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USE OF BIOFERTILIZERS TO ENHANCE RICE YIELD, NITROGEN UPTAKE AND FERTILIZER-N USE EFFICIENCY IN SALINE SOILS

SIKANDER ALI, NAIMA HAMID, GHULAM RASUL AND KAUSER A. MALIK

National Institute for Biotechnology & Genetic Engineering, Jhang Road, P.O.Box 577, Faisalabad, Pakistan.

Abstract

Azolla, plant growth promoting rhizobacteria (PGPR) and chemical fertilizer alone and in different combinations were used to study the effect of nitrogen biofertilizers on rice biomass, nitrogen uptake and fertilizer-N use efficiency. ^{15}N labelled ammonium sulphate was used to trace its uptake into rice plant and compute fertilizer-N recovery. Maximum rice biomass (straw+grain), its N yield and fertilizer-N recovery were obtained in the treatment where PGPR inoculum with 30 KgN/ha was used followed by treatment where in addition to PGPR and fertilizer-N, *Azolla* cover was also present. Use of biofertilizers alongwith a low input of chemical fertilizer-N soils was useful for increasing rice biomass, N uptake and fertilizer-N recovery in rice grown in saline soils.

Introduction

Nitrogen is the most common limiting nutrient for crop production. Although an increased rate of fertilizer-N application has been advocated to meet the growing demand for food, it is unrealistic to advise the farmers to apply fertilizers they could hardly afford. In addition, its excessive use leads to the environmental pollution. Biofertilizers are much more economical and safer source of plant nutrients. *Azolla-Anabaena* symbiosis is considered to be the most suitable and efficient biofertilizer in flooded conditions of rice fields (Lumpkin & Plucknett, 1982) which can fix nitrogen comparable to legumes. The beneficial effect of diazotrophs/PGPR (Roger *et al.*, 1993) and *Azolla* (Watanabe & Liu, 1992) has been reported to increase rice yield. Since rice is generally grown during reclamation of saline soils, the study was therefore undertaken to observe the effect of combined use of *Azolla* and PGPR on rice yield, nitrogen uptake and fertilizer-N use efficiency in rice ecosystem of saline soils.

Materials and Methods

A marginally saline soil was used (Table 1). Total N was estimated with regular Kjeldahl method, and KCl extractable NH^{+}_{4} and NO^{-}_{3}-N by using MgO-Devarda alloy steam distillation method (Keeny & Nelson, 1982). The soil was filled in 25 cm diameter pots @ 16.5 kg/pot and flooded for a few days before rice transplanting. One-month-old rice seedlings of NIAB-6 were transplanted @ 2 seedlings/hill and 5 hills/pot.

For PGPR-inoculation, roots of rice seedlings were dipped for half an hour in a mixture of bacterial culture of *Azospirillum* (strain K-1 & N-4), *Flavobacterium* (96-57) and *Pseudomonas* (96-51), previously isolated by Malik *et al.*, (1994). For *Azolla* inoculation a mixture of 5 *Azolla* strains viz., *A. pinnata* var. *pinnata* and var. imbricata (local), A. *microphylla*, *A. filiculoides*, *A. caroliniana* and Rong ping (hybrid) was inoculated @ 10.2 g f.wt/pot (200g/m^2) after 7 days of transplanting (DAT). To supply phosphorous to rice and enhance *Azolla* growth 5 Kg P_2O_5/ha (superphosphate) was applied into all pots after 3 weeks of *Azolla* inoculation. ^{15}N labelled ammonium sulphate (5% abundance) was applied in solution form, 3-5 cm below soil surface, 7 DAT to the pots in T6-T12 treatment (Table 2). After *Azolla* inoculation, it was allowed to grow (without mixing into soil) for *Azolla* cover treatments, while half of *Azolla* mat was incorporated into soil and half left as inoculum for *Azolla* incorporated treatments. Two such incorporation at 55 and 91 DAT were made during rice growth. During *Azolla* growth, its biomass and total N were estimated and floodwater pH was measured with a portable pH meter. Rice plants at maturity were harvested, dried at 60°C and biomass recorded. ^{15}N was determined using mass spectrometer (Hauk, 1982).

Results and Discussion

Growth of *Azolla* increased after application of phosphorous and its fresh biomass estimated after 7 weeks of its first inoculation was 31-61 g/pot (600-1250g/m^2) being 3-6 times of the inoculated amount. Higher quantity of N 2.5g/m^2 (25 kg/ha) in *Azolla* mat was found for *Azolla* cover+30 kgN/ha treatment (Fig.1). The use of P fertilizer for enhancing *Azolla* growth has been recommended by different workers (Lumpkin & Plucknett, 1982; Motsara *et al.*, 1995; Watanabe & Liu, 1992), and increase in N content of *Azolla* due to N in the culture medium was reported by Kumarasinghe & Eskew (1993). The floodwater pH varied during day time and it was generally lower for *Azolla* treatments (T2&T3) and nitrogen + Azolla treatment (T7) as compared to nitrogen fertilizer treatments (T6 & T11) and control (Fig.2).

