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Influence of microbial culture in combination with micronutrient in improving the groundnut productivity under alluvial soil of India

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ABSTRACT

A field experiment was carried out to study the effect of cobalt, *Rhizobium* and phosphobacterium inoculations on growth, yield and nutrient uptake of summer groundnut in an alluvial soil at three levels of cobalt viz., zero, 0.21 kg and 0.42 kg ha^{-1} with four levels of inoculations viz., uninoculation, inoculation with *Rhizobium*, inoculation with phosphobacterium and inoculation with both *Rhizobium* and phosphobacterium. Results indicated that combined application of *Rhizobium* and phosphobacterium inoculation promoted higher dry matter production, pod yield, oil content and nutrient uptake as compared to application of either of the inoculations. Cobalt @ 0.21 kg ha^{-1} proved to be better to other doses of cobalt. The percent increase in pod yield of different treatment combinations over untreated control varied from, 2.4% with no inoculation + cobalt @ 0.42 kg ha^{-1} to 35.6% with *Rhizobium* + phosphobacterium + cobalt @ 0.21 kg ha^{-1}. The uptake of N, P and K by groundnut was significantly higher in the treatments receiving both inoculation and cobalt at 0.21 kg ha^{-1} than sole application of either inoculation or cobalt.

Key Words: Groundnut, cobalt, Rhizobium, phosphobacterium, nutrient uptake.

IZVLEČEK

VPLIV MIKROBNE KULTURE IN MIKROHRANIL NA IZBOLJŠANJE PRIDELOVANJA ZEMELJSKIH OREŠKOV NA ALUVIJALNIH TLEH V INDIJI

Poljski poskus je bil izveden, da bi raziskali vpliv kobalta, *Rhizobium*-a in fosfornih bakterij na rast, pridelek in privzem hranil zemeljskih oreškov, pridelanih na aluvijalnih tleh, s tremi nivoji gnojenja s kobaltom (brez, 0,21 kg in 0,42 kg ha^{-1}) in štirimi različnimi inokulacijami (kontrola, inokulacija z *Rhizobium*-om, inokulacija s fosfornimi bakterijami in kombinirana inokulacija z obojim). Rezultati kažejo, da kombinirana inokulacija najbolj vpliva na povečanje pridelka sušine, pridelek strokov, vsebnost olja in privzem hranil. Odmerek gnojenja s 0.21 kg ha^{-1} kobalta je bil optimalen. Privzem N, P in K je bil pri zemeljskih oreških značilno večji pri

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kombinirani inokulaciji ter pri odmerku 0.21 kg ha^{-1} kobalta v primerjavi z drugimi kombinacijami tretiranj.

Ključne besede: zemeljski oreški, kobalt, Rhizobium, fosforne bakterije, privzem hranil

1 INTRODUCTION

Among all the oilseed crops, groundnut (*Arachis hypogaea* L.) has the first place in India accounting for more than 28% acreage and 32% production in the country (Anonymous, 2004). Although the country is steadily progressing in oilseeds production, but the average yields of most oilseeds are still extremely low when compared to those prevailing in other countries of the world. This is because oilseeds are cultivated mostly in marginal and sub-marginal land of semi-arid areas under unirrigated condition, hence remain vulnerable to vagaries of nature. Beneficial effects of *Rhizobium* inoculation has been observed by several workers (Naidu, 2000) who reported an increase in yield and oil content of groundnut with such inoculation. Phosphorus deficiency is probably the major limitation to the growth of legumes in many soils. Phosphobacterium, an organism turning the phosphate present in the soil from unavailable to available form, has an indirect but definite effect on the nodulation and yield of groundnut. The seed and soil inoculations with phosphobacterium inoculants can significantly increased the pod yield of groundnut (Balasubramanian and Palaniappan, 1994) as compared to no inoculation. Along with other micronutrients like Mo, Zn, B, Co also takes an important place for nitrogen fixation in the legumes and in the root nodules of non-legumes. Cobalt application increases the formation of leghaemoglobin required for nitrogen fixation thereby improves the nodule number per plant and ultimately pod yield of groundnut (Yadav and Khanna, 1988). Three specific cobalamine dependent enzyme systems in *Rhizobium* which may account for the influence of cobalt on nodulation and nitrogen fixation are: methionine synthase, ribonucleotide reductase and methylmalonyl co-enzyme A mutase (Das, 2000).

