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Effect of *Rhizobium* Inoculation and Organic Manures on Growth and Yield of Groundnut (*Arachis hypogaea* L.) in Malakal, Sudan*

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Abstract: Field experiments were conducted for two rainfed seasons (2006 and 2007) and one irrigated season (2008) at the Agricultural Farm of the Upper Nile University in Malakal (Sudan). The objective of the experiments was to study the effect of each of farmyard manure, water hyacinth compost, *Rhizobium* seed inoculation and in dual combinations on groundnut symbiotic performance and growth and yield under both rainfed and irrigation systems. The organic fertilizers were applied at the rate of 15 t ha^{-1} each. The results showed that each organic manure alone significantly increased shoot dry weight, number of nodules $plant^{-1}$ and pod and hay yields than the control in both rainfed and irrigation systems. *Rhizobium* seed inoculation with either manure significantly increased plant shoot and plant root dry weights, plant nodulation, early pod formation, absorption of plant N and plant P compared to the control. Moreover, these treatments significantly increased groundnut pod and hay yields over that of the control.

Key words: *Rhizobium* inoculation; organic manures; groundnut; Malakal (Sudan)

INTRODUCTION

Root-nodule bacteria (rhizobia) are symbiotic nitrogen fixers that are associated with the roots of leguminous plants. Rhizobia are common soil

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bacteria, but they often fail to produce effective symbiosis with some leguminous crops, either because of low population, weak competition and/or lack of compatibility with the crop. In many situations, establishment of effectively nodulated legumes requires the addition of specific rhizobia in a process known as *Rhizobium* seed inoculation (Mahdi 1993). The importance of biological nitrogen fixation was recognized in Sudan as early as 1925, when the Gezira scheme was started. In this regard, the amount of nitrogen fixed by the *Rhizobium*-groundnut symbiosis has been estimated to be 72% to 80 % of the crop requirement of nitrogen at the Gezira Research Station (Adlan and Mukhtar 2004). Therefore, nitrogen fixation through seed inoculation is most needed to reduce the cost of the relatively high expensive nitrogen fertilizer input.

Groundnut (*Arachis hypogaea* L.) is an important oilseed crop in Sudan. The early maturing Spanish-type cultivars are grown in rainfed areas of western and southern Sudan, whereas the Virginian types are grown in the irrigated areas of the central and eastern parts of the country. According to Mukhtar and Yousif (1979), Virginian-type varieties do not respond to nitrogen fertilization and/or seed inoculation, while the varieties of the Spanish-type respond.

The beneficial effect of *Rhizobium* inoculation on the growth, symbiotic properties and yield of groundnut has been recorded. In this respect, Sulfab (2010) found that variety Sodari (a Spanish type) required seed inoculation at Malakal. However, Haddad *et al*. (1986) stated that the native rhizobia were not able to supply the total nitrogen requirements of groundnut cultivars in the Sudan, and that inoculation with efficient, competitive rhizobia may be a beneficial practice.

Mukhtar (1993) found that addition of organic fertilizer (manure or compost) significantly increases the growth and yield of groundnut. This is due to the nitrogen and phosphorus present in the manure. Muller and Pereira (1995) reported that farmyard manure (FYM) treatment increased the nodule numbers and nodule mass by 34% and 36% of common bean, respectively, compared with the control.

Balasubramanian and Palaniappan (1994) reported that the use of microbial inoculants in combination with FYM significantly increased groundnut production. Radwan and Awad (2002) found that the addition of biofertilizer with either rice straw compost or water hyacinth compost significantly increased the yield of groundnut as compared to organic manure alone or chemical fertilizer application. The organic manure treatments and biofertilizer exhibited relatively high values of the soil nutrients N, P, K. Ca, and S (Stephen and Nybe 2003). Elsheikh et *al*. (2006) reported that inoculation of *Sinorhizobium* with chicken manure significantly increased nodulation and forage yield of alfalfa (*Medicago sativa* L.).