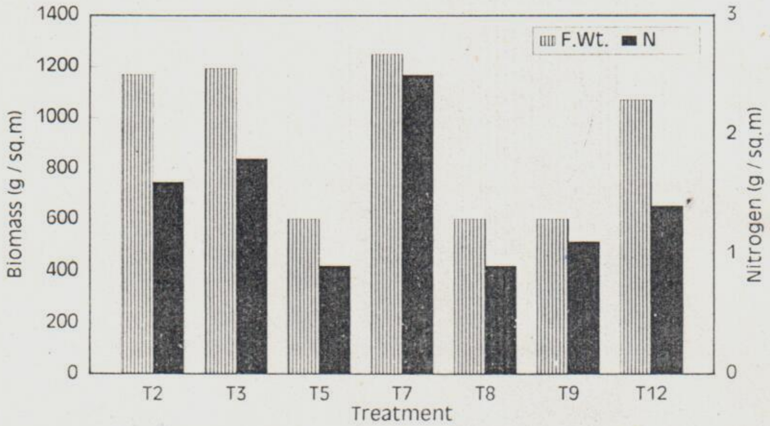

Fig.1.Fresh biomass and nitrogen content of *Azolla* in rice-Azolla culture. (For treatment details, see table 2).

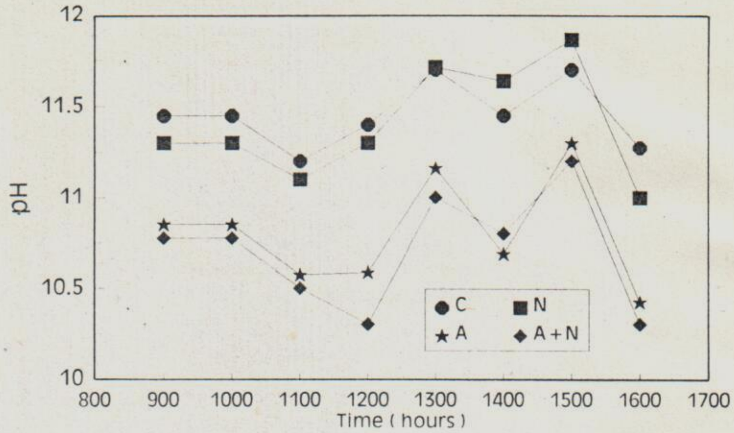

Fig.2.Kinetics of floodwater pH during day time in rice-*Azolla* culture (C = control, N = nitrogen avg. of T6 & T11, A = Azolla avg. of T2 & T3, A+N = T7).

Rice straw and its N yield was higher in treatments where bacteria+30 kgN/ha, N+bacteria+Azolla and bacteria were used indicating the availability of bacterial-N to early stages of rice growth (Table 2). The beneficial effect of PGPR on crop yield has also been reported by Roger *et al.*, (1993). Similarly, the grain and its N yield was also higher for N+bacteria, N+bacteria+*Azolla* cover and *Azolla* incorporated treatments, indicating late availability of *Azolla*-N due to slow decomposition of its lignified tissue (Watanabe *et al.*, 1991). The positive effect of *Azolla* on rice grain yield has also been reported from different countries (Kumarasinghe & Eskew, 1993). The benefit of *Azolla* for increasing rice yield was reported to be positively correlated with number and amount of incorporations (Watanabe, 1987). In the present study very low positive effect of *Azolla* on rice yield may be due to late and only two incorporations made during rice growth. The overall low response to biofertilizers and chemical N fertilizer may be due to the high levels (25 mg/Kg) of available NH^+_4 and NO^-_3-N in this soil (Table 1), as 21 mg N/kg soil, of KCl-extractable $(NH^+_4+NO^-_3)$-N is considered the critical level (Hussain *et al.*, 1994).

Table 1. Chemical properties of the experimental soil.

EC (saturation extract)	4.87 dS/m
pH (soil paste)	7.8
K (saturation extract)	0.15 meq/L
Na (saturation extract)	66.0 meq/L
Ca (saturation extract)	4.1 meq/L
Total N	0.04%
Available NH^+_4-N	11.5 mg/Kg
Available NO^-_3-N	13.4 mg/Kg

Table 2. Effect of biofertilizers on dry matter and total nitrogen yield of rice.

