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Effect of copper fertilization on plant growth, seed yield, copper and phosphorus uptake in soybean (*Glycine max*) and their residual availability in Mollisol

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ABSTRACT

A field experiment was conducted during the rainy season of 1997 and 1998 to study the effect of copper fertilization on plant growth, seed yield of soybean [*Glycine max* (L.) Merr.], copper and phosphorus uptake and their residual availability. Sixteen treatment combinations consisted of 4 varieties ('Bragg', 'PK 262', 'PK 416' and 'PK 1042') as main plots and 4 copper levels (0, 2.5, 5 and 10 kg Cu/ha) as subplot treatments were evaluated in split-plot design with 3 replications. Application of 2.5 kg Cu/ha significantly increased the number of nodules and their dry weight, dry-matter yield/plant and leaf-area index at 60 days, while leaf-area index increased significantly up to 5 kg Cu/ha. The seed yield did not increase with copper application. However, the interactions between varieties and copper levels revealed that variety 'PK 416' at 5 kg Cu/ha gave the highest number of nodules, their dry weight, leaf-area index and dry-matter yield at 60 days, seed yield, pods/plant, seed copper and phosphorus uptake at harvest, while 'Bragg' yielded lowest. Copper content in soybean seed was higher than its shoot. Shoot and seed phosphorus content and their uptake significantly increased with 2.5 kg Cu/ha and decreased with 10 kg Cu/ha. Copper application significantly increased the copper availability in soil at harvest, whereas the increase in available soil phosphorus was non-significant in northern Indian Mollisols. Differential response of varieties revealed that 'PK 416' showed a trend of increased growth and seed yield, while 'Bragg showed susceptibility to increasing copper and 'PK 1042' and 'PK 262' gave intermediate responses.

Key words : Copper fertilization, Soybean varieties, Growth, Yield, Copper, Phosphorus, Residual availability

Soybean with 40–42% protein and 20–22% oil is one of the major rainy-season cash crop and its cultivation is fast spreading in northern India. The importance of

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phosphate supply for the production of legume-based cropping system is well established. Interactions involving phosphorus and copper have been reported where heavy and prolonged application of phosphate fertilizers have induced or intensified copper deficiency, but the interactions with the limited experimental evidence of both antagonistic in citrus (Spencer, 1968) and synergistic relationship in subterranean clove plants (Greenwood and Hallsworth, 1960) have been reported under different set of experimental conditions. Such an effect in soybean has not yet been reported and needs to be studied in foothills of Uttranchal, Uttar Pradesh where soil is fertile.

MATERIALS AND METHODS

An experiment was conducted in 2 different fields during the rainy season of 1997 and 1998 at Crop Research Centre, Pantnagar. Wheat (*Triticum aestivum* L. emend. Fiori & Paol.) was grown before and after soybean experiment during both the years without any copper fertilization. The soil was silty clay loam with pH 7.3, containing organic carbon 0.793%, available N,P,K 325, 10.5 and 132 kg/ha respectively and 0.87 mg/kg soil available copper (0.005 m DTPA-extractable) which was sufficiently high for soybean. Sixteen treatment combination consisted of 4 varieties ('Bragg', 'PK 262', 'PK 416' and 'PK 1042') as main plots and 4 copper levels (0, 2.5, 5 and 10 kg Cu/ha) as subplot treatments were evaluated in split-plot design with 3 replications in plot of 6 m × 3.6 m. Copper was applied as $CuSO_4.5H_2O$ at the time of sowing and thoroughly mixed in 0–15 cm soil layer. The seeds of different soybean varieties were inoculated by *Bradyrhizobium japonicum* culture and was sown on 4 July in each year. The maturity period of the varieties used were 114, 125 and 120 days for 'Bragg', 'PK 262', 'PK 1042' respectively. Observations on nodulation/plant, dry-matter accumulation and leaf area were recorded from the observation strip at 60 days after sowing. Both plant phosphorus (by vanadomolybdo phosphoric acid yellow method in nitric acid system with spectrophotometer) and copper content (analysed by atomic absorption spectrophotometer) in seed and shoots of soybean at harvest were determined after triacid digestion (HNO_3 : H_2SO_4 : $HClO_4$ in ratio of 10:1:4 v/v). The nutrients uptake (shoot, grain and total) was calculated using the following formula :

$$\text{Nutrient uptake (kg/ha)} = \frac{\text{Nutrient content (\%)}}{100} \times \text{yield (kg/ha)}$$

The available soil phosphorus (Olsen's P) and copper (Lindsay and Norvell, 1978) after soybean cultivation were analysed to assess their residual status.

