

## Minerals Composition of Hyacinth Bean (*Dolichos hyacinth* L.) Seeds as Influenced by *Bradyrhizobium* Inoculation and/or Chicken Manure or Sulphur Fertilization

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**Abstract:** Field experiments were carried out at Shambat, Sudan (Latitude 15° 40' N and Longitude 32° 32' E) in three consecutive seasons (2000/03) to investigate the effect of *Bradyrhizobium* inoculation and chicken manure or sulphur fertilization on minerals composition of hyacinth bean. The results obtained showed that inoculation, chicken manure, sulphur and their interactions significantly ( $P = 0.05$ ) improved both major and trace minerals composition of the seeds. The results also indicated that all measured parameters increased with increasing level of chicken manure or sulphur and the highest value of each mineral was observed with either 10 tone/fed chicken manure or 100 kg/fed sulfur with or without *Bradyrhizobium* inoculation. The study also showed that the residual effect of chicken manure or sulphur significantly ( $P = 0.05$ ) improved minerals content particularly at 10 tone/fed chicken manure or 100 kg/fed sulfur with or without *Bradyrhizobium* inoculation.

**Key words:** *Bradyrhizobium*, inoculation, chicken manure, hyacinth bean, mineral composition

### Introduction

The importance of legumes as food lied primarily in their high protein content that averages 20-25%. Hyacinth bean (*Dolichos hyacinth* L.) is an Asia origin crop and was probably domesticated in India as a cultivated grain legume. The crop is widespread in India, Egypt and Sudan as well as in Madagascar and Ethiopia. Hyacinth seed contains an average of 17% protein with *in vitro* protein digestibility of 80% (Murphy and Colucci, 1999). Leiner (1976) found that the calcium content of hyacinth bean was ranged from 167.4 to 231.1, magnesium from 38.7 to 48.6, potassium from 185.3 to 214, sodium from 6 to 9, iron from 11.7 to 18.1, copper from 1.4 to 2.5, manganese from 2.5 to 3.0 and zinc from 3.6 to 4.5 mg/100g. These nutritional characteristics coupled with the other environmental benefits make hyacinth bean a suitable food and fodder crop for the tropics. Hyacinth is an old established irrigated crop in Sudan. The crop is used for shifting cultivation or shorter rest periods of one or a few years for the usual method of maintaining soil fertility and organic matter (English *et al.*, 1999). Grain legumes generally fix about 15-210 kg/ha/y (Dakora and Keya, 1997). It has been found that inoculation of soybean by *Bradyrhizobium japonicum* significantly increased nodulation, yield and seed quality (Okereke and Onochie, 1996). Chicken manure amendments significantly improved the physical properties of the soil, such as water infiltration rate, water holding capacity, texture, reducing bulk density and hence increasing

porosity. Physical improvement of soil properties improves the uptake of nutrients and makes energy available for soil biota and thus influences many of biological processes of the plant which was found to affect the seed quality of faba bean (Elsheikh and Alzidany, 1997a,b). Chicken manure is considered to have fertilizing properties intermediate between mineral fertilizers and farmyard manure and it has an appreciable residual effect (Abdelgani *et al.*, 2003). Elemental sulphur has a variety of uses as soil amendment. The oxidation of elemental sulphur to  $H_2SO_4$  is particularly beneficial in alkaline soils in reducing the pH, supplying  $SO_4^{2-}$  to plants, making phosphorus and micronutrients more available and reclaiming soils (Lindemann *et al.*, 1991). The effectiveness of elemental sulphur depends upon the soil type, pH, organic matter content, clay minerals, depth of soil profile and drainage status. However, Ghani *et al.* (1997) reported that microbial population in soil is not a limiting in elemental sulphur oxidation. Efforts throughout the world are directed towards improving the quality of food crops by increasing the nutritional value of the grains and decreasing the antinutrients level. Breeding, fertilization programmes and genetic engineering are directed towards improving seed quality. *Rhizobium* inoculation of faba beans was reported to increase yield and protein content (Babiker *et al.*, 1995; Elsheikh and Osman, 1995). Addition of sulphur to faba bean plants increased seed yield by

33% and significantly increased the amount of protein and the concentration of methionine and cystine in seeds (Farrag *et al.*, 1992). No trials were conducted to study the effect of chicken manure or elemental sulphur in combination with *Bradyrhizobium* inoculation on chemical composition of plant seeds. Therefore, in this study we would like to investigate the effect of *Bradyrhizobium* inoculation, sulphur or chicken manure fertilization on major and trace minerals composition.