The aim of the present experiment is to evaluate the effect of *Rhizobium,* phosphobacterium and cobalt on yield, oil content and nutrient uptake of groundnut under field condition.

2 MATERIALS AND METHODS

Field experiment was conducted for two years (2001-2002) with groundnut (*Arachis hypogaea* Linn) variety, JL-24 in the pre-kharif season (Feb-June) in the alluvial soil of eastern part of West Bengal, India. The soil characteristics of the experimental site was sandy clay loam with neutral pH (7.2). The organic carbon, available N, P, K and Co content was 0.66%, 109.5 ppm, 22.4 ppm, 85.3 ppm and 0.03 ppm, respectively. The weather of the experimental site was warm and humid with average maximum temperature of 32.7 ± 1.57^0C, minimum temperature of 22.8 ± 2.54^0C, average rainfall of 117.6 ± 96.8 mm, maximum relative humidity of 94.6 ± 3.08% and minimum relative humidity of 63.7 ± 14.6%. The experiment was laid out with two factors, i. e., inoculation with *Rhizobium* (I_R) (strain: M 10), inoculation with phosphobacterium (I_P) (*Bacillus polymyxa*), inoculation with both *Rhizobium* and phosphobacterium and uninoculation (I_0) and three levels of cobalt viz., no cobalt, 0.21 kg and 0.42 kg cobalt ha^{-1} designated as C_0, $C_{.21}$ and $C_{.42}$ respectively. Inoculations were allotted to main plots and levels of cobalt to the sub plots under each main plot. Thus, 12 treatments were arranged in a split plot design with three replications.

2.1 Analysis of soil and plant

Soil samples were collected from the experimental site before and after the experiment to know the fertility status of the soil. Processed samples were analyzed for different physicochemical properties of soil viz., pH, organic carbon, available nitrogen, available phosphorus and available potassium by following standard procedures (Jackson, 1973). Cobalt was estimated by DTPA extraction method (Lindsay and Norvell, 1978).

Haulms of groundnut were dried at 72^{o}C for 48 hours in oven, ground and subsequently used for its chemical analysis. The total N and P was determined by modified Kjeldahl method and vanado-molybdate blue color method respectively (Chapman and Pratt, 1961). K and Co was determined by following the wet digestion method (Jackson, 1973).

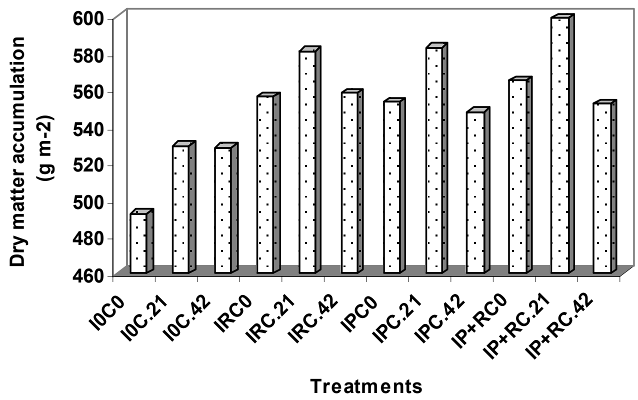
2.2 Statistical analysis

The data were analyzed statistically by applying analysis of variance for split plot design. Least significant differences (LSD) were conducted at a 5% level of probability (Gomez and Gomez, 1976).

