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**N<sub>2</sub> FIXATION, PLANT MINERAL NUTRITION AND C METABOLITES IN  
COWPEA/MAIZE CROPPING SYSTEMS.**

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## ABSTRACT

The objective of this study was (1) quantify N<sub>2</sub> fixation in sole and mixed cropped cowpea, (2) To access nutrient assimilation by component species in the cropping system, and (3) to determine the effects of cropping system on C metabolites.

At harvest the dry mass of both cowpea and maize were highest in sole crops. There was a significant difference in % nitrogen of maize and cowpea plants from the different cropping systems. Total nitrogen content was highest in sole cropped maize and cowpea plants with sole cowpea fixing the most N<sub>2</sub>.  $\delta^{15}\text{N}$  values were lower for the shoots of symbiotic cowpea compared to the maize. Root  $\delta^{15}\text{N}$  values were also generally lower in cowpea than in maize. The %N derived from fixation (Ndfa) was similar for sole, intra and intercropped cowpea. However the amount of N fixed was significantly greater in sole cowpea compared to intra- and intercropped cowpea.

When nutrient uptake and assimilation was assessed in the cropping system, it was found that mineral elements such as Ca, Mg, Cu, Zn and B occurred in significantly greater concentrations in the N<sub>2</sub>- fixing legume compared to the cereal partner.

However the concentration of soluble sugars and starch remained the same for both shoot and roots of cowpea and maize plants in the cropping system.

## **INTRODUCTION**

Africa is under threat due to the widening gap that exists between food production and population growth (Rao and Matuva 2000). Food crop yields have declined over the years as a result of nutrient depletion, This stems from the fact that crops are rarely fertilised, especially when dealing small scale farms. Fertilizer use is low because of unavailability at the right time, high costs as well as risks from erratic and limited rainfall (Rao and Mathuva 2000).

Food legume yields decreased in Africa by 17% between 1979 and 1993; this is high compared to a 4% decrease in South America, and about 157% increase in Europe. Today yields of cereal do not exceed  $1 \text{ t ha}^{-1}$  and legumes  $0.5 \text{ t ha}^{-1}$  per crop season under small farmer conditions (Tiffen et al 1994).

Many factors contribute to the low productivity of African soils, but soil constraints play a major role in limiting crop yields.

African soils are generally infertile and highly heterogeneous. Soil N, P and organic matter are low in most African soils especially those of the Savanna (Dakora and Keya 1997). Soils are also insufficient in nutrients such as N, P, Ca, K and S. Of all the nutrients in the soil, N is the major limiting factor in food production worldwide.

The use of forage legumes in cropping systems, either as intercrops or rotations has become a well-known practice in improving soil fertility.

Biological nitrogen fixation by rhizobial bacteria in legume root nodules plays a major role in maintaining soil fertility and productivity in low input farming systems in addition to their ability to capture organic matter into the soil (Wortmann et al 2000).

Intercropping and rotational cropping are practiced in many parts of the world (Francis 1986), including the tropics (Olasanta et al 1996) and temperate environments (Li et al 1999). In Africa, these cropping systems are very common, though scientifically not well understood.

There are many benefits to mixed cropping systems (sole cropping, intercropping and intracropping). These farming systems contribute to weed suppression (Manyon et al 1996, Oswald et al 2002). The intercropped legume provides soil cover and reduces soil and water loss (Wortmann et al 2000, Olasantan et al 1996). They ensure good ground cover and increase infiltration of rainwater; this also increases earthworm activity (Lal 1983) and reduces soil temperatures. Not only does the farmer make profits from one crop but from two thus increasing returns and reducing risks.

Small-scale farmers have also been known to practice mixed crop and livestock systems (Rao and Matuva 2000). In this case not only do they generate cash income through sale of livestock but also improve soil chemical, physical and biological properties when animal manure is added to the soil (Haque et al 1995).

Perennial tree legumes have greater scope to replenish soil fertility than annual grain legumes due to their ability to exploit residual water and subsoil nutrients. Their all year round growth may also leads to higher N<sub>2</sub> fixation (Giller et al 1997).

There are many constraints to symbiotic N<sub>2</sub> fixation by legumes in Africa. The extent of nodulation and nitrogen fixation is constrained by mineral nutrition, genetic traits and a host of environmental factors. These factors have an effect on both the plant and the microsymbiont. Drought, nutrient imbalance, soil heterogeneity, extreme temperature and humidity all affect legumes differently. Studies in the West African savanna show that sole cropped legumes can contribute significantly to cereal crop yields (Dakora et al 1997). Grain yields of maize following sole cropped legumes were 2 fold higher than under intercropped conditions.

Compared with corresponding sole crops, yield advantages have been recorded in many intercrop systems including maize/soybean (West and Griffith 1992), sorghum / soybean (Elmore and Jackobs 1986) and maize/ lucerne (Smith and Confer 1998). A 26% increase in maize yield was accompanied by a 27% yield reduction of soybean in a maize/soybean intercrop (West and Griffith 1992). Studies in semi arid Kenya on maize intercropped with cowpea (Rao and Mathuva 2000) show that maize can remove as much as 25 to 40 N ha<sup>-1</sup> per season. Cowpea nodulated with *Bradyrhizobium* species could fix up to 200kg N ha<sup>-1</sup> (Pilbeam et al 1995).