The objective of the present research was to study the effect of *Rhizobium,* farmyard manure and water hyacinth compost on growth, symbiotic properties and yield of the Spanish-type groundnut cultivar Sodari.

MATERIALS AND METHODS

Field experiments were conducted on groundnut under rain for two consecutive seasons (2006 and 2007) and one season under irrigation (2008) at the Agricultural Farm of the Upper Nile University in Malakal. Seeds of groundnut (*Arachis hypogaea* L.) variety Sodari were obtained from Elobeid Agricultural Research Station. Variety Sodari was chosen because it is early maturing (90 to 100 days) upright Spanish type, tolerant to water stress and is expected to suit the southern Sudan conditions.

Bradyrhizobium strain (Tal 1371), used in this study, was obtained from the National Centre for Research in Khartoum. The soil at the site has high clay content (>50%) and low water permeability. It is non - saline and non – sodic and has about 0.07% nitrogen, 0.44% organic carbon and somewhat moderate amount of available phosphorus (12 mg kg^{-1} soil). The soil is classified as Typic Haplusterts, fine, smectitic, isohyperthermic (Soil Survey Staff 1999). The total rainfall during July – October was 590 mm and during April - July was 380 mm. The land was

ploughed, harrowed and leveled. It was then divided into 5 x 3.6 m subplots. Five ridges per plot were prepared in north – south direction with 60 cm between ridges.

Nitrogen was added in the form of urea at the rate of 86 kg N ha^{-1} and phosphorus as a basal dose in the form of triple supper phosphate (TSP) at the rate of 43 kg P_2O_5 ha^{-1}. The treatments were the control, farmyard manure (15 t ha^{-1}), water hyacinth compost (15 t ha^{-1}), inoculation, inoculation plus farmyard manure and inoculation plus water hyacinth compost.

Each of the farmyard manure and the water hyacinth compost was added at the rate of 15 t ha^{-1}. The inorganic fertilizers were broadcast and incorporated into the soil before sowing. The manures were broadcast and incorporated into the soil three weeks before sowing. Some chemical properties of the manures were determined using micro-Kjeldahl method for nitrogen, Nelson and Sommers (1982) for organic carbon and Olsen and Sommers (1982) for phosphorus. The FYM contained 1.79% nitrogen, 14.3% organic carbon and 0.45% phosphorus, and the water hyacinth compost contained 1.26% nitrogen, 22.6% organic carbon and 0.32% phosphorus.

In both seasons, the seeds were sown by hand in holes on the top of the ridge at a depth of 4 to 5 cm and 15 cm between holes. Two seeds were planted per hole, as recommended by the Agricultural Research Corporation, Sudan. The sowing started in the first week of July in the rainfed season, when rainfall reached 25 mm or more. Pre-watering was done for the irrigated crop twice before sowing for closing the soil cracks and for weed control. The sowing date was the first week of April. Hand-weeding was carried out, starting at two weeks after sowing (WAS) and then after two weeks until 8 WAS.

The treatments were replicated four times in a completely randomized block design. Five plant samples were taken from each plot at 4 WAS and then three plant samples were taken at each of 6 and 8 WAS. A circular tunnel was made around the sampled plant to avoid the destruction of the lateral roots. A spade was used to lift up the soil clump inside the circle.

Each sample was put in a labeled paper bag and taken to the laboratory. The parameters measured were number of nodules plant^{-1}, shoot and root dry weights, early pod setting and shoot nitrogen content. Plant phosphorus percentage was determined according to Jackson (1958). At harvest, pod and hay yields per unit area were determined.

The data were subjected to analysis of variance, using the SAS statistical package (SAS 1982), and the means were separated by Duncan`s Multiple Rang Test.

RESULTS

Shoot and root dry weights

The data of the rainfed groundnut indicate that the inoculated treatments with either manure significantly ($P \leq 0.05$) increased shoot dry weight over that of each of all other treatments at 4 WAS (Table 1). Each of the remaining treatments significantly ($P \leq 0.05$) produced more shoot dry weight. At 6 WAS and 8 WAS, each of the treatments significantly ($P \leq 0.05$) increased shoot dry weight except for *Rhizobium* inoculation. As for the irrigated groundnut at 4 WAS through the 8th WAS, each treatment significantly ($P \leq 0.05$) increased shoot dry weight.