### Materials and Methods

**Materials:** Hyacinth bean (*Dolichos lablab* L.) cultivar used in this study was supplied kindly by Dr. Abd El Wahab, H. Abdalla, Department of Agronomy, Faculty of Agriculture, University of Khartoum, Shambat, Sudan. *Bradyrhizobium* (TAL 102) was obtained from the Biofertilization Department, Environment and Natural Resources Institute, National Centre for Research, Khartoum, Sudan. The Yeast Extract Mannitol (YEM) medium was prepared according to Cleyet-Marel (1993) method. Chicken manure was obtained from the Top Farm of Faculty of Agriculture, University of Khartoum, Shambat, Sudan. Elemental sulphur was obtained from El Geneed Sugar Industry, Sudan. Strains of *Bradyrhizobium* were preserved by streaking on YEM agar, mixed with 3.0g of calcium carbonate per litre, in slants in screw-caps test tubes and kept in the refrigerator at 4°C. Seeds were inoculated by mixing with a thick suspension of charcoal based *Bradyrhizobium* inoculum, with average count of  $1 \times 10^9$  cfu/g. Arabic gum solution (40%) was added for good adhesion. Seed inoculation was carried out in the farm and seeds were immediately sown in the soil and irrigated. Unless otherwise stated all chemicals and reagents used in this study are of reagent grade.

**Field experiments:** Three field experiments were conducted at the Demonstration Farm of the Faculty of Agriculture, Shambat, University of Khartoum, Sudan (Latitude 15° 40' N and Longitude 32° 32' E). The experiments were conducted during the seasons 2000/2001, 2001/2002 and 2002/2003.

**Chicken manure and sulphur application:** Two separate experiments were conducted, for one experiment chicken manure was applied at different levels (0, 2.5, 5.0, 7.5 and 10.0 t/fed). The fertilizer was distributed along the ridges and mixed with the soil and then the soil divided into plots. Thereafter, the plots were irrigated twice for two weeks before sowing. For the other experiment elemental sulphur was applied at different levels (0, 25, 50, 75 and 100 kg/fed), distributed along the ridges and mixed with the soil. The soil was divided into plots and the plots were irrigated twice before sowing for two weeks.

**Treatments:** The treatments were replicated three times in a split-split plot design. The treatments used during the first and second seasons were divided into groups as follows:

1. **Uninoculated:** Only chicken manure was applied at different levels (0, 2.5, 5.0, 7.5 and 10.0 t/fed).
2. **Inoculated:** The seeds were inoculated and the soil was amended with chicken manure at different levels (0, 2.5, 5.0, 7.5 and 10.0 t/fed).
3. **Uninoculated:** Only elemental sulphur was applied at different levels (0, 25, 50, 75 and 100 kg/fed).
4. **Inoculated:** The seeds were inoculated and the soil was amended with elemental sulphur at different levels (0, 25, 50, 75 and 100 kg/fed).

In the third season the residual effect of either chicken manure or sulphur in the presence or absence of *Bradyrhizobium* inoculation was investigated.

**Sample preparation:** Three samples from each plot were taken randomly after seeds matured. The seeds were dried by direct sun drying. The seeds were cleaned manually to remove husks, damage seeds and other extraneous materials. To determine the chemical composition, tannin and *in vitro* protein digestibility the cleaned seeds were ground to pass a 0.4 mm screen.

**Total mineral determination:** Minerals were extracted from the samples by the dry ashing method described by Walsh (1980). About 1.0 g sample was acid-digested with diacid mixture ( $\text{HNO}_3$ :  $\text{HClO}_4$ , 5:1, v/v) in a digestion chamber. The digested samples were dissolved in double-distilled water and filtered (Whatman No. 42). The filtrate was made to 50 ml with double-distilled water and was used for the determination of total minerals. The amount of iron, zinc, manganese and copper were determined using atomic absorption spectroscopy (Perkin-Elmer 2380, USA). Calcium and magnesium were determined by the titration method described by Chapman and Pratt (1961). Sodium and potassium were determined using a flame photometer (CORNIG EEL, London, UK) according to the AOAC (1995) method.