There is a close relationship between yield increase and nutrient uptake by intercropped species (Mortis and Garnity 1993). These cropping systems can have a significant effect on total nutrient uptake (Olasantun et al 1996). Because roots of the

legume and non-legume partner can get interwoven belowground a high interspecific competition can develop between cereal and legume plants leading to differential nutrient uptake and increased N transfer from cereal to legume.

This study addresses three main questions:

1. What is the amount of N fixed in cowpea grown as sole or intra- and intercropped with maize
2. What is the effect of cropping system on nutrient uptake by legumes and cereals?
3. How does sole, intra- and intercropping affect the level of photosynthetic C stored as soluble sugars or starch?

## **MATERIALS AND METHODSD**

### DESCRIPTION OF EXPERIMENTAL SITE

Field experiments were conducted at Nietvoorbij Farm (33° 54 'S, 18° 14'E) in Stellenbosh, Western Cape region. The site lies in the winter rainfall region of the Western Cape at an elevation of 146 m above sea level. The mean annual rainfall on the farm is 713.4 mm and mean annual temperatures range from 22.6 °C during the day to 11.6 °C during the night (Willem Lansbscher).

The soils are Glenrosa. At the experimental site the 0-400 mm soil layer was found to be fine sandy loam in texture (19% clay, 13% silt and 68% fine sandy loam). At depths of 400-900 mm below ground, the soil texture is described as clay loam and contains 35% clay, 17% silt and 48% clay loam.

The experimental site is used to cultivate grapes, used commercially in the making of white and red wine.

## **EXPERIMENTAL TREATMENTS**

The study evaluated four cropping systems as treatments

1. Cowpea grown alone
2. Maize grown alone
3. Maize intracropped with cowpea
4. Maize intercropped with cowpea

There were 4 replicates of each treatment or cropping system.

The sole maize plants were grown with 40 cm spacing within the row and 90 cm spacing between the rows. Plant density of maize was 55555 per hectare.

Sole cowpea plants were grown with spacing of 20 cm within the row and 60 cm spacing between the rows. Cowpea plant density was 166666 per hectare.

The cowpea in maize/cowpea intercrop was spaced at 20 cm within the row and 90 cm between the rows (or 45 cm between maize rows). The density of cowpea in these treatments was 11111 per hectare. The spacing of the maize plants was 40 cm within the row and 90 between the rows like sole maize treatments. Thus the density of the maize plants was the same as that of the sole maize.

Intracropped treatments followed the same spacing as intercropped treatments.

However instead of having sole crop within the row we had both cowpea and maize.

The cowpea in maize/cowpea intercrop was also spaced at 20 cm within the row and 90 cm between the rows (or 45 cm between maize rows).

## **DATA COLLECTION AND DATA ANALYSIS**

### **Protocol For Metabolite Analysis**

#### **Sugar**

Use 0.1g of the sample

Add 10ml of 80% ethanol

Incubate at 0°C for three days 72 hours to be exact

Centrifuge

Dilute the supernatant to 25ml with 80% ethanol

Preparation of sample for spectrophotometer determination

Add the following to a test-tube:

0.5 ml sample

1ml of 28% phenol

5ml of H<sub>2</sub>SO<sub>4</sub>

Mix the content of the tube by vortex and allow to stand for 15 minutes before taking readings on the spectrophotometer.

Include the blank:

0.5 ml distilled water

1ml phenol

5 ml H<sub>2</sub>SO<sub>4</sub>

Readings were done within an hour with absorbance set at 490 nm

### Starch:

- Dry the sample pellet from sugar extract
- Add 10 ml of 32% HCl
- Put in a boiling water bath for 3 hours
- Centrifuge
- Prepare sample and for spectrophotometric determination as for sugars

### Standards:

Run a set of standards with glucose concentrations in the range of 20-80  $\mu\text{g/ml}$

Glucose stock solution: 0.1 g/L

20 ml stock solution dilute to 100ml with distilled water:	20 $\mu\text{g/ml}$
30 ml	: 30 $\mu\text{g/ml}$
40 ml	: 40 $\mu\text{g/ml}$
50 ml	: 50 $\mu\text{g/ml}$
60 ml	: 60 $\mu\text{g/ml}$
70 ml	: 70 $\mu\text{g/ml}$
80 ml	: 80 $\mu\text{g/ml}$

### HARVESTING

Plant were harvested mid May 2002. The plant materials were separated into shoots and roots oven dried at 50°C, weighed, and ground to fine powder for  $^{15}\text{N}$  isotope analysis.

### <sup>15</sup>N Isotope Analysis And Estimation of N fixed

Weighted samples were run on a mass spectrophotometer (CHN analyser model NA 1500 NC) connected with Conflo device MAT 252, from this we obtained <sup>15</sup>N/<sup>14</sup>N ratios. After every five samples 0.5 mg of gelatin (a nitrogenous compound) was included as standard to assess the precision of the mass spectrophotometer.

From the <sup>15</sup>N/<sup>14</sup>N and % N values obtained for legume and reference plant, nitrogen content in shoots and roots was calculated as well as total nitrogen derived from fixation (Ndfa). Ndfa was calculated using the equation of Bergersen and Turner (1983):

$$\text{Ndfa} = \frac{\delta^{15}\text{N}_{\text{ref}} - \delta^{15}\text{N}_{\text{leg}}}{\delta^{15}\text{N}_{\text{ref}} - B}$$

Samples were sent the Department of Agriculture in Stellenbosh for macro and micro nutrient analysis. Macronutrient analysis was done for the elements P, Ca, K, Mg and Na. Micronutrient analysis included elements such as Fe, Cu, Zn, Mn, Al and B.

