PN-ABD-573
63341

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NITROGEN DYNAMICS OF THE *ACACIA SENEGAL* AGROFORESTRY SYSTEM IN SUDAN

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Progress Report For the
Period January to June 1989

Prepared for the
Agency for International Development
S&T/FNR
Room 515D SA-18
Washington, D.C. 20523

Under
Grant No. DPE-5542-G-SS-7040-00
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Submitted by the
Old Dominion University Research Foundation
P.O.Box 6369
Norfolk, Virginia 23508-0369

July 1989

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Introduction

The first half of this reporting period witnessed the completion of field surveys in Sudan. Civil unrest, wildcat strikes, shortages of goods and services in addition to actual combat operations between the government army units and the SPLA (Sudanese People Liberation Army) close to two of our four research sites were not conducive to scientific field surveys. These conditions lengthened the time required to complete the field work. However, the samples we collected and the data gathered are adequate for us to proceed to the next stage of laboratory investigations and successfull conclusion of our project.

Mr.Saad participated in an international agroforestry symposium held at Washington State University in Pullman Wa.; the symposium theme was "Planning For Agroforestry". He presented a paper entitled " Planning for agroforestry research in Western Sudan: approaches, issues, and lessons learned", which he authered with Dr. Jan C. Noel, director of the International Programs at WSU and Dr. Trent Bunderson, associate professor at WSU. We will present a poster entitled "Effects of soil, seed source and planting density on nodulation and biomass production of *Acacia senegal* in Sudan" at the 40th annual meeting of the American Institute of Biological Sciences to be held at the University of Toronto, Canada in August.

We acknowledge the assistance provided by Dr. George Ghobrial in the USAID mission in Khartoum for writing an introductory letter to the US Embassy who granted Mr. Saad and his wife entry visas to the USA. We also wish to thank Dr. Ahmed A. Mahdi and Mr. Othman Saad in the Faculty of Agriculture, Khartoum University for allowing the use of their laboratory facilities and for accompanying Mr. Saad in his field trips. We also thank Dr. Habbani in the Physics Department for analyzing some of the soil samples using the fast neutron generator technique.

Field surveys

Our field surveys in Singa site were completed in January. The security situation there deteriorated a great deal, but we carried out our work. We collected soil and tissue samples from a water-receiving site on which *Acacia senegal* grew with vigor even late in the dry season. We also sampled two dry sites, one with a natural stand and the

other with an Acacia plantation.

The closure of Khartoum University in March 1989 due to a series of wild cat strikes by all levels of university staff caused some rescheduling of our activities. Drying and grinding of plant tissues, nutrient assays using the neutron generator system, and the inoculation experiments could not be done in Sudan as was previously planned. Samples of soil and tissues were arranged for shipment to the USA for analyses in managable quantities.

Variation among the four study sites in seed size was investigated (Table 1). We chose ten trees from each site at random and collected seeds from each one of them separately. We then took five samples from each tree and determined their weights. We then calculated the number of seed per gram for each site.

No significant difference among sites was detectable using seed size variations. The data were also separated according to soil types (Elobeid and Dubeibat sand against Gedaref and Singa clay). Again the difference with respect to soil type was insignificant. On the same type of soil however, there seems to be a weak trend towards larger seed number per gram i.e. smaller seeds with higher rainfall (Table 2).

This might indicate adaptation of the tree species to dry conditions. Indeed its distribution ceases at higher isohyets.

Our plan to pay a last visit to Elobeid and Dubeibat sites to launch a soil moisture study was aborted by lack of aviation fuel and repeated strikes of Sudan Airways pilots. However, we managed through radictelephone communication to collaborating scientists in WSARP Elobeid station (Dr. Mekki and Mr. Mohammed Mukhtar) to get them to lay out the study as we proposed it. We wish to acknowledge their assistance.

