

Performance of cowpea (*Vigna unguiculata* L.) as influenced by inoculation with indigenous Omani rhizobia and mineral nitrogen fertilization.

M. A. Hadad^{1*}, H.S. Al-Hashmi², N. Al-Wihaibi² and M. M. Samoal¹

¹Department of Soils And Water, Sudan University Of Science & Technology, Shambat, Sudan.

²Agricultural Research Center, P.O.Box 50, P.C.121, Ministry Of Agriculture & Fisheries, Sultanate of Oman.

*Corresponding author

The current research was undertaken to investigate the response of a popular fodder crop (cowpea) to Inoculation with indigenous strains of *Rhizobium* from Oman, and to mineral nitrogen additions (urea and ammonium sulfate) at two different salinity levels of irrigation water. Both *Rhizobium* strains from Batinah and Sharqia consistently improved nitrogen fixation parameters including tissue dry weight and nodulation. The low tissue nitrogen obtained was explained as due to the dilution effect resulting from the high vegetative growth obtained with *Rhizobium* inoculation. The strain from Sharqia showed a relatively superior performance as compared to the Batinah strain, being highly infective (nodulation) and effective (tissue nitrogen). Addition of mineral nitrogen significantly enhanced growth parameters and out yielded the two tested *Rhizobium* strains in all growth parameters. The tested local *Rhizobium* strains were therefore unable to supply the tested cowpea with the needed nitrogen for optimum growth. The effect of salinity was negligible probably due to the fact that most of the fodder crops recommended for production were preselected by the research center for high salinity tolerance. Further research is being stressed to select highly infective and effective *Rhizobium* strains capable of supplying fodder crops with the optimum nitrogen for healthy growth to attain high productivity.

Key words: Inoculation, *Rhizobium*, Mineral fertilizers, *Vigna unguiculata*, Nitrogen fixation.

Despite the significance of agricultural production to the Omani farmer as a means of generating additional income, no available research information to guide the farmer to apply the recent technological advances in this field. The information available is so scanty, which necessitates serious investigations in this regard.

The sandy soils of Oman are deficient in organic matter (Soil Survey Report, 1994). Also serious nutrient deficiencies were noted in all the traditionally grown crops (Parkash, 1989). The salinity of irrigation water and secondary salinization of the soils is a serious obstacle for stable production of economical crops in Oman for the Omani farmer as well as to the concerned researcher in this field (ARC report, 1990). All these add to the arid dry weather which hinders feasible agricultural production. This merits serious experimentation with all the

factors of production to make available to the farmer the necessary information and technology he needs for maximizing his production and therefore his income.

This study was therefore undertaken to investigate the productivity of Omani lubia (*Vigna unguiculata*) commonly grown in Oman through improving the fertilization practice as compared to the use of *Rhizobium* inoculation by exploiting the available indigenous nitrogen fixing bacteria as a cheap source of nitrogen to maximize the farmers income. The effect of salinity was also explored in an attempt to select the most efficient *Rhizobium* strains suitable for the most adaptable cultivars for the country.

MATERIALS AND METHODS

A greenhouse experiment was established at Rumais ARC (Agricultural

Research Center, Oman). Cowpea (*Vigna unguiculata*), a local variety traditionally grown in the Batinah area in Oman was included in the trial. The seeds obtained from the local farmers, were pregerminated in petri dishes after surface sterilization with 95% ethyl alcohol followed by three successive washes with sterile tap water. The soils were selected based on salinity level, where the one with the lowest level was chosen (the properties of the selected soil was shown in table 1). Six kilograms were thereafter transferred to each plastic pot with the capacity of 8 kilograms. Nonsaline water was added to each pot to field capacity. Five sterile seeds were carefully transferred to each pot at 2-cm depth. Methods of rhizobia isolations, purification, authentication, and preparation for inoculation were as described earlier by Vincent (1972), Habte (1985) and as reported earlier by Hadad et al (1998). One ml of each of the specific selected cowpea rhizobia (Batina & Al-Sharquia) was aseptically added to the sown seeds in each pot which were irrigated immediately with nonsaline water to avoid desiccation and to hasten germination. Nitrogen was added either in the form of ammonium sulfate or urea three times during the growth period; at sowing, 2 weeks from sowing, and at the flowering stage i.e. 35 days from sowing. The fertilization dose was calculated based on the recommendation issued by the research center and as practice by Omani farmers (200 kg/ha), and was calculated for each pot and split as described earlier. Sowing, inoculation and the first dose of fertilization were completed on the same day. Two water salinity levels were used for irrigating the pots i.e. 1 & 8 ds/m. The saline waters were selected from the natural wells prevailing in the area (Farm No 1 & Barka farm of the ARC). Little adjustments were made by mixing saline waters to get the desired salinity level. A complete nutrient solution lacking nitrogen (Halliday, J. 1981, Asad et al, 1991) was added to all the plots interchangeably with the added irrigation water during the whole growth period. With one legume used in the trial, 5 treatments, 2 salinity levels, and 4 replication, a total of 40 pots were laid out in a split plot design under the shade. The pots were weekly rerandomized. Daily observations included color rating plus other general observations relating to vegetative growth as influenced by salinity levels, diseases & insect infestations. Two samples were taken for analysis during the whole growth period