**Statistical analysis:** Experimental data were analyzed by using the general linear models procedure, the ANOVA procedure and Duncan's multiple range test (Duncan, 1955). Least significant differences were computed at  $P = 0.05$ . Data were also analyzed using the correlation procedure (Pearson's correlation coefficients) in SAS (1995).

### Results and Discussion

**Effect of treatments on major minerals content:** As shown in Table 1, *Bradyrhizobium* inoculation showed a significant ( $P = 0.05$ ) increase in calcium (Ca) content of hyacinth bean seeds in all seasons. Chicken manure

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Table 1: Effect of *Bradyrhizobium* inoculation and chicken manure or sulphur fertilization on calcium (Ca) content of hyacinth bean grown for three consecutive seasons

Ca content (mg/100g)									
Treatment	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means
Chicken M.									
Control	127.80	155.50	141.70	124.90	164.40	144.65	129.40	149.20	139.30
2.5 t/fed	155.50	159.30	157.40	155.60	175.54	165.57	17.00	151.80	144.40
5.0 t/fed	157.70	171.10	164.40	169.93	167.73	168.83	148.00	164.20	156.10
7.5 t/fed	173.50	181.30	177.80	166.67	183.33	175.00	160.80	176.00	168.40
10 t/fed	178.10	186.70	182.40	170.00	204.46	187.23	184.60	200.20	192.40
Means	158.52	170.78		157.42	179.10		151.96	168.28	
Overall			164.65			168.26			160.12
Sulphur									
Control	115.10	125.50	120.30	83.33	116.67	100.00	126.30	132.30	129.30
25 kg/fed	124.30	136.30	130.30	122.20	116.66	119.43	138.40	152.20	145.30
50 kg/fed	127.80	141.00	134.40	133.27	166.67	149.97	150.00	162.40	156.20
75 kg/fed	133.20	163.00	148.10	166.6	150.00	158.33	170.50	184.30	177.40
100 kg/fed	170.70	178.10	174.40	194.40	183.26	188.83	189.20	201.60	195.40
Means	134.22	148.78		139.97	146.65		154.88	166.56	
Overall			141.50			162.87			160.72
LSD (5%)									
Treatment(s)	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual 3 <sup>rd</sup> season		
Amendments	13.72			15.62			17.12		
Inoculated	8.68			6.80			10.15		
Amendments X Inoculated	22.06			22.10			24.84		
Amendment X level	36.63			36.30			38.21		
Amendment X Inoculated X level	53.04			50.02			54.73		

Table 2: Effect of *Bradyrhizobium* inoculation and chicken manure or sulphur fertilization on magnesium (Mg) of hyacinth bean grown for three consecutive seasons

Mg Content (mg/100g)									
Treatment	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means
Chicken M.									
Control	42.20	48.40	45.30	43.50	49.30	46.40	44.30	50.50	47.40
2.5 t/fed	47.30	55.50	51.40	48.10	54.70	51.40	48.00	58.80	53.40
5.0 t/fed	58.00	64.00	61.00	52.30	61.90	57.10	54.20	68.20	61.20
7.5 t/fed	66.40	70.20	68.30	57.40	68.20	62.80	72.40	78.50	75.50
10 t/fed	68.50	72.30	70.40	64.00	72.80	68.40	75.30	81.30	78.30
Means	56.48	62.08		53.06	61.38		58.84	67.40	
Overall			59.28		57.22				62.16
Sulphur									
Control	40.10	44.30	42.20	45.30	50.50	47.90	42.40	46.20	44.30
25 kg/fed	42.80	48.00	45.40	50.50	58.30	54.40	47.60	57.00	52.30
50 kg/fed	46.00	50.60	48.30	56.10	64.70	60.40	53.80	65.00	59.40
75 kg/fed	51.30	57.10	54.20	62.00	70.60	66.30	70.20	74.00	72.10
100 kg/fed	6.50	68.30	64.40	70.80	78.00	74.40	73.50	81.10	77.30
Means	48.14	53.66		56.94	64.42		57.50	64.66	
Overall			50.90			60.68			61.08
LSD (5%)									
Treatment(s)	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
Amendments	10.36			10.47			11.71		
Inoculated	5.64			4.48			7.31		
Amendments X Inoculated	15.07			15.12			16.22		
Amendment X level	24.21			24.20			24.72		
Amendment X Inoculated X level	33.93			32.81			33.35		

and sulphur fertilization were significantly ( $P = 0.05$ ) increased Ca content of uninoculated seeds with increasing level of each. However, after inoculation it was greatly increased in all seasons with maximum values obtained when 10t/fed of chicken manure was

applied (186.70, 204.46 and 200.20 mg/100g) for the first, second and residual season, respectively) or 100kg/fed of sulphur (178.10, 183.26 and 201.60 mg/100g for the first, second and residual seasons, respectively). The results obtained for both fertilizers