3 RESULTS AND DISCUSSION

3.1 Growth parameters

Table 1 and Figure 1 shows that there was a significant influence of different treatments of inoculation and cobalt on the plant height, number of branches per plant and dry matter production at harvest and leaf area index (LAI), number of nodules per plant at 50 days after sowing (DAS) of groundnut. Combined application of *Rhizobium* and phosphobacterium inoculation was better as compared to use of either of the inoculation at a given dose of cobalt. These parameters were also significantly influenced by different levels of cobalt. The highest dry matter production in application of both *Rhizobium* and phosphobacterium was due to the fact that it produced maximum shoot length, higher number of branches per plant and leaf area index (LAI) (Chetti et al., 1995; Naidu, 2000). The maximum values were found with Co @ 0.21 kg ha^{-1} followed by Co @ 0.42 kg ha^{-1}. The minimum values of all the growth parameters observed in case of without cobalt application. Increase in dry matter production due to cobalt application has been reported by Joshi et al. (1987). The combination of *Rhizobium* inoculation, phosphobacterium inoculation and cobalt at lower dose (@ 0.21 kg ha^{-1}) proved superior to any other combinations of inoculation and cobalt. There was no significant difference between *Rhizobium* inoculation and phosphobacterium inoculation.



(Note: I_0=No inoculation, I_R=Inoculation with *Rhizobium* and I_P=Inoculation with Phosphobacterium. C_0, $C_{.21}$, $C_{.42}$ = Cobalt @ zero, 0.21 and 0.42 kg ha^{-1} respectively.)

Fig. 1: Dry matter accumulation by groundnut at harvest as influenced by different levels of inoculations and cobalt (Pooled mean of two years).

3.2 Pod yield, haulm yield and oil percent

There was a significant influence of different treatments of inoculations and cobalt on pod yield, haulm yield and oil percent of groundnut (Table 2). Pod yield in all the treatment combinations were significantly higher than the untreated control. Again pod yield, averaged across three levels of cobalt, was recorded to be highest for inoculation with both *Rhizobium* and phosphobacterium, which was 19.7% and higher over no inoculation. Beneficial role of *Rhizobium* in the N nutrition through nodulation and a consequently better growth or development attributed for this yield advantage

(Subramaniyan and Kalaiselven, 2000). In general, average (across three inoculations) pod yield of groundnut was about 14.5% higher with Co @ 0.21 kg ha^{-1} than without cobalt. However, increasing the level of cobalt to 0.42 kg ha^{-1} did not significantly influence the pod yield. Lower dose of cobalt helps in better nodulation and consequently a better growth and yield, but at a higher level cobalt reduced the bacterial population in the rhizosphere and as a result nodulation was hampered which led to a lower growth and yield of the crop (Jana and Sounda, 1994). Application of both *Rhizobium* and phosphobacterium inoculation along with cobalt at 0.21 kg ha^{-1} showed 35.6% higher pod yield than untreated control. Balamurugan and Gunasekaran (1996) reported that the combined inoculation of *Rhizobium* and phosphobacterium gave maximum crop growth, nodulation and yield in groundnut. Similar trend was followed in case of haulm yield. As in pod yield different levels of inoculations and cobalt significantly influenced oil percent. Average oil percent increased by about 12.4% over without cobalt and enhancing the cobalt level beyond 0.21 kg ha^{-1}, did not significantly increase the oil percent. Combined application of *Rhizobium* inoculation, phosphobacterium inoculation and Co @ 0.21 kg ha^{-1} gave higher values than single application of either inoculation or cobalt. Similar observations were reported by Raj and Rao (1996).

Table 1: Growth parameters of groundnut as influenced by cobalt and inoculations (Pooled mean of 2001-2002)

Treatment combinations	Plant height (cm)	Number of branches plant^{-1}	LAI[1]	[1]Number of nodules plant^{-1}
I_0C_0	45.7	9.5	2.58	146.4
$I_0C_{.21}$	50.8	11.4	3.17	155.0
$I_0C_{.42}$	46.9	9.7	2.95	151.7
I_RC_0	52.8	11.4	3.01	181.3
$I_RC_{.21}$	60.2	14.0	3.70	193.2
$I_RC_{.42}$	55.0	11.5	3.41	189.9
I_PC_0	52.5	10.8	3.01	157.7
$I_PC_{.21}$	58.4	13.4	3.72	172.7
$I_PC_{.42}$	54.1	11.1	3.51	166.3
$I_{P+R}C_0$	55.1	11.8	3.22	190.2
$I_{P+R}C_{.21}$	61.3	14.9	3.85	214.8
$I_{P+R}C_{.42}$	55.2	11.6	3.67	198.5
LSD (P=0.05)				
At same inoculation	4.23	1.23	0.57	7.83
At same cobalt level	1.68	NS	NS	6.01
Inoculation X Cobalt interaction	7.08	3.02	1.07	9.93

