Nitrogen fixation and yield of two cowpea varieties under legume-cereal cropping sequence

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Field experiment was conducted to determine the biological nitrogen fixation and yields of two cowpea varieties, IT90K-277-2 and Oloyin, under legume-cereal cropping sequence. The experimental design was Randomized Complete Block design (RCBD) with three replicates. Data were collected on growth, and yield parameters which include number of nodules, nodule dry weight, percentage N derived from atmosphere (% Ndfa), grain yield, chlorophyll content, shoot dry weight and total N fixed. Result showed significant varietal differences ($P<0.05$) in plant height and number of leaves between the two varieties with Oloyin having taller plant height and higher number of leaves. There was no significant difference between the two varieties in the number of nodules, nodule dry weight, percentage N derived from the atmosphere (% Ndfa) and total N fixed (kg/ha). The percent Ndfa for the two varieties was relatively high; IT90K-277-2 had 79% while Oloyin had 77%. The shoot dry weight and grain yield of the two varieties were also not significantly different. Based on the results, IT90K-277-2 and Oloyin had similar potential for biological N fixation and grain yield production and can be used as food security item in Nigeria.

Key words: IT90K-277-2, Oloyin, legume-cereal, cropping system

INTRODUCTION

Nitrogen is a nutrient that is important to plant and without it, the plants will suffer. This nutrient occurs in a variety of forms and it is affected by chemical and biological processes that can change its form and can transfer it to or from water, soil, biological organisms, and the atmosphere (Mueller and Helsel, 1996). It is the nutrient that is most commonly deficient in soils, contributing to reduced agricultural yields throughout the world. Nitrogen can be supplied to crops by biological nitrogen fixation (BNF), a process which is becoming more important for not only reducing energy costs, but also in seeking more sustainable agricultural production. Nitrogen fixing micro-organisms could therefore be an important component of sustainable agricultural systems. It is utilized by plants to synthesize amino acids, the building blocks of proteins. Nitrogen is also required by plants for other vital compounds, such as chlorophyll, nucleic acids, and enzymes. Chlorosis is usually more pronounced in older tissue. Since nitrogen is mobile within plants, it tends to move from older to younger tissue when in short supply.

Legume cover crops can supply N to succeeding crops and increase crop yields compared with non-legume or no cover crops (Hargrove, 1986; Clark et al., 1994, Kuo et al., 1997b). In contrast, non-legume cover crops are effective in increasing soil organic matter by supplying C through increased biomass production (Kuo et al., 1997a, 1997b; Sainju et al., 2000) compared with legume or no cover crops. Non legume cover crops also reduce NO_3–N leaching from the soil profile better than legume or no cover crops do (Meisinger et al., 1991; McCracken et al., 1994). A mixture of legume and non-legume cover crops would be ideal to supply both C and N inputs inadequate amounts that help to improve soil quality and reduce N leaching compared with legumes and increase crop productivity compared with non-legumes. As legumes and non-legumes are grown together, N is transferred from legumes to non-legumes, thereby resulting in better N nutrition of non-legumes and increasing the herbage and protein yields of the biculture crops (Ta and Faris, 1987; Russelle and Hargrove, 1989).

Biological N_2 fixation (BNF) and mineral soil or fertilizers N are the main sources of meeting the N requirement of high yielding cowpea. However, antagonism between nitrate concentration in the soil solution and the N_2 fixation process in the nodules is the main constraint the crop faces in terms of increasing N uptake (Streeter, 1988) when no other abiotic stress that reduce BNF activity occurs, e.g. soil moisture (Purcell et al., 2004), soil pH (Parker and Harris, 1977) or soil temperature. If the overall N supply does not meet soybean requirements, the crop will remobilize N accumulated in leaves to the grain, which diminishes the photosynthetic capacity of the canopy and thus limits yield potential. Van Kessel and Hartley (2000) suggested that N_2 fixation will increase in high-yielding environments since the nitrogenase, located in the nodules, will adjust its activity to the demand of the legume (Mengel, 1994). Since nitrogen is one of the limiting elements in the soil, as a legume, cowpea fixes its own nitrogen, and does not need nitrogen fertilizer. In fact, some of the vigorous, vining, varieties of cowpeas are excellent nitrogen producers as a cover crop. Seed should be inoculated with the appropriate *Rhizobium* species for optimum nitrogen fixation, but nodules will generally form on cowpeas.