RESULTS AND DISCUSSION

Growth

Varietal differences, the effect of copper application as well as their interaction on number of nodules, their dry weight, dry-matter yield and leaf-area index/plant of soybean were significant (Table 1) at 60 days after sowing. 'PK 262' produced significantly highest nodules/plant and their dry weight except 'PK 416' for nodules/plant at 60 days, which were at par. Increase in copper application from 0 to 2.5

Table 1. Main and interactive effects between varieties and copper levels on nodulation, dry matter, leaf-area index and pods/plant of different varieties of soybean (pooled data of 2 years)

Variety	Copper levels (kg Cu/ha)				
	0	2.5	5	10	Mean
		Nodules/plant at 60 days			
'Bragg'	45.2	28.1	21.8	18.0	28.3
'PK 262'	45.8	63.1	56.1	47.1	53.0
'PK 416'	35.4	48.4	67.1	41.7	48.1
'PK 1042'	38.4	43.6	42.9	38.1	40.8
Mean	41.2	45.8	46.9	36.3	
CD (P=0.05)	5.64*	4.41**	8.83***	9.47****	
		Nodule dry weight/plant at 60 days (g)			
'Bragg'	0.34	0.25	0.20	0.16	0.237
'PK 262'	0.35	0.47	0.38	0.31	0.377
'PK 416'	0.27	0.34	0.49	0.33	0.395
'PK 1042'	0.25	0.26	0.29	0.31	0.279
Mean	0.303	0.336	0.340	0.278	
CD (P=0.05)	0.05*	0.03**	0.06***	0.07****	
		Dry-matter yield/plant at 60 days (g)			
'Bragg'	27.70	23.12	18.00	15.37	21.04
'PK 262'	34.24	41.36	34.60	28.15	34.59
'PK 416'	39.27	50.51	61.19	43.43	48.60
'PK 1042'	32.51	40.73	45.95	37.31	37.12
Mean	33.43	38.93	39.93	31.06	
CD (P=0.05)	4.44*	1.60**	3.21***	5.22****	
		Leaf-area index/plant at 60 days (g)			
'Bragg'	7.52	4.76	4.54	3.69	5.12
'PK 262'	7.87	9.33	9.71	7.92	8.70
'PK 416'	6.22	6.85	9.08	6.65	7.20
'PK 1042'	5.86	6.71	8.04	7.80	7.10
Mean	6.86	6.91	7.84	5.51	
CD (P=0.05)	0.95*	0.40**	0.79***	1.17****	
		Pods/plant			
'Bragg'	48.8	49.0	37.3	30.7	40.2
'PK 262'	59.4	84.6	68.6	56.8	67.3
'PK 416'	83.9	102.9	114.6	88.7	91.5
'PK 1042'	57.8	72.3	81.7	69.6	70.3
Mean	62.5	75.9	75.6	61.4	
CD (P=0.05)	2.57*	2.75**	5.51***	5.41****	

*For comparing main effects of varieties;**for comparing main effects of copper levels; ***for comparing interaction effect of 2 copper levels at same variety; ****for comparing interaction effect of 2 varieties atsame or different copper levels.

kg Cu/ha produced significantly more nodules and their dry weight and thereafter differences between 2.5 and 5 kg Cu/ha for nodule dry weight were non-significant. The interaction effect (Table 1) revealed that 'PK 416' at 5 kg Cu/ha and 'PK 262' at 2.5 Cu/ha produced maximum nodules and their dry weight at 60 days which were significantly more than that of other varieties at same or different copper levels, and at 10 kg Cu/ha nodulation significantly decreased. Similar increase in nodulation due to copper application were also reported (Cartwright and Hallsworth, 1970) in subterranean clover.

Dry-matter yield/plant increased with the crop age and at 60 days 'PK 416' produced significantly highest dry matter compared with all the varieties and 'Bragg recorded lowest. Application of 5 kg Cu/ha produced significantly highest dry-matter yield/plant, showing 19.4% more dry matter over the control, but the differences between 2.5 and 5 kg Cu/ha were non-significant. The interaction effect showed that variety 'PK 416' at 5 kg Cu/ha produced significantly highest plant dry matter at 60 days (Table 1) compared with other varieties at some or different copper levels and it was significantly reduced at 10 kg Cu/ha in all the varieties.

Highest leaf-area index was recorded in 'PK 262', followed by 'PK 416' and 'PK 1042' which were significantly higher than 'Bragg'. At 60 days, application of 5 kg Cu/ha recorded highest leaf-area index due to increased trifoliates/plant. The interaction effect (Table 1) at 60 days showed consistent increase of leaf-area index up to 5 kg Cu/ha in all the varieties except 'Bragg'. Thus, soybean varieties appeared to have different in their ability to utilise applied copper at 60 days, indicating the need of copper at peak growth stage for increasing trifoliates/plant, leaf area and ultimately increasing dry-matter yield/plant and copper uptake. This confirms the findings of Walker and Loneragan (1981) in subterranean clover that greater requirement of copper is necessary for producing new trifoliates after 30 days and also increased dry-matter yield/plant.