Starch and sugar analyses were done in the lab at the University of Cape Town using the sugar and starch protocols described above.

A program called Statistica was used for statistical analysis, factorial ANOVA was the analysis used and quick specs dialog the method used.

## RESULTS

At harvest total plant biomass was significantly ( $p \leq 0.05$ ) higher in sole maize compared to intercropped and intracropped maize (Figure 1). Similarly, sole cowpea plants showed significantly ( $p \leq 0.05$ ) greater growth and biomass compared to intercropped and intracropped counterparts (Figure 2).

The concentration of N in tissues differed with cropping systems. The shoots of sole maize had significantly ( $p \leq 0.05$ ) higher % N compared to intercropped and intracropped maize (Table 1). With cowpea, there were no significant differences in shoot % N in relation to cropping system. However the % N of cowpea shoots were markedly greater than those of mixed crop maize cereal. With roots, there was little difference in % N between cropping systems, except for intracropped cowpea, which was significantly lower than sole or intercropped counterparts.

The  $\delta^{15}\text{N}$  values of maize shoots were significantly ( $p \leq 0.05$ ) higher than those of  $\text{N}_2$ -fixing cowpea. The high values clearly indicate that the cereal depended on soil N for its requirements. The  $\delta^{15}\text{N}$  of roots was significantly different for maize, with the intracropped being markedly lower than sole and intercrop. The roots of cowpea also showed lower  $\delta^{15}\text{N}$  values compared to the maize cereal (Table 2).

Total N content per plant was significantly ( $p \leq 0.05$ ) higher in sole maize compared to intercropped and intracropped maize (Figure 5). Similarly sole cowpea plants showed significantly ( $p \leq 0.05$ ) greater total nitrogen content compared to intercropped and intracropped counterparts (Figure 6).

There was no significant difference ( $p \leq 0.05$ ) in percent nitrogen derived from fixation, by cowpea plants in relation to cropping systems (Figure 3). However fixed nitrogen was significantly ( $p \leq 0.05$ ) higher in sole cowpea compared to intercropped and intracropped cowpea (Figure 4).

Nutrients concentrations varied within cropping systems depending on the type of nutrient, crop type and cropping system. Calcium levels were significantly ( $p \leq 0.05$ ) higher in  $N_2$  fixing cowpea compared to the reference plant. However these were not significantly different in cowpea exposed to different cropping systems. Maize calcium levels were also not significantly different ( $p \leq 0.05$ ) when exposed to different cropping systems (Figure 7). Similarly with roots, calcium levels were significantly ( $p \leq 0.05$ ) higher in cowpea compared to reference plant. Root calcium levels were also significantly ( $p \leq 0.05$ ) higher in cowpea intracropped compared to sole and intercropped cowpea. With maize there was no significant ( $p \geq 0.05$ ) difference in calcium levels in relation to cropping system.

Potassium levels were significantly higher in the maize treatments compared to those of the  $N_2$  fixing cowpea. There was no significant difference in shoot potassium levels of maize plants in relation to cropping system. Intracropped cowpea showed significantly less potassium than sole cropped and intercropped cowpea (Figure 7).

With roots cowpea had significantly ( $p \leq 0.05$ ) higher levels of root potassium levels than maize. Sole cowpea was significantly ( $p \leq 0.05$ ) lower in potassium levels than intracropped cowpea, but similar ( $p \geq 0.05$ ) to intercropped cowpea. There was no difference ( $p \geq 0.05$ ) in root potassium levels of maize plants (Figure 7).

Magnesium shoot levels were significantly ( $p \leq 0.05$ ) higher in cowpea treatments compared to the maize treatments, however there was no significant ( $p \geq 0.05$ ) difference in magnesium levels of cowpea in relation to cropping system. Similarly there was no significant difference in maize magnesium levels. Magnesium levels in roots were significantly ( $p \leq 0.05$ ) higher in  $N_2$  fixing cowpea than the maize reference plant. There was no significant ( $p \leq 0.05$ ) difference in root magnesium levels in maize plants exposed to different crop systems. Cowpea intracropped had significantly ( $p \leq 0.05$ ) higher magnesium root levels than sole cropped and intercropped cowpea counterparts (Figure 7).

However intracropped cowpea had significantly ( $p \leq 0.05$ ) higher phosphorus levels compared to sole cowpea and was similar ( $p \geq 0.05$ ) to intercropped cowpea cropping system. Shoot phosphorus levels were the same ( $p \geq 0.05$ ) in maize cropped system. With roots phosphorus levels were significantly ( $p \leq 0.05$ ) higher in intercropped maize compared to sole maize and intracropped maize counterparts. There was no significant difference in cowpea root phosphorus levels (Figure 7).

Shoot sodium levels were significantly ( $p \leq 0.05$ ) higher in maize compared to the  $N_2$  fixing legume. Sodium levels were not significantly ( $p \geq 0.05$ ) different in both

cowpea and maize cropped system. Similarly in root, sodium levels were significantly ( $p \leq 0.05$ ) higher in cowpea compared to maize. Sodium levels were also not significantly ( $p \geq 0.05$ ) different in both cowpea and maize cropped system (Figure 7).