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Table 3: Effect of *Bradyrhizobium* inoculation and chicken manure or sulphur fertilization on potassium (K) content of hyacinth bean grown for three consecutive seasons

Treatment	K Content (mg/100g)								
	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means
Chicken M.									
Control	172.40	178.20	175.30	165.30	170.90	168.10	170.20	174.40	172.30
2.5 t/fed	180.00	184.40	182.20	171.50	177.30	174.40	180.00	184.80	182.40
5.0 t/fed	185.10	191.70	188.40	180.00	186.40	183.20	188.40	194.40	191.40
7.5 t/fed	190.30	200.10	195.20	192.00	198.80	195.40	200.20	214.40	207.30
10 t/fed	200.00	220.80	210.40	210.70	226.50	218.60	224.60	232.20	228.40
Means	185.56	195.04		183.90	191.98		192.68	200.04	
Overall			190.30			187.94			196.36
Sulphur									
Control	167.90	173.30	170.60	162.10	169.90	166.00	168.40	172.40	170.40
25 kg/fed	174.10	178.50	176.30	172.50	179.70	176.20	179.10	185.30	182.20
50 kg/fed	182.00	192.80	187.40	180.40	188.20	184.30	189.50	193.59	191.50
75 kg/fed	198.00	210.40	204.20	192.10	199.90	196.00	201.10	217.70	209.40
100 kg/fed	215.30	219.50	217.40	209.50	229.30	219.40	225.00	237.60	231.30
Means	187.46	194.90		183.32	193.40		192.62	201.3	
Overall			191.18			188.36			196.96
LSD (5%)									
Treatment(s)	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
Amendments	8.26			9.87			9.87		
Inoculated	3.30			3.89			5.42		
Amendments X Inoculated	10.65			13.96			13.96		
Amendment X level	18.30			22.07			22.07		
Amendment X Inoculated X level	27.06			31.22			31.22		

Table 4: Effect of *Bradyrhizobium* inoculation and chicken manure or sulphur fertilization on sodium (Na) content of hyacinth bean grown for three consecutive seasons

Treatment	Na Content (mg/100g)								
	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means
Chicken M.									
Control	5.40	6.20	5.80	5.00	5.80	5.40	4.80	5.00	4.90
2.5 t/fed	5.80	6.40	6.10	6.00	6.60	6.30	5.40	5.80	5.60
5.0 t/fed	6.10	6.70	6.40	6.90	7.30	7.10	6.00	6.40	6.20
7.5 t/fed	6.90	7.50	7.20	7.70	7.90	7.80	6.60	6.80	6.70
10 t/fed	7.50	8.10	7.80	8.20	8.60	8.40	7.10	7.30	7.20
Means	6.34	6.98		6.76	7.24		5.98	6.26	
Overall			6.66			7.00			6.12
Sulphur									
Control	6.00	6.60	6.30	5.20	5.60	5.40	5.10	5.70	5.40
25 kg/fed	6.90	7.10	7.00	5.80	6.20	6.00	5.90	6.10	6.00
50 kg/fed	7.30	7.70	7.50	6.50	6.90	6.70	6.50	6.90	6.70
75 kg/fed	8.00	8.60	8.30	7.10	7.30	7.20	7.05	7.35	7.20
100 kg/fed	8.50	9.10	8.60	7.80	8.40	8.10	7.90	8.30	8.20
Means	7.34	7.82		6.48	6.88		6.49	6.87	
Overall			7.58			6.68			6.68
LSD (5%)									
Treatment(s)	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
Amendments	0.83			0.33			0.13		
Inoculated	0.51			0.14			0.09		
Amendments X Inoculated	1.17			0.47			0.18		
Amendment X level	1.86			0.73			0.29		
Amendment X Inoculated X level	2.62			0.97			0.41		

indicated that fertilization of hyacinth bean by chicken manure or sulphur greatly increased Ca content of the crop seeds. Moreover, fertilization of inoculated seeds caused further increase in Ca content. Elsheikh and Mohamedzein (1998) reported that groundnut seed Ca

content did not show any pattern in response to *Bradyrhizobium* and/or VA mycorrhiza. However, Kawai and Yamamoto (1986) reported that inoculation with VAM increased plant development through supply of some elements such as Ca. Moreover, Giri (1993) reported