At 4 WAS, each of the rainfed treatments did not significantly increase the root dry weight. However, at 6 WAS through the 8th WAS each of the treatments significantly ($P \leq 0.05$) increased the root dry weight (Table 1). As for the irrigated groundnut at 4 WAS, each of the organic manure treatments alone significantly ($P \leq 0.05$) produced more root dry weight. At 6WAS through the 8th WAS each of the treatments significantly ($P \leq 0.05$) increased root dry weight.

Early pod setting and plant nodulation

There was no pod formation in both rainfed and irrigated groundnut at 4 WAS (Table 2). However, at 6 WAS, the rainfed groundnut showed that each of the treatments significantly ($P \leq 0.05$) produced more pod weight. At 8 WAS, inoculated treatments with either manure significantly ($P \leq 0.05$) produced more pod weight than the other treatments. Each of the

remaining treatments significantly ($P \leq 0.05$) increased pod weight. As for the irrigated crop, at 6 WAS through the 8th WAS, each of the treatments significantly ($P \leq 0.05$) increased pod weight.

At 4 WAS through the 8th WAS, rainfed treatments significantly increased plant nodulation. Each of the treatments significantly ($P \leq 0.05$) increased plant nodulation, (Table 2). For the irrigated crop at 4 WAS through the 8th WAS each treatment significantly ($P \leq 0.05$) increased groundnut nodulation. The best treatments were RM and RWHC at the 6th and 8th WAS (Table 2)

Plant shoot N and P percentages

At 4 WAS and the 6th WAS, the rainfed groundnut in each treatment significantly ($P \leq 0.05$) increased shoot N uptake (Table 3). At 8 WAS, each of the treatments invariably significantly ($P \leq 0.05$) increased shoot N content. As for the irrigated groundnut at 4 WAS through the 8th WAS, each of the treatments significantly ($P \leq 0.05$) produced more shoot N content than that of the control.

At 4 WAS, the rainfed groundnut in each treatment significantly ($P \leq 0.05$) increased shoot P content except for the *Rhizobium* inoculation alone (Table 3). However, at 6 WAS and 8 WAS, all the treatments significantly ($P \leq 0.05$) increased plant P content. As for the irrigated groundnut at 4 WAS, no treatment had any significant increase in plant P content. However, at 6 WAS and 8 WAS, all the treatments significantly ($P \leq 0.05$) increased plant P content (Table 3).

Pod and hay yields (t ha^{-1})

For both rainfed and irrigated groundnut, each of the inoculated treatments with either manure significantly ($P \leq 0.05$) produced more groundnut pod yield than that of each of the other treatments (Table 4). All the remaining treatments significantly ($P \leq 0.05$) increased pod yield. However, all the treatments significantly ($P \leq 0.05$) increased groundnut hay yield (Table 4).

Table1. Effect of *Rhizobium* inoculation and organic manures on shoot and root dry weights (g plant^{-1}) of rainfed and irrigated groundnut