Legume-cereal cropping sequence is an important agricultural system that is popular among smallholder farmers. There are different reasons for varietal preference in legume-cereal cropping system and one of it is the ability of legume to minimize the cost incurred on N fertilizer for the next crop. Information is lacking on the contribution of some legume varieties to the soil N and productivity in some crop research in FUNAAB. Hence, this study aims to determine the nitrogen fixation of the local and improved varieties and the yield of Oloyin and IT90K-277-2 under legume-cereal cropping sequence.

MATERIALS AND METHODS

Experimental site

The experiment was carried out at the Directorate of University Farms (DUFARMS) at the Federal University of Agriculture Abeokuta (FUNAAB), Ogun State. The geographical location lies in the South-Western Nigeria (Latitude 7° 15' N and Longitude 3° 28' E). The altitude was 76 m above sea level with a prevailing tropical climate and a mean annual rainfall above 1300 mm. The soil classification is typical Kandiudalf (Soil Survey Staff, 2006).

Soil sample preparation and analysis

The soil sample collected using the soil auger were air-dried, crushed gently and passed through a 2-mm sieve. After this, it was analyzed for pH, organic carbon, total N, available P, Mg, K, Ca, Na and particle size analysis. Particle size was determined by hydrometer method (Gee and Bander, 1986). Available P was determined by Bray 1 method (Anderson and Ingram, 1993). Total N was determined by micro-kjeldahl method (Brookes et al., 1985). Soil reaction (pH) was determined in a 1:2 soil to water suspension using a pH meter (Maclean, 1982). Ca and Mg was determined using atomic absorption spectrophotometer, while k and Na were read from a flame photometer (IITA, 1979). Organic carbon was determined by the Walkley and Black dichromate wet oxidation method (Nelson and Summers, 1982).

Cropping history

In the year 2012, maize was cultivated during the early planting season, while cowpea was planted during the late season. Rice was cultivated during the 2013 early season while groundnut and soybean were planted in the late season. In the year 2014, rice was planted during the early season while cowpea was planted during the late season. In 2015 during the early season, maize was planted. The experiment was conducted in the late season of 2015. No fertilizer was added to the land.

Source of plant materials

Two varieties of cowpea were used for the experiment and were obtained from the International Institute of Tropical Agriculture (IITA), Ibadan, Oyo State, Nigeria.

Experimental design

The experiment was laid out in Randomized Complete Block Design (RCBD) with three replicates. The treatment was two cowpea varieties (IT90K-277-2 and Oloyin). The plot size was 5 m × 4 m.

Land preparation and planting

The land was ploughed and stumped. The field was divided into three plots using measuring tape, pegs and ropes. Soil samples were taken randomly from the field at three different spots and at a depth of 15 cm using the soil auger. 3 – 4 seeds of each variety were planted per hole with an inter row spacing of 0.75 m and intra row spacing of 0.25 m. Seedlings were thinned at 2 weeks after planting to 2 per hole and lost stands were supplied. Weeding was done at 4th and 6th WAP. Spraying of insecticides cypermethrin at the rate of 45.2 ml was done to control insect's attacks.

Data collection

The following data were collected from five randomly selected plants that were tagged. These data were taken at two weeks interval.

Growth parameter: Plant height, number of leaves, leaf area, chlorophyll content, stem girth, canopy height, number of branches, nodule number, nodule weight, above ground dry weight.

Yield parameter: 100% grain weight, shelling %, harvest index, grain yield/plant, grain yield/ha, fodder weight (dry and fresh), number of pod/plant, number of seeds/pod, pod weight/plant, seed weight/plant, days to 50% flowering.