(LAI and number of nodules plant^{-1} at 50 DAS; I_0=No inoculation, I_R=Inoculation with *Rhizobium* and I_P=Inoculation with Phosphobacterium. C_0, $C_{.21}$, $C_{.42}$ = Cobalt @ zero, 0.21 and 0.42 kg ha^{-1} respectively. LSD: Least significant difference)

3.3 Nutrient uptake

Levels of inoculation and cobalt significantly influenced the total uptake of N, P, K and Co by groundnut at harvest (Table 3). *Rhizobium* inoculation recorded 5.48% higher N uptake over phosphobacterium inoculation. The beneficial effect of *Rhizobium* was also observed by Saad et al. (1998) and Naidu (2000). Average N uptake (across three cobalt value) was maximum in combined application of *Rhizobium* and phosphobacterium inoculation as compared to single application of either of the inoculations and it was 22.6% and 29.33% higher over *Rhizobium* and phosphobacterium inoculation respectively. Shasidhara and Sreenivasa (1994) obtained the same results. On an average, the average value of N uptake (across three inoculation levels) increased when cobalt was applied @ 0.21 kg ha^{-1} and enhancing the cobalt level to 0.42 kg ha^{-1}, had no positive effect. The maximum N uptake value was obtained with *Rhizobium* + phosphobacterium inoculation + Co @ 0.21 kg ha^{-1} which was followed by *Rhizobium* inoculation + Co @ 0.21 kg ha^{-1} and the difference was 27.4%. The cause of maximum uptake of N in combined application of *Rhizobium* and phosphobacterium inoculation with cobalt @ 0.21 kg ha^{-1} was that, it had increased the population of nitrogen fixing bacteria (*Rhizobium*) in the rhizosphere which led to more infection as well as nodule formation (Yadav and Khanna 1988). Phosphorus uptake also followed the same trend as N. Groundnut inoculated with *Rhizobium* + phosphobacterium culture resulted in 29.4% and 31.6% higher average P uptake over application of either *Rhizobium* inoculation or phosphobacterium inoculation respectively. But there was no significant difference between P uptake at *Rhizobium* and phosphobacterium inoculation. Cobalt applied @ 0.21 kg ha^{-1} significantly increased the average P uptake over without cobalt; the extent of increase was about 21.0%. The maximum value was obtained with

Rhizobium + phosphobacterium + cobalt @ 0.21 kg ha^{-1}. Similar observations regarding the effect of inoculation and cobalt on nutrient uptake were also reported by and Raj and Rao (1996). Uptake of K also followed the similar trend as N and P. The uptake of cobalt was higher with phosphobacterium inoculation than *Rhizobium* inoculation and the difference was 28.2%. Maximum uptake of cobalt was found in use of both the inoculation along with Co @ 0.42 kg ha^{-1} followed by phosphobacterium + cobalt at @ 0.42 kg ha^{-1}.

Table 2: Pod yield, haulm yield, oil content and protein content of groundnut as influenced by cobalt and inoculations (Pooled mean of 2001-2002)

Treatment combinations	**Pod yield (kg ha^{-1})**	**Haulm yield (kg ha^{-1})**	**Oil content (%)**	**Protein content (%)**
I_0C_0	1701.6	2603.6	42.9	20.2
$I_0C_{.21}$	1810.4	3302.3	47.5	28.9
$I_0C_{.42}$	1742.4	3042.6	45.7	27.3
I_RC_0	1888.8	3809.8	45.9	33.2
$I_RC_{.21}$	2158.6	4148.8	48.7	35.1
$I_RC_{.42}$	1954.8	3967.8	47.6	34.6
I_PC_0	1868.9	3759.2	45.8	30.9
$I_PC_{.21}$	2166.1	4125.2	48.8	32.9
$I_PC_{.42}$	1982.0	3973.6	47.0	31.7
$I_{P+R}C_0$	1914.6	3875.2	46.2	32
$I_{P+R}C_{.21}$	2307.3	4305.4	49.8	33.8
$I_{P+R}C_{.42}$	2065.3	4056.2	47.3	32.6
LSD (P=0.05) At same inoculation	109.95	619.77	1.73	5.42
At same cobalt level	70.64	155.60	1.48	2.66
Inoculation X Cobalt interaction	205.86	700.98	2.56	5.59