Treatment	Rf			Ir		
	4 WAS	6 WAS	8WAS	4 WAS	6 WAS	8WAS
Shoot dry weight						
Control	2.1^{c}	5.9^{de}	12.2^{c}	0.80^{d}	3.3^{d}	9.4^{d}
M	4.2^{b}	12.2^{a}	18.9^{ab}	1.63^{a}	4.9^{bc}	12.6^{c}
WHC	4.1^{b}	10.3^{b}	17.8^{b}	1.48^{bc}	5.1^{b}	12.8^{c}
R	4.0^{b}	6.4 ^{d}	13.3^{c}	1.40^{c}	4.6^{c}	12.4^{c}
RM	5.0^{a}	10.3^{b}	20.1^{a}	1.58^{ab}	6.0^{a}	15.3^{a}
RWHC	4.9^{a}	8.5^{c}	20.3^{a}	1.53^{b}	5.6^{a}	14.1^{b}
SE±	0.5	1.1	2.0	0.1	0.4	0.7
CV (%)	22.5	25.1	24.4	15.6	14.1	10.2
Root dry weight						
Control	0.46^{a}	0.70^{c}	0.70^{d}	0.15^{c}	0.33^{d}	0.69^{d}
M	0.69^{a}	1.24^{a}	1.45^{c}	0.25^{a}	0.75^{b}	1.22^{a}
WHC	0.58^{a}	1.15^{ab}	1.92^{a}	0.28^{a}	0.76^{ab}	1.20^{a}
R	0.59^{a}	1.06^{b}	1.42^{c}	0.15^{c}	0.52^{b}	0.90^{c}
RM	0.69^{a}	1.13^{b}	1.49^{bc}	0.19^{b}	0.84 ^{a}	1.20^{a}
RWHC	0.65^{a}	1.22^{a}	1.62^{b}	0.19^{b}	0.74^{b}	1.02 ^{b}
SE±	0.1	0.1	0.15	0.03	0.09	0.09
CV (%)	28.8	24.7	25.6	27.04	23.9	16.8

Means followed by the same letter(s) in a column are not significantly at $P \leq 0.05$, according to Duncan's Multiple Range Test.

Rf = rainfed groundnut; Ir = irrigated groundnut; R = *Rhizobium* seed inoculation; M = farmyard manure; WHC = water hyacinth compost; WAS = weeks after sowing

Table 2. Effect of *Rhizobium* inoculation and organic manures on early pod setting (g plant^{-1}) and number of nodules of rainfed and irrigated groundnut

Treatment	Rf			Ir		
	4 WAS	6 WAS	8WAS	4 WAS	6 WAS	8 WAS
Early pod setting						
Control		1.3^{d}	9.0^{e}		0.3^{d}	4.5^{e}
M		5.8^{c}	17.3^{d}		4.8^{c}	16.7^{b}
WHC		10.4^{a}	20.7^{c}		10.2^{a}	13.4^{c}
R		7.2^{c}	23.4^{b}		7.3^{b}	9.0^{d}
RM		8.9^{b}	30.2^{a}		8.7^{b}	19.4^{a}
RWHC		11.4^{a}	29.8^{a}		11.5^{a}	16.4^{b}
SE±		1.4	2.7		1.4	1.6
CV (%)		25.9	24.8		22.0	19.3
Number of nodules						
Control	36^{c}	52^{c}	145^{c}	16^{c}	34^{d}	64^{e}
M	58^{b}	111^{a}	254^{a}	40^{a}	80^{c}	105^{d}
WHC	59^{b}	94^{b}	271^{a}	40^{a}	87^{c}	101^{d}
R	63^{ab}	111^{a}	207^{b}	30^{b}	77^{c}	127^{c}
RM	69^{a}	96^{b}	253^{a}	37^{a}	131^{a}	167^{a}
RWHC	68^{a}	98^{ab}	274^{a}	37^{a}	108^{b}	139^{b}
SE±	6.0	13.7	29.5	3.7	13.6	11.0
CV (%)	23.5	34.8	28.5	20.7	25.6	16.6

Means followed by the same letter(s) in a column are not significantly at $P \leq 0.05$, according to Duncan's Multiple Range Test.

Rf = rainfed groundnut; Ir = irrigated groundnut; R = *Rhizobium* seed inoculation; M = farmyard manure; WHC = water hyacinth compost; WAS = weeks after sowing

Table 3. Effect of *Rhizobium* inoculation and organic manures on the nitrogen and phosphorus in the shoots of groundnut