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Table 5: Effect of *Bradyrhizobium* inoculation and chicken manure or sulphur fertilization on iron (Fe) content of hyacinth bean grown for three consecutive seasons

Treatment	Fe Content (mg/100g)								
	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means
Chicken M.									
Control	8.14	8.38	8.26	7.75	7.85	7.80	7.10	7.80	7.45
2.5 t/fed	8.56	8.72	8.64	7.90	8.10	8.00	7.92	8.40	8.16
5.0 t/fed	9.38	9.48	9.43	8.24	8.32	8.27	8.50	9.04	8.77
7.5 t/fed	9.74	9.96	9.85	8.65	8.83	8.74	9.12	9.68	9.40
10 t/fed	10.28	10.48	10.36	8.94	9.18	9.06	9.74	10.32	10.03
Means	9.22	9.40		8.29	8.45		8.47	9.05	
Overall			9.31			8.37			8.76
Sulphur									
Control	8.12	8.28	8.15	8.05	8.15	8.10	6.94	7.62	7.28
25 kg/fed	8.38	8.56	8.47	8.32	8.46	8.39	7.67	8.09	7.88
50 kg/fed	8.92	9.38	9.15	8.74	8.92	8.85	8.16	8.84	8.50
75 kg/fed	9.58	9.76	9.67	9.04	9.22	9.13	8.93	9.41	9.17
100 kg/fed	10.19	10.39	10.29	9.10	9.24	9.17	9.52	10.08	9.80
Means	9.03	9.27		8.65	8.79		8.24	8.80	
Overall			9.15			8.72			8.52
LSD (5%)									
Treatment(s)	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
Amendments	0.16			0.18			0.22		
Inoculated	0.08			0.10			0.14		
Amendments X Inoculated	0.23			0.26			0.31		
Amendment X level	0.38			0.41			0.44		
Amendment X Inoculated X level	0.55			0.58			0.57		

Table 6: Effect of *Bradyrhizobium* inoculation and chicken manure or sulphur fertilization on copper (Cu) content of hyacinth bean grown for three consecutive seasons

Treatment	Cu Content (mg/100g)								
	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means
Chicken M.									
Control	1.22	1.26	1.24	1.12	1.21	1.17	1.04	1.62	1.33
2.5 t/fed	1.31	1.59	1.45	1.32	1.46	1.39	1.64	2.04	1.83
5.0 t/fed	1.68	1.86	1.77	1.74	1.92	1.83	2.07	2.71	2.39
7.5 t/fed	1.98	2.18	2.08	2.02	2.06	2.04	2.78	3.14	2.96
10 t/fed	2.28	2.46	2.37	2.12	2.16	2.14	1.94	3.02	2.48
Means	1.69	1.87		1.66	1.76		1.89	2.51	
Overall			1.78			1.71			2.20
Sulphur									
Control	1.12	1.28	1.15	1.08	1.16	1.12	1.03	1.59	1.31
25 kg/fed	1.22	1.28	1.25	1.24	1.38	1.36	1.60	1.98	1.79
50 kg/fed	1.31	1.59	1.45	1.56	1.72	1.64	2.02	2.64	2.33
75 kg/fed	1.66	1.88	1.77	1.89	2.03	1.96	2.71	3.19	2.95
100 kg/fed	1.92	2.18	2.05	2.09	2.19	2.14	3.22	3.76	3.49
Means	1.44	1.64			1.57	1.69		2.11	2.63
Overall			1.54			1.63			2.37
LSD (5%)									
Treatment(s)	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
Amendments	0.02			0.40			0.014		
Inoculated	0.01			0.02			0.009		
Amendments X Inoculated	0.05			0.06			0.020		
Amendment X level	0.07			0.08			0.032		
Amendment X Inoculated X level	0.20			0.13			0.045		

that application of 25 kg N/ha to groundnut increased crop uptake of Ca. The difference in response to such treatments could be attributed to the difference in cultivars as well as the growing environment. Chicken manure and sulphur application significantly ( $P = 0.05$ )

increased Magnesium (Mg) content of uninoculated seeds with increasing level of each (Table 2). However, after inoculation of the seeds the rate of improvement greatly increased in all seasons with maximum values obtained when 10t/fed of chicken manure was applied