Ureide analysis

This was done using the hot water extraction method. The stem of the cowpea plants was harvested with the leaves separated from the stem segment. The plant samples were then put in labeled paper bags to be dried at 65-80°C in a forced-air oven for 2-3 days. The total dry weight was taken and tissue was grinded to pass a 60-mesh (1.0 mm) screen. Contents were packed and stored in a cool dry place after this.

Subsamples dried and grounded weighing 0.5 g were taken and transferred to a 100-ml beaker. Distilled water of 25 ml was added to each subsamples and boiled for 1-2 min using electric heater. Filtration was done immediately while still hot through a funnel and 15 cm filter paper into a 50-ml volumetric flask. Residue was washed onto the filter and rinsed with a little distilled water. After the contents of the flask were cooled the volume was made to 50 ml with distilled water. The extract was then stored in a freezer in small plastic containers until the analysis of N solutes. Ureides were extracted from ground tissues. Samples of approximately 0.1 g dry weight were extracted with 1.0 cm^3 of 0.2 kmol m^{-3} NaOH in a boiling water bath for 30 min. The Ureide extracts was determined using the method of Young and Conway (1942). Nitrate was measured by the salicylic acid technique (Cataldo et al., 1975).

Data analysis

The data were subjected to Analysis of Variance (ANOVA) using Genstat 12th edition statistical software application. The means were separated using Least Significance Difference (LSD).

RESULTS

The result of the soil physico-chemical properties as presented in Table 1 shows that the soil pH (H_2O) is neutral (6.8) with a moderate organic matter content (2.408). The result also shows that the total Nitrogen and the available Phosphorus of the soil were low (0.12% and 12.451 mg/kg respectively). The exchangeable bases (Na, K, Ca and Mg) were also low (0.428, 0.612, 2.691 and 1.082 Cmol/kg respectively). The soil textural class was sandy loam.

The growth performance of the two cowpea varieties is presented in Table 2. There

Table 1. Physical and chemical properties of experimental soil.

Soil properties	Values
pH(H_2O)	6.800
Organic carbon (%)	1.397
Organic matter (%)	2.408
Total nitrogen (%)	0.120
Available phosphorus (mg/kg)	12.451
Exchangeable acidity (cmol/kg)	0.400
Clay (%)	10.40
Sand (%)	75.40
Silt (%)	14.20
Na (Cmol/kg)	0.428
K (Cmol/kg)	0.612
Ca (Cmol/kg)	2.691
Mg (Cmol/kg)	1.082
Textural class	Sandy loam

Table 2. Effect of cropping sequence on plant height, number of leaves and stem girth.

Variety	Plant Height	No. of leaves	Stem Girth
IT90K-277-2	61.4[b]	15.87[b]	0.25[a]
Oloyin	65.33[a]	18.13[a]	0.35[a]
LSD (5%)	1.434	1.518	0.1083

Values having the same letter are not significantly different at 5% P.

was significant difference in the plant height and number of leaves of the two varieties. The local variety Oloyin had significantly higher height and number of leaves than variety IT90K-277-2.

The performance of the two cowpea varieties in terms of number of nodules, nodules fresh weight and nodules dry weight is shown in Table 3. IT90K-277-2 has the highest number of nodules and nodules fresh weight. There was no significant difference observed in the number of nodules. There was no significant difference observed in the nodules fresh weight and the nodules dry weight of the two varieties.

Table 4 shows the nitrogen use efficiency of the two cowpea varieties. It shows and explains the higher potential of Oloyin over IT-90K-277-2 variety to supply more nitrogen to the soil when used for green manuring to enhance soil fertility.

Table 3. Effect of cropping sequence on number of nodules, nodules fresh weight and nodules dry weight.

Variety	No. of nodules	Nodules Fresh weight	Nodules dry weight
IT90K-277-2	390756a	2.74a	0.8a
Oloyin	307911b	2.10a	1.25a
LSD (5%)	356450.9	3.143	2.00

Values having the same letter are not significantly different at 5% P.

Table 4. The Nitrogen use efficiency.

Variety	Yield (kg/ha)	Nitrogen use efficiency (kg/kg)	% Nitrogen use efficiency
IT-90K-277-2	3700	30833	48.61
Oloyin	7200	60000	51.38
LSD (5%)	993.7	8280.5	

Also, there was no significant difference in the chlorophyll content of the cowpea (Figure 1). Oloyin had the highest chlorophyll content.