Treatment	Rf			Ir		
	4 WAS	6WAS	8WAS	4 WAS	6 WAS	8WAS
Shoot N percentage						
Control	1.72^{d}	1.64^{d}	1.26^{d}	1.72^{d}	1.26^{b}	1.29^{d}
M	2.11^{b}	1.91^{c}	1.61^{b}	1.95^{c}	1.93^{a}	1.86^{a}
WHC	2.09^{bc}	2.20$^{a\ b}$	1.63^{b}	2.08^{b}	1.95^{a}	1.79^{b}
R	2.00^{c}	1.96^{c}	1.61^{b}	2.02^{b}	1.88^{a}	1.70^{c}
RM	2.00^{c}	2.10^{b}	1.72^{a}	1.97^{bc}	1.99^{a}	1.82^{ab}
RWHC	2.8^{a}	2.23^{a}	1.53^{c}	2.23^{a}	1.99^{a}	1.68^{c}
SE±	0.1	0.1	0.07	0.1	0.07	0.06
CV (%)	11.8	15.9	10.4	7.7	7.2	7.1
Shoot P percentage						
Control	0.20^{c}	0.161^{c}	0.12^{d}	0.21^{a}	0.18^{b}	0.11^{d}
M	0.23^{a}	.191^{ab}	0.15^{c}	0.22^{a}	0.21^{a}	0.18^{b}
WHC	0.23^{a}	0.185^{b}	0.16^{b}	0.22^{a}	0.22^{a}	0.20^{a}
R	0.21^{bc}	0.198^{a}	0.17^{a}	0.21^{a}	0.22^{a}	0.18^{b}
RM	0.22^{ab}	0.189^{b}	0.17^{a}	0.22^{a}	0.22^{a}	0.21^{a}
RWHC	0.22^{ab}	0.190^{b}	0.16^{b}	0.22^{a}	0.22^{a}	0.17^{b}
SE±	0.013	0.007	0.006	0.012	0.01	0.01
CV (%)	14.7	8.8	8.03	10.7	6.5	8.8

Means followed by the same letter(s) in a column are not significantly at $P \leq 0.05$, according to Duncan's Multiple Range Test.

Rf = rainfed groundnut; Ir = irrigated groundnut; R = *Rhizobium* seed inoculation; M = farmyard manure; WHC = water hyacinth compost; WAS = weeks after sowing

Table 4. Effect of *Rhizobium* inoculation and organic manures on pod and hay yields (t/ha^{-1}) of rainfed and irrigated groundnut

Treatment	Rf		Ir	
	Pod yield	Hay yield	Pod yield	Hay yield
Control	3.4 ^{c}	2.41^{d}	2.9^{c}	2.0^{c}
M	6.5^{b}	3.03^{c}	5.0^{b}	2.8^{b}
WHC	6.5^{b}	3.05^{c}	5.3^{ab}	2.9^{b}
R	6.9^{a}	3.00^{c}	5.1^{ab}	3.0^{b}
RM	6.7^{ab}	3.41^{b}	5.3^{ab}	3.4^{a}
RWHC	6.8^{ab}	3.63^{a}	5.4^{a}	3.5^{a}
SE±	0.3	0.1	0.3	0.12
CV (%)	8.3	11.7	7.0	12.3

Means followed by the same letter(s) in a column are not significantly at $P \leq 0.05$, according to Duncan's Multiple Range Test.

Rf = rainfed groundnut; Ir = irrigated groundnut; R = *Rhizobium* seed inoculation; M = farmyard manure; WHC = water hyacinth compost

DISCUSSION

Addition of manure with *Rhizobium* inoculation resulted in significant increase in shoot and root dry weights. These results are in agreement with those of Babiker *et al*. (2009) who found that the treatments containing *Bradyrhizobium* strains and chicken manure significantly increased shoot dry weight of guar. Also, Elsheikh *et al*. (2006) reported that inoculation of *Sinorhizobium* with chicken manure significantly increased nodulation and forage yield of alfalfa (*Medicago sativa* L.). Similarly, Abdelgani (1997) found that *Rhizobium* inoculation significantly increased shoot dry weight, nodulation, and shoot nitrogen content of a fenugreek (*Trigonella foenum-graecum* L) cultivar.