Treatment	Dry matter yield			Total nitrogen yield		
	Straw (g/pot)	Grain (g/pot)	Straw + Grain (g/pot)	Straw (mg/pot)	Grain (mg/pot)	Straw + Grain (mg/pot)
T1. Control	37.4 e	27.9 de	65.4 cd	139 def	278 de	418 def
T2. *Azolla* Cover	36.7 e	25.1 fg	61.8 d	136 ef	252 f	388 fgh
T3. *Azolla* Incorporated	36.2 e	30.7 bc	66.9 c	130 f	293 bcd	423 de
T4. Bacteria	44.6 abcd	20.2 h	64.8 cd	167 ab	210 g	377 gh
T5. Az. Cover-Bacteria	42.6 cd	22.9 g	65.5 cd	148 cde	225 g	374 h
T6. 30 Kg N/ha	44.1 bcd	30.2 bcd	74.2 b	149 cde	284 cde	433 cde
T7. 30 Kg N/ha + Az. Cover	41.5 d	26.5 ef	68.0 c	145 def	261 ef	407 efg
T8. 30 Kg N/ha + Bacteria + Az. Incorp.	47.6 ab	28.7 cde	76.2 b	163 abc	284 cde	447 bcd
T9. 30 Kg N/ha + Bacteria + Az. Cover	46.3 abc	31.2 abc	77.5 b	167 ab	303 bc	470 b
T10. 30 Kg N/ha + Bacteria	48.6 a	33.5 a	82.1 a	175 a	334 a	509 a
T11. 60 Kg N/ha	42.3 cd	26.8 ef	69.1 c	156 bcd	269 def	425 de
T12. 30 Kg N/ha + Bact + Az. Cov + BGA	43.7 bcd	31.9 ab	75.6 b	148 cde	308 b	457 bc

N-15 labelled ammonium sulphate was applied to soil for treatment T6 to T12.

Means followed by the same letter are not statistically different at 5% level.

Abbreviations: Az. = Azolla, Bact = Bacteria, BGA = Blue-green algae, Incorp. = Incorporated.

Table 3. Nitrogen derived from fertilizer, fertilizer-N yield and fertilizer-N recovery in rice.

Treatment		N derived from fertilizer		Fertilizer-N Yield		Fertilizer-N recovery	
		Straw (%)	Grain (%)	Straw (mg/pot)	Grain (mg/pot)	Straw (%)	Grain (%)
T6.	30 Kg N/ha	6.74 d	5.87 c	10.04 d	16.70 d	6.56 c	10.92 d
T7.	30 Kg N/ha + Azolla Cover	5.46 e	4.56 d	7.94 e	11.92 e	5.19 d	7.79 e
T8.	30 Kg N/ha + Bacteria + Az. Incorp.	10.82 a	7.76 a	17.60 a	22.08 bc	11.51 a	14.43 c
T9.	30 Kg N/ha + Bacteria + Az. Cover	10.32 b	8.01 a	17.15 ab	24.21 b	11.21 ab	15.82 b
T10.	30 Kg N/ha + Bacteria	9.06 c	8.29 a	15.88 b	27.67 a	10.38 b	18.09 a
T11.	60 Kg N/ha	8.95 c	8.37 a	13.97 c	22.50 bc	4.56 a	7.35 e
T12.	30 Kg N/ha + Bacteria + Az.Cov + BGA	7.17 d	7.09 b	10.66 d	21.89	6.97 c	14.31 c

N-15 labelled ammonium sulphate was applied to soil for treatment T6 to T12.

Means followed by the same letter are not statistically different at 5% level.

Abbreviations: Az. = Azolla, Bact = Bacteria, BGA = Blue-green algae, Incorp. = Incorporated.

The proportion of ^{15}N labelled fertilizer to the total N in rice straw was higher in treatment where bacteria+*Azolla* were used while in grain it was higher in treatment with bacteria alone or along with *Azolla* and 60 kgN/ha (Table 3). The fertilizer-N recovery, traced with ^{15}N labelled fertilizer, indicated that it was higher in the presence of bacteria or bacteria+*Azolla* (Table 3). A better utilization of fertilizer-N for PGPR inoculation may be due to their growth promoting effect (Arshad & Frankenberger, 1993) on rice roots (Okon & Kapunlnik, 1986) as more root biomass helps in higher uptake of nutrients from soil. *Azolla* has been reported to increase fertilizer-N use efficiency by lowering the floodwater pH thereby reducing ammonia volatilization losses (Ali & Malik, 1988; Kumarasinghe & Eskew, 1993).

The study indicated that use of biofertilizers is useful for increasing rice yield, nitrogen uptake and fertilizer-N use efficiency in rice ecosystem.

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