which continued for 2-month. Upon harvest, soil samples were taken for analysis. Fresh top weights, nodule number, and fresh root weights were also taken. The fresh tissue was dried in an oven at 72°C for 48 hours and the dry weights were taken. Tissue nitrogen was determined using conventional Kjeldahl method. MstatC was used for statistical analysis.

RESULTS AND DISCUSSION

The data are presented in tables (1-3). At the lower salinity level (1 DS/ m), both *Rhizobium* strains from Batinah and Sharquia out-yielded all other treatments in tissue dry weight including mineral nitrogen addition treatments (urea and ammonium sulfate). The control treatment was significantly lower than all the four other treatments. The same trend was observed with the other measured parameters including root dry weight and nodulation. This super performance of the *Rhizobium* strains, however, was not reflected in the tissue nitrogen obtained. The addition of ammonium sulfate significantly out yielded all other treatments followed by urea. The higher tissue dry weight obtained with the addition of rhizobia may have exerted a dilution effect on tissue nitrogen.

Raising the salinity to 8 DS/m resulted in rather an erratic data and no apparent effect was noticed on the nitrogen fixation traits measured. The soil chemical properties presented in table 1 reflected a dramatic increase in soil EC after treatment with the higher saline irrigation water i.e. 8DS/m. Both nitrogen percent and phosphorus levels declined following the addition of the higher saline water. This could be attributed to the high salinity tolerance of the cultivars tested when used in this experiment since they were preselected for tolerance to saline water by the research center in Oman to cater for the intrusion of sea water in Oman.

Singleton et al, 1982 could not find variations among the cowpea cultivars they inoculated under different saline irrigation water. They concluded that the effect of saline water is dependant among others on the tolerance of the cultivars to water salinity. The addition of urea resulted in a significant increase in tissue dry weights over all other treatments except for the values obtained with the strain from Sharquia. This strain, however, increased tissue dry weights and gave values comparable to those obtained with urea addition. Significant increase in nodulation

Table 1: Chemical & Physical properties of the soils used in the trial at the beginning and at the end of the experiment

	ECe DS/m	pH	Cl me/l	Na me/l	Ca me/l	N (%)	K me/l	CaCO ₃ (%)	O.M. (%)	Avail. P. ppm
*	6.75	8	27.15	19.5	6.25	0.001	0.4	40.2	0.2	6.16
**	5.065	7.05	36.25	25.15	9.5	0.0085	2.95	34.0	0.2	11.7
***	11.04	6.8	93.25	31.5	23.5	0.0135	3.65	40.1	0.16	7.5

* Initial soil at the beginning of the experiment

** Upon irrigation with 1 DS/m

*** Upon irrigation with 8 DS/m

Table 2. The Performance of *Rhizobium* strains with the tested cowpea cultivar as a response to mineral nitrogen fertilization under 1 DS/m irrigation water.