Copper levels in the shoots were significantly ( $p \leq 0.05$ ) higher in cowpea than in the reference plant. However there was no significant difference in shoot copper levels in both cowpea and maize cropped systems. With roots copper levels were similar across all treatments (cowpea and maize cropped systems) except in sole cowpea, which was significantly ( $p \leq 0.05$ ) lower than the intracropped and intercropped counterparts (Figure 8).

Zinc shoot levels were also significantly ( $p \leq 0.05$ ) higher in the  $N_2$  fixing cowpea compared to the reference plant. There was no significant difference ( $p \geq 0.05$ ) in zinc shoot levels in cowpea cropped systems as well as maize cropped systems. Similarly root zinc levels were markedly higher in cowpea compared to maize. There was no significant difference in root zinc levels in relation maize cropping systems.

Intracropped cowpea was significantly ( $p \leq 0.05$ ) higher in zinc levels than sole cowpea and similar to cowpea cropped system (Figure 8).

Shoot boron levels were significantly ( $p \leq 0.05$ ) higher in cowpea compared to maize plants. There was no significant difference ( $p \geq 0.05$ ) in shoot boron levels in both cowpea and maize cropped systems. Root boron levels showed similar patterns, there was no significant difference ( $p \geq 0.05$ ) in shoot boron levels, in both cowpea and maize cropped systems. Similarly root boron levels were significantly ( $p \leq 0.05$ ) higher in cowpea compared to maize cropped systems (Figure 8).

There was no defined pattern in shoot iron levels, when comparing maize and cowpea cropped systems. In maize cropped systems intercropped maize had significantly ( $p \leq 0.05$ ) higher iron levels compared to the sole maize and intracropped maize counterparts, which were similar ( $p \geq 0.05$ ). Sole cowpea shoot iron levels were significantly ( $p \leq 0.05$ ) higher than intracropped iron levels, however levels were similar to those of intercropped cowpea. Iron levels in maize root were significantly ( $p \leq 0.05$ ) higher compared to the  $N_2$  fixing cowpea. However there was no significant ( $p \geq 0.05$ ) difference in maize cropped systems. Intercropped cowpea was significantly ( $p \leq 0.05$ ) higher compared to intercropped and intracropped cowpea.

There was also no defined pattern when comparing manganese level in cowpea and maize shoots. There was no significant difference ( $p \geq 0.05$ ) in manganese shoot levels in both maize and cowpea cropped system. There was no significant ( $p \geq 0.05$ ) difference in manganese root levels in all cropped systems (including both cowpea and maize cropped systems), except in sole cowpea, significantly ( $p \leq 0.05$ ) lower than the other cropped system (Figure 8).

Aluminium also showed no defined pattern when comparing maize and cowpea shoots. Sole maize aluminium levels were significantly ( $p \leq 0.05$ ) lower compared to intercropped and intracropped maize. Intercropped cowpea had significantly ( $p \leq 0.05$ ) higher shoot aluminium levels than the sole maize and intracropped maize counterparts. There was no significant difference ( $p \geq 0.05$ ) in root aluminium levels in relation to maize cropping system. Intercropped cowpea had significantly ( $p \leq 0.05$ )

higher levels of root aluminium compared to the sole cowpea and intracropped counterparts (Figure 8).

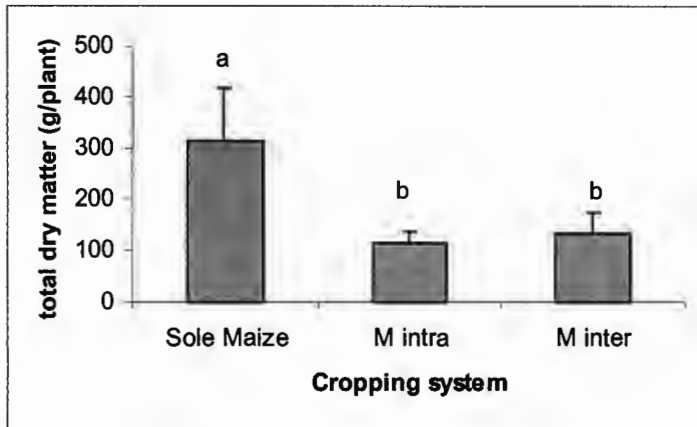
The accumulation of soluble sugars and starch remained the same for both shoots and roots cowpea and maize plants in cropping system (Figures 9, 10, 11 and 12). There was no significant difference in all cropping system.

**Table 1:** Nitrogen concentration (%) in shoots and roots of cowpea and maize plants. Values followed by dissimilar letters are significantly different at  $p \leq 0.05$ .

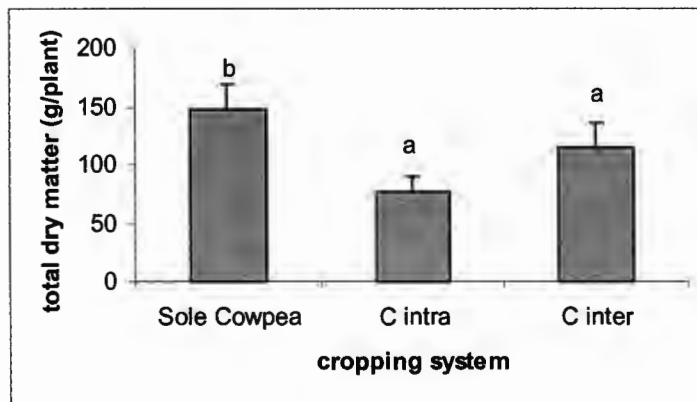
Treatment	Shoot (%)	Root (%)
<b>Maize</b>		
Sole maize	2.33 ± 0.40a	0.77 ± 0.3a
Maize intracropped	1.76 ± 0.40b	1.39 ± 0.66ab
Maize intercropped	1.80 ± 0.26b	1.16 ± 0.50ab
<b>Cowpea</b>		
Sole cowpea	2.56 ± 0.41a	1.98 ± 0.36b
Cowpea intracropped	2.11 ± 0.12ab	0.85 ± 0.10a
Cowpea intercropped	2.37 ± 0.32a	1.81 ± 0.90b