The study investigates the effects of tree cover and temperature on soil moisture relationships through a period of 9 months covering the latter, more difficult part of the dry season, the wet season and first part of the dry season (the driest and wettest times of year). We utilized the gypsom blocks, the Bouyoucos moisture meter and the max., min. and soil thermometers. Daily records of soil moisture, air and soil temperature will be kept by a resident forest overseer stationed in Demokeya forest reserve outside Elobeid.

All field supplies and equipment were provided to Mr. Mohammed Mukhtar, the gum arabic research officer of WSARP.

Laboratory investigations

The seed/soil combination experiment reported previously was concluded. To recapitulate we collected soil and seed from naturally-growing Acacia senegal stands from each of our four sites. We then raised seedlings from each seed type on each soil type, getting a 4 X 4 design of 16 seed/soil combinations. Eight weeks later we dug out the seedlings, counted nodules and dried shoot and root separately at 80 C for 48 hours. We recorded the dry weights. The object of this experiment was to examine the effect of seed sources and soil types on nodulation, germination and biomass production. The soils collected from the four study sites were basically of two types : sand and clay. A T-test based on soil types revealed no significant difference in nodule numbers and germination percentages. Plant biomass was found to be affected by soil types. Duncan Multiple Range Test on nodules showed sites G, S and E are not significantly different with respect to soil types. Site D, however, varies significantly from them, giving lower nodule numbers and biomass. With respect to seed sources Duncan test showed no significant difference between the four sites tested.

We selected the combination that resulted in aoove average nodule formation. These were 3 strains from G site and 3 from S site; both have clay soil. We isolated bacteria strains from each separately in a nutrient agar medium and inoculated seedlings from site E seeds raised on sterile clay. The selected strains are marked (*) in Table 3 above. Our basic plan was to measure the rate of nitrogen fixation using a gas chromatograph. We were unable to locate one while in the field, so we used nodule numbers instead with two assumptions:

1) that small nodules increase in size with time,

2) and that rate of nitrogen fixation is proportional to nodule number.

The effect of seedlig competition on nodulation and biomass production was also investigated (Table 4). Three rates of competition reflected as plant density per unit area were tested :

1) high competition of an average seedling number of about 100, 2) moderate competition of 70, and (3) slight competition of about 6. Each set of seedlings were placed in a clay pot 20 cm in diameter that tapered to 8.0 cm and 20 cm height. The experiment was replicated three times.

There was a strong inverse relationship between plant density and nodulation, but there were minimal effects on total plant biomass. However, competition reduced individual plant biomass significantly.

Field samples were shipped in double polythene bags airfreight to Old Dominion University laboratories where additional lab investigations began. The soil sample types are either sandy or clayey. The former are lose fine sand particles that do not require any further processing prior to chemical analyses. However, the clay soils are cemented into crumby structure that needs to be crushed to prepare them for chemical analyses. Tissue sample processing in the laboratory was started by drying them in a mechanical convection oven at 80 C for 48 hours. The tissue types include leaves, twigs, bark, roots and wood. We collected one sample of thorns and another of root bark to preliminarily investigate their nutrient dynamics. The dried tissues were ground using a Wiley mill.

We will carry out chemical analyses to determine total, nitrate and ammonia nitrogen content in leaves, wood, bark and root tissues. In addition we will determine P, S ,Fe and also pH. We will also analyze soil samples for the above and also Mo.

The genetic variability among the <u>Acacia</u> <u>senegal</u> populations in our study sites is being investigated in the laboratory. We conducted an initial run of gel electrophoresis using germinated seeds from each of the four sites. We wanted to find out whether the system would work with the genetic material available. We placed 10 seeds from each site in a petri plate lined with a moist filter paper, and placed it in an incubator for 4 days. Germinated seeds were crushed in a marked spot plate with an extraction microbuffer. We used Tris citrate (pH 8.0) electrophoresis buffer. We attempted 7 enzyme stain recipes of which the following three were successful:

a) LAP (Leucine aminopeptidase),
b) GOT (Glutamate oxaloacetate transaminase),
c) IDH (Isocitrate dehydrogenase).