Note: I_0=No inoculation, I_R=Inoculation with *Rhizobium* and I_P=Inoculation with Phosphobacterium. C_0, $C_{.21}$, $C_{.42}$ = Cobalt @ zero, 0.21 and 0.42 kg ha^{-1} respectively., LSD: Least significant difference.

3.4 Residual soil fertility

The data on changes in nutrient content of soil over two years (Figure 2) revealed that there was a marginal change under the influence of different inoculations and cobalt levels. After two years the nitrogen content of the soil increased from an initial value of 110 ppm to as high as 150 ppm under combined application of *Rhizobium* and phosphobacterium inoculation. The build-up of P was also greater in application of both *Rhizobium* and phosphobacterium with an increment in average phosphorus content of the soil up to 32 ppm. Cobalt application improved the cobalt accumulation in soil. Increasing the dose of cobalt subsequently increased the cobalt content of the soil with a maximum value of 0.38 ppm. Improvement of the cobalt content of the soil was higher in association with phosphobacterium inoculation than in combination with *Rhizobium* inoculation.

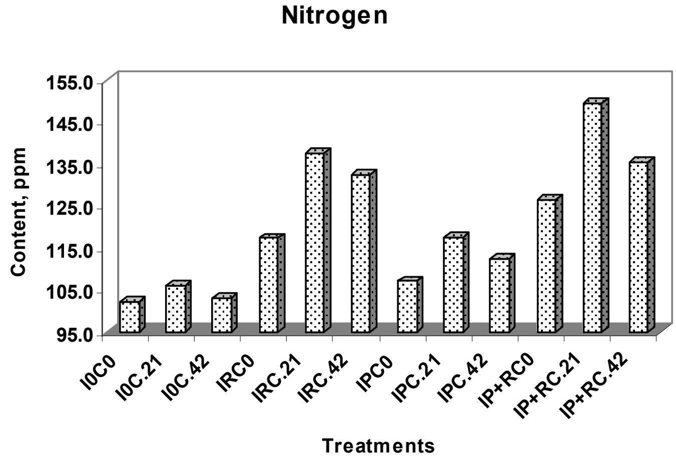
Table 3: Nutrient uptake by groundnut as influenced by cobalt and inoculations (Pooled mean of 2001-2002)

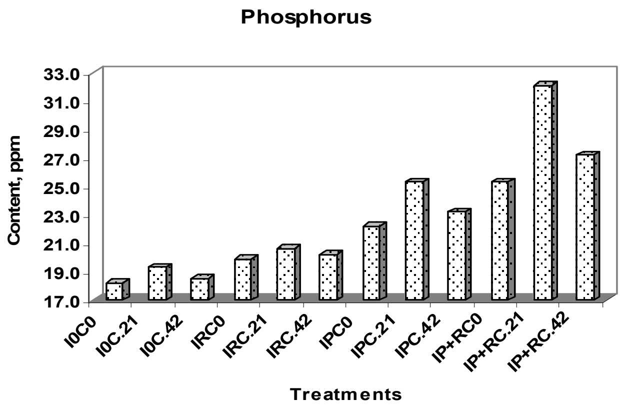
Treatment Combinations	**Nutrient Uptake**			
	kg ha^{-1}			**g ha^{-1}**
	N	**P**	**K**	**Co**
I_0C_0	96.3	10.4	16.3	4.3
$I_0C_{.21}$	103.6	12.6	19.1	7.1
$I_0C_{.42}$	99.1	11.3	17.6	5.6
I_RC_0	120.3	16.2	20.5	8.2
$I_RC_{.21}$	135.2	19.9	26.5	12.3
$I_RC_{.42}$	125.4	17.9	22.9	10.5
I_PC_0	111.1	16.1	21.4	11.3
$I_PC_{.21}$	129.2	19.3	24.2	14.9
$I_PC_{.42}$	120.5	17.7	23.4	13.4
$I_{P+R}C_0$	140.5	21.5	24.4	14.1
$I_{P+R}C_{.21}$	172.2	26	30.5	19.3
$I_{P+R}C_{.42}$	153.9	22.3	25.5	16.2
LSD (P=0.05)	11.31	4.49	2.13	5.61
At same inoculation				
At same cobalt level	8.06	2.51	1.85	2.64
Inoculation X Cobalt interaction	25.11	4.87	5.99	2.21