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Table 7: Effect of *Bradyrhizobium* inoculation and chicken manure or sulphur fertilization on zinc (Zn) content of hyacinth bean grown for three consecutive seasons

Treatment	Zn Content (mg/100g)								
	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means
Chicken M.									
Control	1.23	1.67	1.45	1.17	1.79	1.48	1.26	1.72	1.49
2.5 t/fed	1.71	1.99	1.85	1.34	1.92	1.63	1.48	1.86	1.67
5.0 t/fed	2.03	2.63	2.33	1.56	1.98	1.77	1.76	2.10	1.93
7.5 t/fed	2.71	2.89	2.80	1.76	2.16	1.96	2.04	2.52	2.28
10 t/fed	2.92	3.06	2.99	1.94	2.42	2.18	2.16	2.88	2.52
Means	2.12	2.44		1.55	2.05		1.74	2.22	
Overall			2.28			1.80			1.98
Sulphur									
Control	1.20	1.72	1.46	1.15	1.67	1.41	1.25	1.77	1.51
25 kg/fed	1.65	1.93	1.79	1.43	1.85	1.64	1.50	1.92	1.71
50 kg/fed	1.87	2.13	2.00	1.73	1.97	1.85	1.82	2.14	1.98
75 kg/fed	2.15	2.53	2.34	1.98	2.18	2.08	2.10	2.68	2.39
100 kg/fed	2.30	2.88	2.59	2.19	2.53	2.36	2.21	2.93	2.57
Means	1.83	2.23		1.70	2.04		1.77	2.29	
Overall			2.03			1.87			2.03
LSD (5%)									
Treatment(s)	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
Amendments	0.06			0.07			0.21		
Inoculated	0.04			0.05			0.13		
Amendments X Inoculated	0.09			0.10			0.30		
Amendment X level	0.15			0.17			0.48		
Amendment X Inoculated X level	0.22			0.24			0.68		

Table 8: Effect of *Bradyrhizobium* inoculation and chicken manure or sulphur fertilization on manganese (Mn) content of hyacinth bean grown for three consecutive seasons

Treatment	Mn Content (mg/100g)								
	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means	Uninoculated	Inoculated	Means
Chicken M.									
Control	1.94	2.32	2.13	1.82	2.28	2.05	1.89	2.35	2.12
2.5 t/fed	1.99	2.43	2.1	1.89	2.41	2.15	1.98	2.48	2.23
5.0 t/fed	2.17	2.69	2.43	1.97	2.59	2.28	2.15	2.67	2.41
7.5 t/fed	2.43	2.95	2.69	2.09	2.73	2.41	2.46	2.96	2.71
10 t/fed	2.68	3.12	2.90	2.34	2.92	2.63	2.64	3.14	2.89
Means	2.24	2.70		2.02	2.58		2.22	2.72	
Overall			2.47			2.30			2.47
Sulphur									
Control	2.00	2.42	2.21	1.92	2.54	2.23	1.82	2.34	2.08
25 kg/fed	2.24	2.68	2.46	1.99	2.63	2.31	1.96	2.42	2.19
50 kg/fed	2.41	2.95	2.68	2.17	2.85	2.51	2.13	2.69	2.41
75 kg/fed	2.82	3.08	2.95	2.32	2.96	2.64	2.53	2.97	2.75
100 kg/fed	2.98	3.34	3.16	2.64	3.12	2.88	2.69	3.23	2.96
Means	2.49	2.89		2.20	2.82		2.23	2.73	
Overall			2.69			2.51			2.48
LSD (5%)									
Treatment(s)	1 <sup>st</sup> season			2 <sup>nd</sup> season			Residual (3 <sup>rd</sup> season)		
Amendments	0.10			0.06			0.22		
Inoculated	0.06			0.04			0.13		
Amendments X Inoculated	0.14			0.09			0.32		
Amendment X level	0.22			0.15			0.50		
Amendment X Inoculated X level	0.33			0.21			0.70		