**Table 2:**  $\delta^{15} \text{N}$  values of shoots and roots of cowpea and maize plants. Values followed by dissimilar letters are significantly different at  $p \leq 0.05$

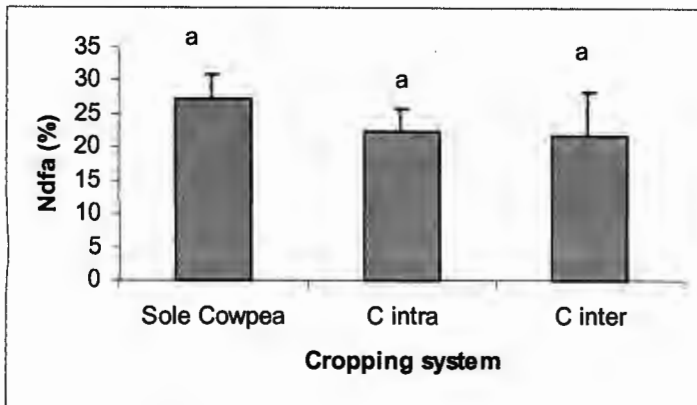
Treatment	$\delta^{15} \text{N}$ Shoot ‰	$\delta^{15} \text{N}$ Roots ‰
<b>Maize</b>		
Sole Maize	3.84±0.72a	1.99±0.34b
Maize intracropped	3.73±0.24a	1.06±0.366b
Maize intercropped	3.65±1.14a	1.65±0.48b
<b>Cowpea</b>		
Sole Cowpea	2.01±0.01b	1.32±0.22b
Cowpea intracropped	1.47±0.69b	0.97± 0.81b
Cowpea intercropped	2.01±0.97b	0.90±0.72b



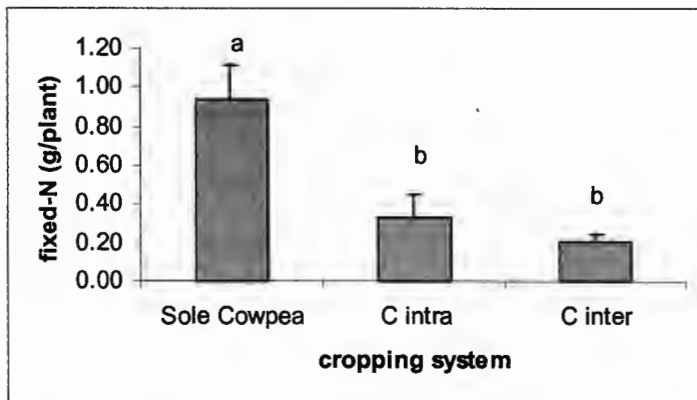
**Figure 1:** Dry matter of maize plants. Values followed by dissimilar letters are significantly different at  $p \leq 0.05$ .



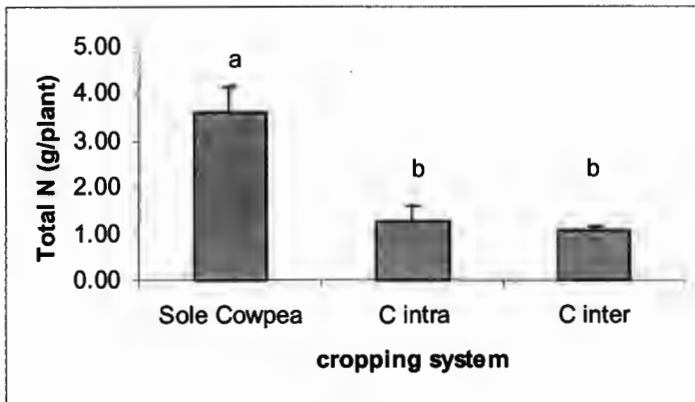
**Figure 2:** Dry matter of cowpea plants. Values followed by dissimilar letters are significantly different at  $p \leq 0.05$ .



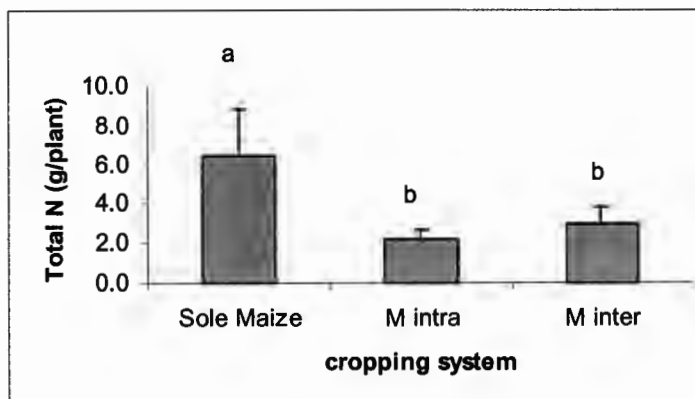
**Figure 3:** Percent nitrogen derived from fixation by cowpea plants. Values followed by the same letters are not significantly different at  $p \geq 0.05$ .