There was no significant difference in the N uptake, shoot dry weight yield, grain yield and N concentration of the two varieties (Figures 2 to 5).

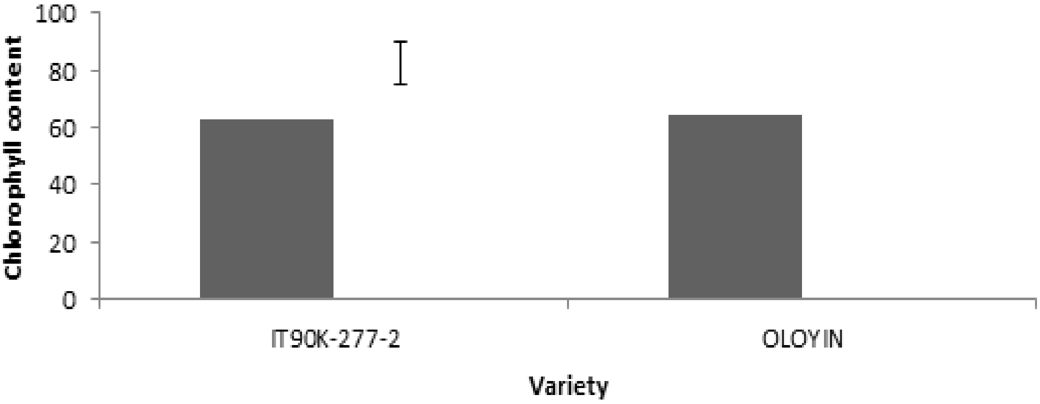

Figure 1. Effect of cropping sequence on chlorophyll content. The vertical bar represents L.S.D at P (5%).

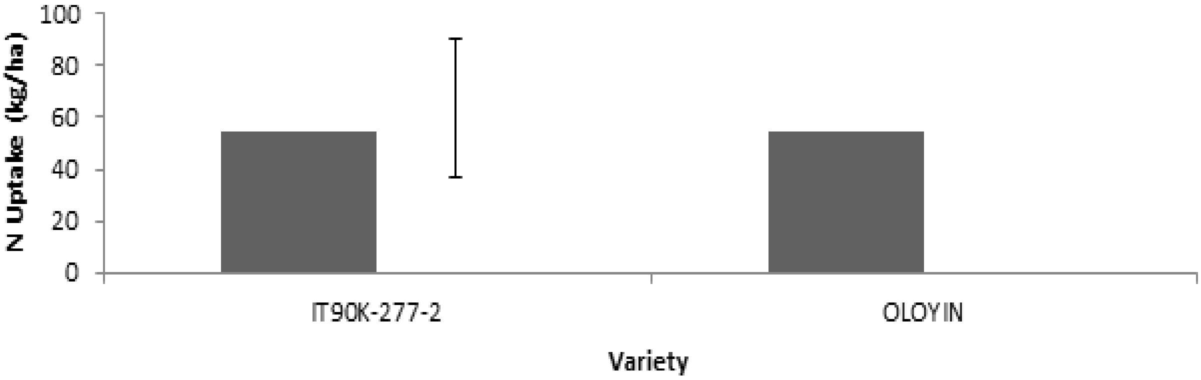

Figure 2. Effect of cropping sequence on nitrogen uptake. The vertical bar represents L.S.D at P (5%).