Note: I_0=No inoculation, I_R=Inoculation with *Rhizobium* and I_P=Inoculation with Phosphobacterium. C_0, $C_{.21}$, $C_{.42}$ = Cobalt @ zero, 0.21 and 0.42 kg ha^{-1} respectively. LSD: Least significant difference

3 CONCLUSIONS

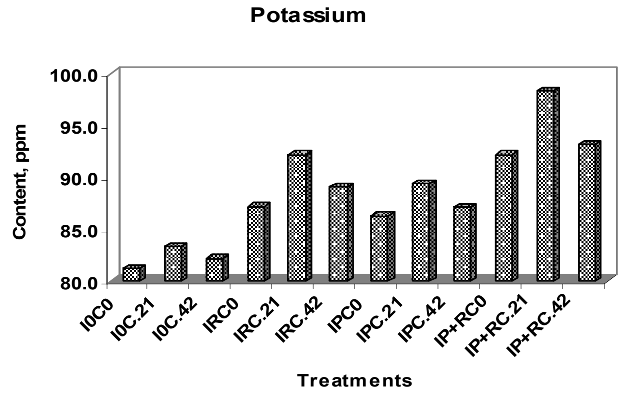
It could reasonably be said that the combined application of *Rhizobium* and phosphobacterium inoculation increased the yield and nutrient uptake of groundnut as compared to no inoculation or application of either of the two inoculation. Lower dose of cobalt (0.21 kg ha^{-1}) resulted in maximum benefit towards crop yield as compared to the higher dose. It could reasonably be said that the combined application of *Rhizobium* culture, phosphobacterium with lower dose of cobalt (0.21 kg ha^{-1}) resulted in maximum benefit towards crop yield as compared to the combined application of phosphobacterium inoculation and cobalt or single application of either inculation or cobalt. Besides the yield advantage, positive residual soil chemical properties in terms of available N, P, K and Co were also noted in the soil under the combined application of inoculations and cobalt as compared to sole application of only inoculation or cobalt.

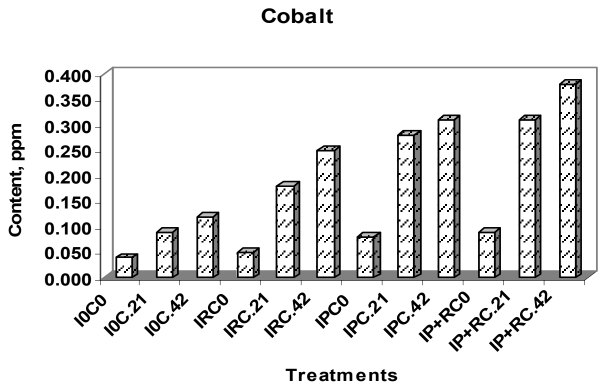




(Note: I_0=No inoculation, I_R=Inoculation with *Rhizobium* and I_P=Inoculation with Phosphobacterium. C_0, C_1, C_2 = Cobalt @ zero, 0.21 and 0.42 kg ha^{-1} respectively.)

Fig. 2: Effect of different levels of cobalt and inoculations on N and P content of soil after two years.





(Note: I_0=No inoculation, I_R=Inoculation with *Rhizobium* and I_P=Inoculation with Phosphobacterium. C_0, C_1, C_2 = Cobalt @ zero, 0.21 and 0.42 kg ha^{-1} respectively.)

Fig. 3: Effect of different levels of cobalt and inoculations on K and Co content of soil after two years.

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