**Figure 4:** Fixed nitrogen in cowpea plants. Values followed by dissimilar letters are significantly different at  $p \leq 0.05$ .



**Figure 5:** Total nitrogen content of cowpea plants. Values followed by dissimilar letters are significantly different at  $p \leq 0.05$ .



**Figure 6:** Total nitrogen content of maize plants. Values followed by dissimilar plants are significantly different at  $p \leq 0.05$ .

**Figure 7:** Concentrations of macronutrient elements in cowpea and maize plant parts.

Treatment	Macronutrient Level									
	P		Ca		K		Mg		Na	
	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root
					(%)					
					(mg/kg)					
<b>Maize</b>										
Maize	0.41±0.04 <b>ab</b>	0.08±0.02 <b>a</b>	0.55±0.08 <b>a</b>	0.20±0.00 <b>a</b>	3.91±0.14 <b>a</b>	0.90±0.28 <b>a</b>	0.16±0.12 <b>a</b>	0.06±0.01 <b>a</b>	632.00±308.30 <b>a</b>	597.50±308.30 <b>ab</b>
Maize intra	0.36±0.12 <b>ab</b>	0.11±0.0 <b>ab</b>	0.52±0.11 <b>a</b>	0.21±0.03 <b>a</b>	3.76±1.13 <b>a</b>	1.13±0.36 <b>a</b>	0.29±0.05 <b>a</b>	0.10±0.04 <b>ad</b>	342.33±204.59 <b>abc</b>	1011.67± 232.20 <b>a</b>
Maize inter	0.28±0.06 <b>a</b>	0.32±0.25 <b>b</b>	0.56±0.01 <b>a</b>	0.21±0.04 <b>a</b>	3.97±0.08 <b>a</b>	1.03±0.51 <b>a</b>	0.28±0.01 <b>a</b>	0.10±0.03 <b>ad</b>	569.50±20.51 <b>ac</b>	1060.00±448.31 <b>a</b>
<b>Cowpea</b>										
Cowpea intra	0.45±0.01 <b>b</b>	0.30±0.03 <b>b</b>	2.86±0.66 <b>b</b>	1.93±1.27 <b>c</b>	1.66±0.15 <b>b</b>	2.31±0.15 <b>b</b>	0.49±0.05 <b>b</b>	0.51±0.04 <b>b</b>	191.50±36.06 <b>b</b>	208.50±45.96 <b>b</b>
Cowpea inter	0.42±0.06 <b>ab</b>	0.28±0.09 <b>b</b>	2.94±0.18 <b>b</b>	0.94±1.05 <b>b</b>	1.74±0.15 <b>c</b>	1.54±0.25 <b>ab</b>	0.50±0.08 <b>b</b>	0.27±0.09 <b>c</b>	171.33±24.21 <b>ab</b>	551.67±298.22 <b>b</b>
Sole Cowpea	0.27±0.01 <b>a</b>	0.23±0.01 <b>b</b>	2.38±0.87 <b>b</b>	1.00±1.15 <b>b</b>	1.77±0.28 <b>c</b>	0.83±0.88 <b>a</b>	0.42±0.01 <b>b</b>	0.20±0.00 <b>cd</b>	230.50±4.95 <b>b</b>	289.50±3.54 <b>bc</b>

**Figure 8:** Concentration of micronutrient elements in cowpea and maize plants parts.

Treatment	Micronutrients level (mg/kg)											
	Fe		Cu		Zn		Mn		Al		B	
	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root
<b>Maize</b>												
Maize	258.85±55.79a	5595.0±141.4a	10.8±0.2a	12.57±0.15a	43.47±1.27a	15.52±6.92a	13.17±0.62a	18.30±1.51a	268.80±106.07a	9570.0±106.07a	8.06±0.4a	10.28±0.05a
Maize Intra	395.20±79.24 ac	4472.88±1155.65a	11.9±1.0a	12.85±2.39a	36.40±10.86a	35.01±17.64ab	18.18±4.10ab	18.72±2.98a	464.23±146.35b	8246.67±523.08a	13.32±11.22a	9.80±0.98a
Maize Inter	1193.40±401.07b	4137.50±989.2a	14.2±1.1a	19.84±8.27a	54.77±13.28ab	48.56±30.90ab	11.78±0.16a	16.59±1.29a	581.75±502.26b	9580.0±360.62a	10.26±1.42a	11.21±0.22a
<b>Cowpea</b>												
Cowpea intra	442.90±97.86ac	376.48±180.91b	21.30±0.4b	15.99±8.39a	63.59±17.56ab	77.14±17.22b	34.55±19.92b	17.65±10.54a	579.80±34.01b	463.66±191.82b	50.45±14.3b	36.74±11.04c
Cowpea inter	653.77±45.900cd	1438.5±626.23c	24.5±4.b	16.46±9.37a	82.10±20.08b	49.28±23.9ab	21.17±4.08ab	10.02±4.90a	1034±144.25c	3219.3±1869.37c	45.07±3.52b	28.03±7.3b
Sole cowpea	911.60±22.06bd	493.90±60.65b	67.77±10.2c	9.74±0.78b	64.24±1.32ab	14.52±0.16a	16.87±3.07ab	3.70±0.94b	430±191.80b	922.95±34.01b	42.14±1.56b	18.20±2.5a