Grain yield

Varieties differed significantly in seed yield, whereas the average effects due to copper application was non-significant (Table 2), but their interaction effect was significant (Table 3). Highest seed yield was recorded in 'PK 416' and 'Bragg' yielded lowest. The differences in seed yield due to 'PK 1042' and 'PK 262' were also significant. Variety 'PK 416' registered significantly more 77.5, 45.6 and 23.7% seed yield than 'Bragg', 'PK 262' and 'PK 1042' respectively. Interaction effect (Table 3) revealed that 'PK 416' at 5 kg Cu/ha gave significantly highest seed yield than the other varieties at same or different copper levels, followed by 'PK 1042' and 'PK 262' each at 2.5 kg Cu/ha, whereas in 'Bragg' it decreased with increasing copper levels showing toxicity. Thus, the level of copper needed for maximum growth and grain yield differed for different varieties of soybean. The increase in seed yield with 'PK 416' at 5 kg Cu/ha may be attributed to an increase in nodulation, leaf-area index which enhanced the supply of metabolites in increasing dry-

Table 2. Effect of copper application on seed yield, phosphorus, copper content, uptake and their residual availability in soil after soybean (pooled data of 1997 and 1978)

Treatment	Seed yield (kg/ha)	Seeds/pod	100-seed weight (g)	P%		P uptake (kg/ha)			Cu (ppm)		Cu uptake (g/ha)			Available P in soil at harvast (kg P_2O_5/ha)	Available Cu Seed in soil at harvest (mg/kg soil)*
				Shoot	Seed	Shoot	Seed	Total	Shoot	Seed	Shoot	Seed	Total		
Variety															
'Bragg'	948	1.98	8.81	0.271	0.353	9.53	3.33	13.02	8.29	21.17	43.01	19.83	62.84	18.99	2.26
'PK 262'	1156	2.04	9.57	0.295	0.434	11.84	5.21	17.05	7.72	19.38	53.53	23.39	76.90	17.93	2.19
'PK 416'	1683	2.09	10.17	0.258	0.432	11.61	7.25	18.86	7.21	16.90	52.88	29.17	82.06	17.28	2.19
'PK 1046'	1360	2.05	9.80	0.275	0.420	12.97	5.72	18.69	8.81	19.21	60.89	26.26	87.16	16.92	2.07
CD (P=0.05)	169	0.07	0.45	0.016	0.018	0.166	0.73	1.10	0.28	1.04	5.32	3.84	4.92	0.86	0.11
CV%	13.2	3.9	4.7	5.8	4.53	14.4	13.5	6.51	3.54	5.40	10.1	15.6	6.3	4.8	5.0
Copper (kg Cu/ha)															
0	1273	2.05	9.41	0.278	0.415	11.95	5.26	17.54	6.43	15.91	44.28	20.24	64.52	18.28	0.81
2.5	1314	2.10	10.00	0.315	0.462	13.30	6.10	19.40	7.32	18.66	53.54	24.35	77.89	19.31	1.23
5	1288	2.05	10.01	0.265	0.406	11.49	5.53	17.02	8.42	20.00	55.32	26.60	81.90	18.18	2.83
10	1273	1.96	8.93	0.236	0.357	9.22	4.62	13.84	9.85	22.08	57.17	27.47	84.64	15.37	3.84
CD (P=0.05)	NS	0.03	0.21	0.018	0.021	1.03	0.34	1.08	0.52	0.58	3.23	1.68	3.22	1.24	0.12
CV%	86	2.8	2.6	8.1	6.2	10.6	7.4	7.5	7.6	3.5	7.2	8.1	4.9	8.3	6.4

NS, Non-significant
*0.005 m DTPA extractable

matter yield/plant, pods/plant, seeds/pod and 100-seed weight (Tables 1, 2). Similar increase in soybean seed yied (Oplinger and Ohlrogge, 1974) and pods/plant (Karlen and Hunt, 1974) due to copper application were also reported.