Each of them resulted in dotted band formation on the gel surface. Based on the satisfactory results that we observed, we ordered the necessary chemicals and are getting ready to proceed with this analysis.

Table 1

Seed size variation with site.
Number of seed per g.

	Elobeid	Dubeibat	Gedaref	Singa
	9.4	8.6	7.7	10.4
	6.7	10.4	8.1	10.9
	10.4	7.8	9.6	10.0
	7.7	10.5	9.9	9.7
	7.6	10.3	10.2	11.3
	7.5	9.7	7.6	9.6
	7.1	11.0	10.4	8.1
	8.2	11.9	8.8	9.8
	7.9	10.8	7.9	10.8
	8.3	9.2	8.6	8.4
Mean	8.08	10.02	8.88	9.90
S.E.	0.34	0.38	0.33	0.33

Table 2

Variation of seed size with rainfall and soil types

Site		Rainfall (mm)	Soil Type	Seed # / g.
ELOBEID	(E)	300	Sand	8.1
DUBEIBAT	(D)	350	Sand	10.0
GEDAREF	(G)	450	Clay	8.8
SINGA	(S)	500	Clay	9.9

Table 3

Effects of Seed/Soil Combination on Nodulation, Biomass Production and % Germination

SER.	Soil Type	Seed Source	% Germin.	Nodule #	Dry Weight (g) Shoot	Root	Plant
1	ET	EP	100.00	6.3	0.225	0.092	0.317
2	ET	GP	57.14	9.8	0.134	0.092	0.220
3	ET	SP	92.86	6.1	0.211	0.085	0.296
4	ET	DP	50.00	10.4	0.194	0.109	0.303
MEAN			75.00	8.15	0.191	0.094	0.284
S.E.			12.543	1.131	0.020	0.005	0.023
5	GT	EP	71.43	14.0	0.307	0.139	0.446
6	GT	GP	57.14	11.75	0.385	0.202	0.587
7	GT	SP	92.96	13.54	0.305	0.136	0.441
8	GT	DP	71.43	7.10	0.323	0.129	0.452
MEAN			73.25	11.57	0.33	0.151	0.481
S.E.			7.385	1.571	0.019	0.016	0.035
9	ST	EP	71.43	13.60	0.409	0.188	0.597
10	ST	GP	50.00	7.43	0.334	0.150	0.484
11	ST	SP	71.43	10.80	0.296	0.151	0.447
12	ST	DP	42.86	10.50	0.430	0.188	0.618
MEAN			58.93	10.575	0.367	0.169	0.536
S.E.			7.362	1.268	0.031	0.011	0.042
13	DT	EP	78.57	5.73	0.207	0.105	0.312
14	DT	GP	28.57	3.75	0.112	0.060	0.172
15	DT	SP	57.14	4.25	0.216	0.112	0.328
16	DT	DP	50.00	1.86	0.110	0.050	0.160
MEAN			53.57	3.85	0.160	0.032	0.243
S.E.			10.309	0.80	0.029	0.015	0.045

(E= ELOBEID, D= DUBEIBAT, G= GEDAREF, S = SINGA, T= SOIL, P= SEED.)

Table 4
Effect of Competition on Nodulation and Biomass Production

Ser.	Plant #	Shoot Wt	Root Wt	Plant Wt	Nodule #
1	100	5.45	1.42	6.90	0
2	107	5.94	1.32	7.26	0
3	92	5.03	1.54	6.57	0
Mean	99.67	5.47	1.43	6.91	0
S.E	4.3	0.265	0.063	0.199	0
4	70	5.06	1.58	6.64	14
5	67	5.17	1.50	6.67	11
6	65	5.20	1.49	6.69	15
Mean	67.33	5.14	1.52	6.66	13.33
S.E	1.453	0.042	0.028	0.014	1.202
7	9	4.17	1.82	5.99	51
8	4	3.02	1.48	4.50	49
9	6	4.08	1.60	5.68	43
Mean	6.33	3.76	1.63	5.39	47.67
S.E	1.453	0.369	0.099	0.454	2.404