Each of the two organic manure treatments alone or with inoculation significantly increased plant nodulation. This result is in conformity with

that of Aouani *et al.* (1997) who showed that the inoculation with *Rhizobium* increased the nodulation of common bean. In this study, the *Rhizobium* inoculation gave 207 nodules plant^{-1} under rainfed condition and 127 under irrigation at 8thWAS. This could probably be attributed to the number of *Rhizobium* in the soil. Muller and Pereira (1995) reported that farmyard manure treatment increased the nodule numbers and nodule mass of common bean by 34% and 36%, respectively. In the present research, farmyard manure gave 75% more groundnut nodules than the control under rainfed condition, whereas the irrigated groundnut gave 64% more nodules than the control at the 8th WAS.

Either of the organic manure treatments alone or with inoculation significantly increased both shoot N and P percentages. This result is in conformity with that of Stephen and Nybe (2003) who showed that treatments containing organic manure and biofertilizer exhibited high values of soil N, K, P, and Ca. *Rhizobium* seed inoculatin increased plant shoot N by 0.35% and 0.41% over the control for the rainfed and irrigated groundnut, respectively, at the 8 th WAS. In this regard, Chafi and Bensoltane (2009) showed that inoculation of faba bean with strains of *Rhizobium leguminosarum* significantly increased plant shoot N compared with the control.

Each of the organic manures in combination with inoculation increased pod yield of rainfed and irrigated groundnut. Similar finding was reported by Radwan and Awad (2002) who found that the addition of biofertilizer with either water hyacinth compost or rice straw compost significantly increased the yield of groundnut as compared to organic manure alone or chemical fertilizer. Also, Mukhtar (1993) showed that addition of organic fertilizers (manures or compost) significantly increased the growth and yield of groundnut. Similar result was obtained by Balasubramanian and Palaniappan (1994) who reported that the use of microbial inoculants in combination with farmyard manure significantly increased groundnut production.

The present work indicated that each of the organic manures with inoculation significantly increased groundnut hay yield. This result is in agreement with that of Elsheikh *et al*. (2006) who reported that inoculation of *Sinorhizobium* with chicken manure increased nodulation and forage yield of alfalfa (*Medicago sativa* L.). Also, Prasad *et al*. (2002) showed that application of green manure in groundnut significantly increased pod and hay yields.

CONCLUSIONS

1-The use of water hyacinth as compost is promising for improving soil fertility, particularly for the lands adjacent to the banks of the White Nile river.

2-Composting by water hyacinth could be used as a beneficial means of getting rid of this water weed that has polluted the White Nile river.

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تأثير التلقيح بالرايزوبيم والأسمدة العضوية على نمو و إنتاجية الفول السوداني في ملكال (السودان)*

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و أدم إبراهيم أدم[3]

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المستخلص : أجريت تجارب حقلية في موسمين مطريين (2006 و 2007) وموسم مروي (2008) بمزرعة كلية الزراعة ، جامعة أعالي النيل بملكال (السودان) بهدف دراسة تأثير كل من السماد البلدي ، والسماد العضوي المصنع (كومبست) المكون من أعشاب النيل، وبكتيريا الرايزوبيا كسماد أحيائي كمعاملات منفردة ً وثنائيه على النمو والخواص التكافلية وإنتاجية الفول السوداني في المواسم الثلاثة. استخدمت الأسمدة العضوية بمعدل 15 طن/هكتار لكل منهما. أثبتت النتائج أن استخدام أي من السمادين العضويين مفرداً أدى إلى زيادة معنوية في الوزن الجاف للنبات، وعدد العقد الجذرية للنبات، وإنتاج قرون الفول والتبن تحت ظروف الري والمطر بالمقارنةً مع الشاهد. أدت المعاملات الملقحة في وجود أي من السمادين العضويين إلى زيادة معنوية في الوزن الجاف للنبات وعدد العقد الجذرية للنبات والتبكير في تكوين الثمار، كما أدت إلى زيادة معنوية في إمتصاص النتروجيين والفسفور مقارنةً بالشاهد. أدت المعاملتان المذكورتان آنفا إلى زيادة معنوية كبيرة في إنتاج قرون الفول السوداني و التبن مقارنةً بالشاهد.

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