(72.30, 72.80 and 81.30 mg/100g for the first, second and residual season, respectively) or 100kg/fed of sulphur (68.30, 78.00 and 81.10 mg/100g for the first, second and residual season, respectively). The results obtained for both fertilizers indicated that fertilization of

hyacinth bean by chicken manure or sulphur greatly increased Mg content of the crop seeds. Moreover, fertilization of inoculated seeds caused further increase in Mg content. Elsheikh and Mohamedzein (1998) reported that inoculation with *Bradyrhizobium* and/or VA

mycorrhiza significantly (~50.05) increased the seed content of Mg. Further they reported that the Mg percentage increased by N and P treatments and decreased when 0.01m ton manure were applied. The seed content of Mg was increased by inoculation. Kawai and Yamamoto (1986) reported that inoculation with VAM increased plant development through supply of some elements such as Mg. Giri (1993) reported that application of 25 kg N/ha to groundnut increased crop uptake of Mg. Chicken manure and sulphur application significantly ( $P = 0.05$ ) increased potassium (K) content of uninoculated seeds with increasing level of each (Table 3). However, after inoculation of the seeds it was greatly increased in all seasons with maximum values obtained when 10t/fed of chicken manure was applied (220.80, 226.50 and 232.20 mg/100g for the first, second and residual season, respectively) or 100kg/fed of sulphur (219.50, 229.30 and 237.60 mg/100g for the first, second and residual season, respectively). The results obtained for both fertilizers indicated that fertilization of soybean by chicken manure or sulphur greatly increased K content of the crop seeds. Moreover, fertilization of inoculated seeds caused further increase in K content. Elsheikh and Mohamedzein (1998) reported that inoculation with *Bradyrhizobium* and/or VA mycorrhiza significantly (~50.05) increased the seed content of K. The seed content of K was increased by inoculation and fertilization of the plant seeds. Chicken manure and sulphur application gradually increased sodium (Na) content of uninoculated seeds with increasing level of each (Table 4). Inoculation of the seeds significantly ( $P = 0.05$ ) increased Na content in all seasons with maximum values obtained when 10t/fed of chicken manure was applied (8.10, 8.60 and 7.30 mg/100g for the first, second and residual season, respectively) or 100kg/fed of sulphur (9.10, 8.40 and 8.30 mg/100g for the first, second and residual season, respectively). The results obtained for both fertilizers indicated that fertilization of soybean by chicken manure or sulphur greatly increased Na content of the seeds. Fertilization of inoculated seeds caused further increase in Na content. Elsheikh and Mohamedzein (1998) reported that inoculation with *Bradyrhizobium* and/or VA mycorrhiza significantly (~50.05) increased the seed content of Na.

**Effect of treatments on trace minerals content:** As shown in Table 5, *Bradyrhizobium* inoculation showed a significant ( $P = 0.05$ ) increase in iron (Fe) content of hyacinth bean seeds in all seasons. Chicken manure and sulphur fertilization were significantly ( $P = 0.05$ ) increased Fe content of uninoculated seeds with increasing level of each. However, after inoculation it was greatly increased in all seasons with maximum values obtained when 10t/fed of chicken manure was applied (10.48, 9.18 and 10.32 mg/100g) for the first,

second and residual season, respectively) or 100kg/fed of sulphur (10.39, 9.24 and 10.08 mg/100g for the first, second and residual seasons, respectively). The results obtained for both fertilizers indicated that fertilization of hyacinth bean by chicken manure or sulphur greatly increased Fe content of the crop seeds. Moreover, fertilization of inoculated seeds caused further increase in Fe content. The results obtained (rate of change) for Cu (Table 6), Zn (Table 7) and Mn (Table 8) were similar to that obtained for Fe. In general proper fertilization and inoculation provides the plant with both major and trace elements which reflects the nutritional status of the plant. Several minerals such as Ca, Fe, K, Na and Mg are essential for human and animal health. Knowledge about their level in different raw foods will provide information on the nutritional adequacy of diets. Other minerals, such as Cu, Mn and Zn though essential, have a limited range between required and toxic levels.

**Conclusion:** Chicken manure or sulfur fertilization in the presence or absence of *Bradyrhizobium* inoculation significantly increased mineral composition of hyacinth bean especially at a level of 10t/fed of chicken manure or 100kg/fed of sulphur. Proper fertilization programmes, focusing on biofertilization should be implemented to improve the productivity of food legumes and thereby increase total food production, improve the supply of good quality proteins as well as minerals in the diet of people who largely depend on food legume crops and improve seed quality. The latter implies processing, consumer, nutritional value and export quality. This investigation also calls food scientists to allow for the previous agronomic treatments, the history of the seeds, their origin and certification, before starting their experiments, analysis or interpreting their data.

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