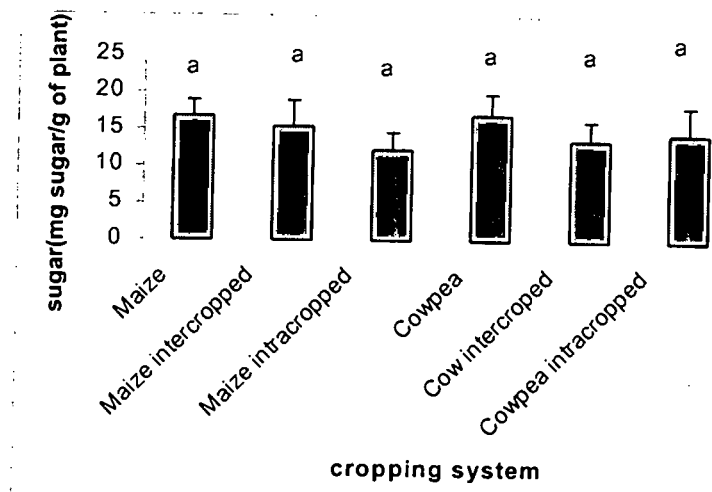


Figure 9: Amount of sugar in shoot of maize and cowpea plants. Values followed by similar letters are similar at  $p \leq 0.05$ .

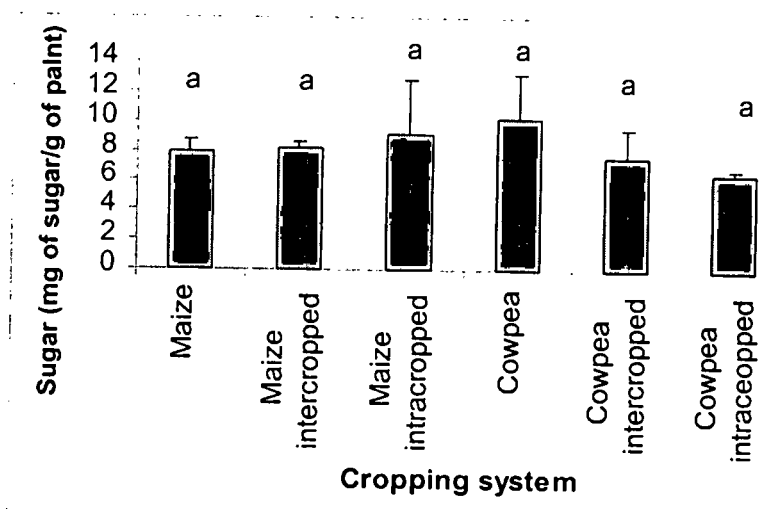


Figure 10: Amount of sugars in roots of maize and cowpea plants. Values followed by similar numbers are similar at  $p \leq 0.05$ .

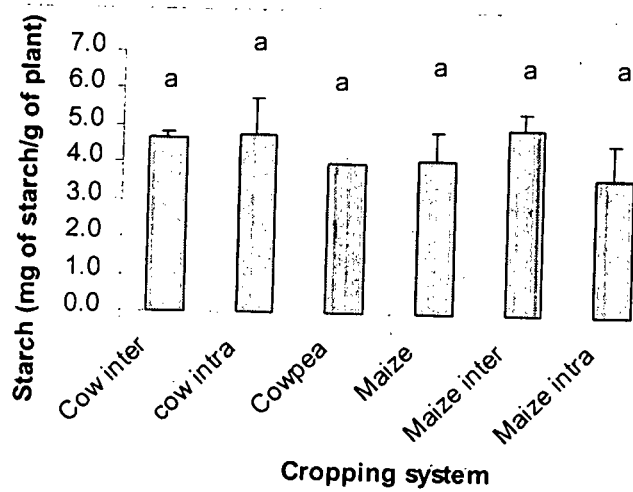


Figure 11: Amount of starch in shoots of cowpea and maize plants. Values followed by similar numbers are similar at  $p \leq 0.05$ .

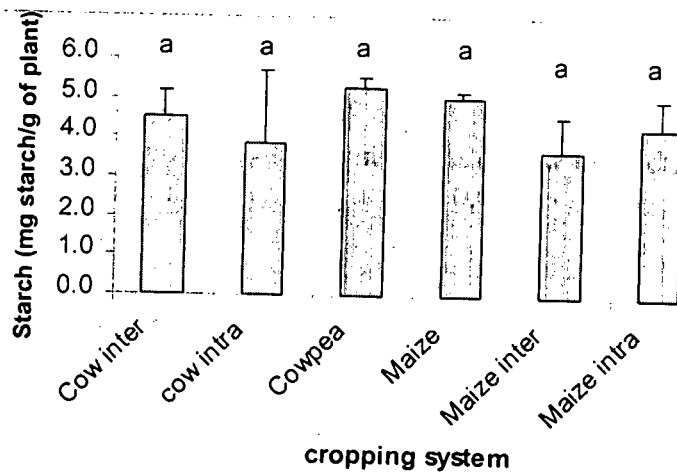


Figure 12: Amount of starch in shoots of cowpea and maize plants. Values followed by similar numbers are similar at  $p \leq 0.05$ .