Treatment	Measured traits			
	Top Fresh Weight(g)	Top Dry Weight(g)	Nodule Number	Nitrogen (%)
Cowpea R. (Batinah)	6.220 a*	0.986 a	41 ab	2.072 c
Cowpea R.(Sharquia)	6.138 a	0.884 a	23 bc	2.300 bc
Urea	5.938 a	0.872 a	11c	2.971 b
Ammonium Sulphate	5.938 a	0.730 b	06 c	4.300 a
Control	5.470 ab	0.718 b	0.0 d	2.523 bc

*Numbers followed by the same letters within columns are not significantly different at the 0.05 level of significance by the Duncan Multiple range Test

Table 3. The Performance of *Rhizobium* strains with the tested cowpea cultivar as a response to mineral nitrogen fertilization under 8 DS/m irrigation water.

Treatment	Measured traits			
	Top Fresh Weight (g)	Top Dry Weight (g)	Nodule Number	Nitrogen (%)
Cowpea R. (Batinah)	4.616 bc*	0.589 bc	10 c	2.155 c
Cowpea R. (Sharquia)	5.549 ab	0.645 b	50 a	2.279 bc
Urea	5.939 a	0.690 b	05 c	4.663 a
Ammonium Sulphate.	4.191 c	0.471 b	13 c	4.394 a
Control	4.110 c	0.490 c	00 d	2.257 bc

*Numbers followed by the same letters within columns are not significantly different at the 0.05 level of significance by the Duncan Multiple range Test.

was also obtained upon addition of the Sharquia strain. This was further reflected in the relatively high percent nitrogen obtained as compared to the Batinah strain and the control treatment. Davis and Somasegaran, 1981 found positive correlation between dry tissue nitrogen and tissue nitrogen percent in many of the leguminous crops tested. The highest nitrogen percentage, however, was obtained with the addition of mineral

nitrogen either in the form of urea or ammonium sulfate.

It could therefore be concluded that the local *Rhizobium* strains tested although gave a good performance regarding plant growth parameters, were not able to supply the crop with the optimum nitrogen needed for growth. The effect of salinity was negligible, although the strain from Sharquia seemed to be more tolerant to higher

salinity effects. This finding was reported earlier by Hadad and Al-Hashmi, 1999 when testing different *Rhizobium* strains from different locations in Oman under laboratory conditions. The strain from Sharqia was superior to all other tested strains when grown under laboratory conditions including some exotic strains from Canada and Australia.

REFERENCES

- Asad, S., K.A. Malik and F.Y. Hafeez. 1991. Competition between inoculated and indigenous *Rhizobium/Bradyrhizobium* spp. strains for nodulation of grain and fodder legumes. Pakistan Biol Fertil Soils 12:107-111.
- Davis, R.J. and P. Somasegaran. 1981. A worldwide network of inoculation trials. p. 515. In A.H. Gibson and W.E Newton (eds) Current Perspectives in Nitrogen Fixation. Australian Acad of Sci, Australia
- Habte, M. 1985. Selective medium for verifying specific populations of rhizobia introduced into tropical soils. Appl Environ Microbiol 50(6):1553-1555.
- Hadad et al , 1998. Annual Agricultural Research Report. Ministry of Agriculture & Fisheries, Sultante of Oman.
- Hadad, M.A. and Hamood, S. H. 2002. Response of fodder legumes to *Rhizobium* inoculation. ARC Annual Report. Sultanate of Oman.
- Halliday, J. 1981. Biological nitrogen fixation: its potential in tropical soils. In S.O. Emejuaiwe, O. Ogunbi and S.O. Sanni (eds) Global Impacts of Applied Microbiology. Academic Press, London pp. 73-84.
- Parkash, S.R.1989. Inoculation of legumes in Oman. ARC Annual Reports. Ministry of Agriculture & Fisheries, Sultanate of Oman.
- Singleton, P.W., S.A. El Swaify and B.B. Bohlool. 1982. Effect of salinity on *Rhizobium* growth and survival. Appl Environ Microbiol 44(4):884-890
- Somasegaran, P. and B.B. Bohlool. 1990. Single-strain vs. multistrain inoculation on rhizobial strain effectiveness and competition for nodulation on chickpeas, soybeans, and dry bean. Appl Environ Microbiol 56(11):3298-3303.
- Soil Survey Report, 1994. Ministry Of Agriculture, Sultanate of Oman.
- Vincent JM, 1972. A Manual for practical study of root nodule bacteria. IBP Handbook No. 15, Oxford: Blackwell Scientific Publications.