78, 8.20 to 8.16 and 8.90 to 8.82). This reduction in soils pH under inoculated treatment is due to acid production, as rhizobia used to inoculation was fast grower and acid producers

Soils pH increased almost up to 18 days and after that either it were constant or declined except in Figure 6 with inoculation. During incubation, Inoculation with Rhizobia

Table 1. Effect of different types of Rhizobia Inoculation on Soil pH.

S/N	Name of Rhizobia	Type of Rhizobia	Initial soil pH	Soil pH after 21 days incubation	
				No- Inoculation	Inoculation
1	Rhizobium leguminosarum phaseoli (French bean)	Rhizobia (Fast Grower)	6.18 <u>+</u> 0.3	6.50 <u>+</u> 0.2	6.35 <u>+</u> 0.3
2	Rhizobium leguminosarum vicieae (Pea)	Rhizobia (Fast Grower)	6.0 + 0.1	6.46 + 0.1	6.32 + 0.2
3	Rhizobium leguminosarum vicieae (Lentil)	Rhizobia (Fast Grower)	6.10 + 0.4	6.56 + 0.3	6.35 + 0.2
4	Bradyrhizobium miscellany (Black gram)	Bradyrhizobium(Slow Grower)	6.10 + 0.2	6.15 + 0.1	6.38 + 0.3
5	Rhizobium leguminosarum phaseoli (French bean)	Rhizobia (Fast Grower)	7.80 + 0.2	7.78 <mark>+</mark> 0.1	7.40 + 0.2
6	Rhizobium leguminosarum phaseoli (French bean)	Rhizobia (Fast Grower)	8.20 + 0.1	8.16 + 0.2	8.06 + 0.1
7	Rhizobium leguminosarum phaseoli (French bean)	Rhizobia (Fast Grower)	8.90 <mark>+</mark> 0.1	6.82 + 0.2	8.40 + 0.2



Figure 2. Rhizobium leguminosarum vicieae (Pea).



Figure 3. Rhizobium leguminosarum vicieae (Lentil).



Figure 4. Bradyrhzobium miscellany (Black gram).

(fast grower) started to increase the soil pH, but increased in soils pH were more in un–inoculated set as compared to inoculated one in acidic conditions (Figures

1, 2 and 3). Under high soil pH, inoculation with Rhizobia (fast grower), drastically reduced soil pH, but latter on it increased constantly during incubation. In un-inoculated



Figure 5. Rhizobium leguminosarum phaseoli (French bean).



Figure 6. Rhizobium leguminosarum phaseoli (French bean).

sets, soils pH also decreased in comparison to initial soils pH. The quantum of decrease was less in un-inoculated sets as compared to inoculated sets. In both soil pH conditions, either acidic or alkali; the impact of inoculation was much more pronounced as compared to no inoculation.

DISCUSSION

This is plausible that Rhizobia (fast grower) are acid producers. Bradyrhizobia (slow grower) are alkali producers. Acid tolerance in rhizobia involves constitutive mechanisms, such as permeability of the outer



Figure 7. Rhizobium leguminosarum phaseoli (French bean).

membrane, together with adaptive responses including the state of bacterial growth and concomitant changes in protein expression (Correa and Barneix, 1997). Inoculation by Bradyrhizobia (slow grower) increased soil pH under acidic conditions, because Bradyrhizobia (slow grower) are alkali producers so the inculation by slow grower increased soil pH. Increase in soils pH was consistently recorded up to 15 days of incubation. However, in uninoculated set, soils pH increased up to 18 days of incubation, but reduced on 21 days of incubation. Slow-growing Rhizobia such as Bradyrhizobia for soybeans, mungbean and peanut produce little basic compounds. After incubation, the pH will increase. (Example, pH before growing = 6.0, after growing pH 6.1 - 6.2) (Anonymous, 2009). In higher soil pH (more than 7.0), pH decreased during incubation, but this decrease was less in no-inoculated set as compared to inoculated sets. Decrease in the soils pH by inoculation was more as compared to no inoculation. Rhizobia- Plants associations have immense significance in soil amelioration programmes for diverse habitats (Sharma et al., 2004). Straub et al. (2003) also reported that Rhizobia (fast grower), that is, *Rhizobium tropici* CIAT899 are considered as an acid-tolerant Rhizobia.

Conclusion

On the basis 21 days study, it was concluded that Rhizobia (fast grower) decreased soil pH during incubation and Bradyrhizobia increased soil pH during incubation. It is explicated from the figures that trends of decreased in soil pH during incubation was consistent by inoculation with Rhizobia and reverse is by inoculation of Bradyrhzobia.

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