## DISCUSSION

Greater nitrogen acquisition by non-legume crops intercropped with legumes is frequently reported in literature (Francis 1986, Vandermeer 1989, Stern 1993). This is usually attributed to two factors. Firstly the difference in competitive abilities of the component species, which causes the none-legume to be more competitive thus taking in more nitrogen from the soil (Li et al 1986). Secondly the transfer of nitrogen from the legume to the none- legume thus increasing % nitrogen in the none-legume.

Trade off between increasing the yield of dominant species and decreasing that of the dominated specie has three outcomes for intercropping systems, yield advantage, yield disadvantage and the intermediate results (Vadermeer 1989). Although these results are not analysing yield, dry mass followed Vandermeer's second principal of yield disadvantage, intercropping and intracropping significantly reduced maize and cowpea dry mass.

These results show that total dry-mass of cowpea and maize plants were significantly reduced in intercropping and intercropping. Nitrogen acquisition by maize intercropped/intracropped with cowpea was significantly decreased when compared with sole maize. However nitrogen acquisition by cowpea was not significantly affected by intercropping and intracropping with maize crops. The results suggest that neither cowpea nor maize benefited from intercropping, in terms of nitrogen acquisition.

This is probably due to the fact that when grown together, maize and cowpea compete for the same natural resources thus negatively affecting each other in terms of growth

and nitrogen acquisition. Contrary to studies by Li et al (2001) on wheat and soybean crops and Callaway (1998) on *Abies lasiocarpa* and *Pinus albicaulis*, this study shows that interspecific and intraspecific crop facilitation did not occur.

Interspecific competition is the interaction of two species that reduces the fitness of one or both species (Crawley 1997). Vandermeer (1989) stated that when two plants are grown in close proximity, basic physiological principle suggest that they will always compete whether or not facilitation is operating

There was no significant different ( $p \geq 0.05$ ) in  $\delta^{15}\text{N}$  values within cowpea, treatments. Cowpea in this study was not affected by the different cropping systems, thus it acquired the same % nitrogen across all treatments.  $\delta^{15}\text{N}$  values for maize are also not significantly different, concluding that maize did not benefit from the nitrogen-fixing legume.

Total nitrogen content in sole cowpea was significantly different ( $p \leq 0.05$ ) to intercropped and intracropped cowpea, showing that total nitrogen acquired was much higher in sole crop than both intercrop and intracrop. This was also the case for the maize cropping systems. This again implies that intercropping and intracropping maize and cowpea imposes no benefits to the crops.

Further evidence is presented by data from nitrogen derived from fixation (Figure 3), indicating no significant difference in Ndfa in cowpea plants exposed to different cropping systems.

Fixed nitrogen (g/plant) was significantly larger in sole cowpea cropping systems compared to intercropped cowpea and intracropped cowpea (Figure 4). This shows cowpea fixes more nitrogen grown alone, then when intercropped or intracropped.

Competition for nutrients between component species in intercropping and intra systems often occurs (Mason et al 1986), first for the mobile resources such as water and nitrogen and then for all other soil nutrients. When nutrient uptake and assimilation was assessed in the cropping system, it was found that mineral elements such as Ca, Mg, Cu, Zn and B occurred in significantly greater concentrations in the N<sub>2</sub>-fixing legume compared to the cereal partner. Contrary to Li et al (2001) the overall results show that maize nutrient uptake was not affected by different cropping systems. In this study, intercropping and intracropping maize with N<sub>2</sub> fixing cowpea did not lead to increased nutrient uptake of the maize plants.

Cropping systems also had no significant effect on the levels of photosynthetic carbon stored as soluble sugars and starch i.e starch and sugar levels were insignificantly different ( $p \geq 0.05$ ) across cropped maize and cowpea plants.

## CONCLUSION

Intercropping and intracropping cowpea with maize had a significant effect on maize shoot and root % nitrogen however it did not have significant effects on  $\delta^{15}\text{N}$  values. Nitrogen fixation by cowpea in this study is neither facilitated nor negatively affected by maize.

Sole maize acquired more shoot nitrogen than mixed crop systems, however it also acquired the least root % nitrogen. Overall (shoots and roots) there was no significant difference in nitrogen acquired by maize plants exposed to different treatments.

Both cowpea and maize grew better separately than when grown together.

Cowpea/maize intracropped and intercropped systems had the least growth due to the extreme competition for nutrients that exist between the two component species.

Total nitrogen content was highest in sole crops suggesting both cowpea and maize do better (by acquiring more nitrogen) when grown as sole crops.

There was no significant difference in nitrogen derived from fixation in cowpea exposed to different cropping systems. Sole cowpea fixed the most nitrogen compared to cowpea grown as mixed crop systems.

Macronutrient and micronutrient acquisition varied within cropping systems, some showing least depletion in sole crops, others no significant differences in cropping systems. However cowpea had significantly more macro and micronutrients compared to the maize counterpart. This is probably because legumes require extra nutrients for

the symbiotic process. The implications are that increased legume cultivation followed by residue removal can lead to nutrient depletion.

Competition seems to play a huge role in cowpea grown with maize. In this study maize (non legume) plants did not benefit from cowpea (legume), due to the high competitive nature of these component species when grown together. Competition can be minimised in intercropping systems by selecting species with different rooting patterns, different nutrient requirements (Willey 1979), proper plant spacing (Dalal 1974) and different timing of peak demand for nutrients.

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