Copper content and uptake

The differences in copper content and uptake due to varieties and copper levels were significant (Table 2), whereas their interaction effect was significant only for grain copper uptake (Table 3). The copper content in seed was higher than that in shoot. 'PK 416' contained significantly lower copper (shoot and grain) than all the varieties, whereas 'Bragg' accumulated significantly highest copper in grain. The copper content and uptake increased significantly with increase in copper levels. Compared with the control increase of 13.8, 30.9 and 53.2% in shoot; and 17.2, 25.6 and 38.8% in seed copper content was observed in 2.5, 5 and 10 kg Cu/ha respectively. 'PK 1042' showed significantly highest copper uptake in shoot and 'PK 416' in seed, mainly due to increased copper content, dry-matter accumulation and seed yield.

The total copper uptake increased up to 5 kg Cu/ha and the difference between 5 and 10 kg Cu/ha was, non-significant. Interaction effect revealed that 'PK 416' and 'PK 1042' were at par for seed copper uptake when compared at same copper levels. Maximum seed copper uptake was obtained in 'PK 416' at 5 kg Cu/ha. Similar variations in copper content and uptake in soybean were also reported (Pyne *et al.*, 1986).

Phosphorus content and uptake

The difference due to varieties and copper levels for phosphorus content and uptake were significant (Table 2), whereas their interaction effect was significant only for seed phosphorus uptake (Table 3). Variety 'PK 262' showed the maximum phosphorus content in shoot, whereas its seed phosphorus content was at par with 'PK 416' and 'PK 1042', while 'Bragg' contained significantly lowest phosphorus both in shoot and seed phosphorus content over the control and thereafter it decreased significantly (ranging 12% to more than 25%) with increased copper levels. 'PK 1042' showed the maximum shoot phosphorus uptake and 'PK 416' showed highest seed phophorus uptake which were significantly more than all other varieties, whereas shoot, seed and total phosphorus yield were significantly increased with 2.5 kg Cu/ha, resulting 11.2, 15.9 and 10.6% increase over the control respectively. Interaction between varieties and copper levels for seed phosphorus uptake (Table 3) revealed that variety 'PK 416' at 5 kg Cu/ha showed the maximum uptake which was significantly higher than other varieties at same or different copper levels. This confirms the earlier results of Greenwood and Hallsworth (1960) in forage legumes with direct enhancement of phosphorus content and uptake by an increased copper supply. Higher copper levels significantly decreased the phosphorus yield due to decreased phosphorus content and seed yield. This may have been the inability of the plant to utilize phosphorus at higher copper levels (Spencer, 1966).

Table 3. Effect of interaction between varieties and copper levels on seed yield, phosphorus and copper uptake of soybean (pooled data of 2 years)

Variety	Copper levels (kg Cu/ha)			
	0	2.5	5	10
		Seed yield (kg/ha)		
'Bragg'	1,080	880	921	913
'PK 262'	1,108	1,387	1,038	1,092
'PK 416'	1,633	1,669	1,785	1,647
'PK 1042'	1,271	1,322	1,409	1,440
CD (P=0.05)		186.9*	233.3**	
		Seed P uptake (kg/ha)		
'Bragg'	3.91	3.80	3.03	2.59
'PK 262'	4.98	6.80	5.03	4.05
'PK 416'	6.88	7.66	8.03	6.42
'PK 1042'	5.25	6.17	6.04	5.43
CD (P=0.05)	0.93*	0.41**		
		Seed Cu uptake (g/ha)		
'Bragg'	17.68	18.50	20.49	22.65
'PK 262'	18.82	25.97	24.73	24.06
'PK 416'	23.32	28.28	33.12	31.98
'PK 1042'	21.11	24.66	28.08	31.21
CD (P=0.05)		3.36*	4.80**	

*For comparing 2 copper levels at same variety; **for comparing 2 varieties at same or different copper levels

Residual soil phosphorus and copper

Significant changes in available phosphorus (0.5 m $NaHCO_3$ extractable Olsen's P) and copper (0.005 m DTPA-extractable Cu) in soil left after harvest of soybean were observed (Table 2). Available soil P and Cu were maximum where 'Bragg' was grown, followed by 'PK 262', 'PK 416' and were significantly higher than 'PK 1042' where their availability were minimum. Application of 2.5 kg Cu/ha did not result any significant increase in soil-available P, whereas 10 kg Cu/ha significantly decreased (15.9%) available soil P and showed negative corelation (r=–0.281) with residual available copper. Again, copper application significantly increased the availability of copper at harvest at all copper levels. Compared to the control increase of 51.8, 249.3 and 374.6% was observed with 2.5, 5 and 10 kg Cu/ha respectively. This increased availability of soil copper due to copper application attributed to antagonistic behaviour of both copper and phosphorus in soil, resulting decreased availability of phosphorus in soil. Similarly, Prasad and Ram (1986) also reported decreased availability of Olsen's phosphorus in soil due to higher rates of copper application in

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