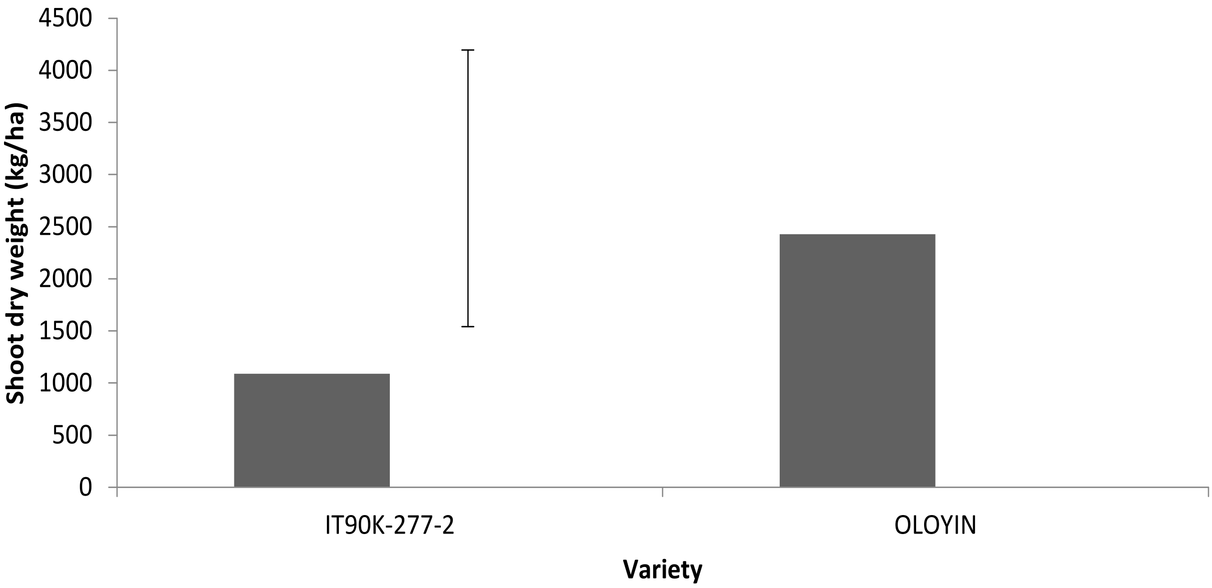

Figure 3. Effect of cropping sequence on shoot dry weight. The vertical bar represents L.S.D at P (5%) .

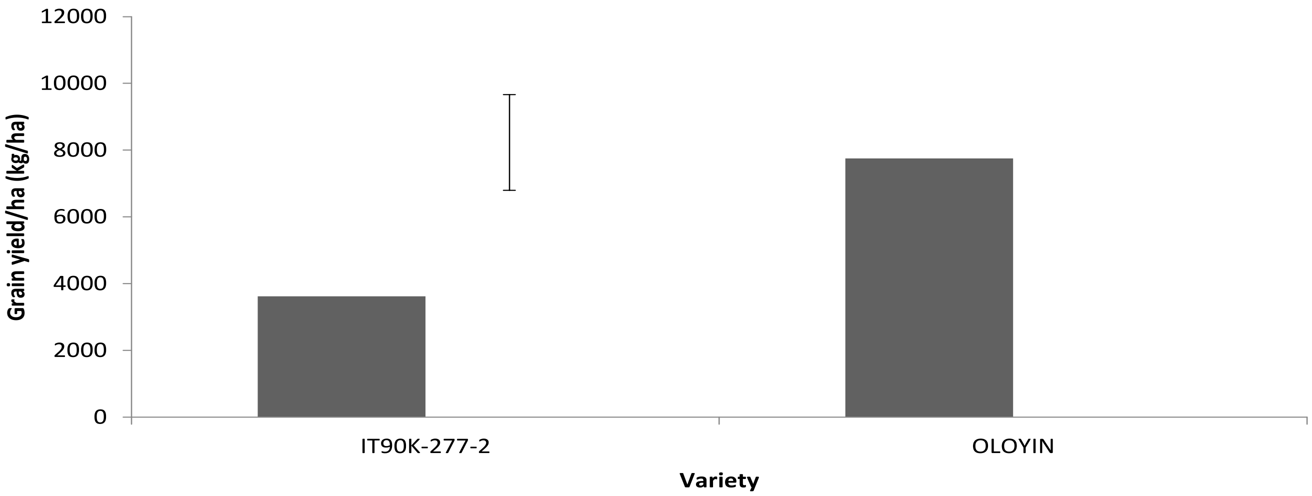

Figure 4. Effect of chlorophyll content on grain yield. The vertical bar represents L.S.D at P (5%)

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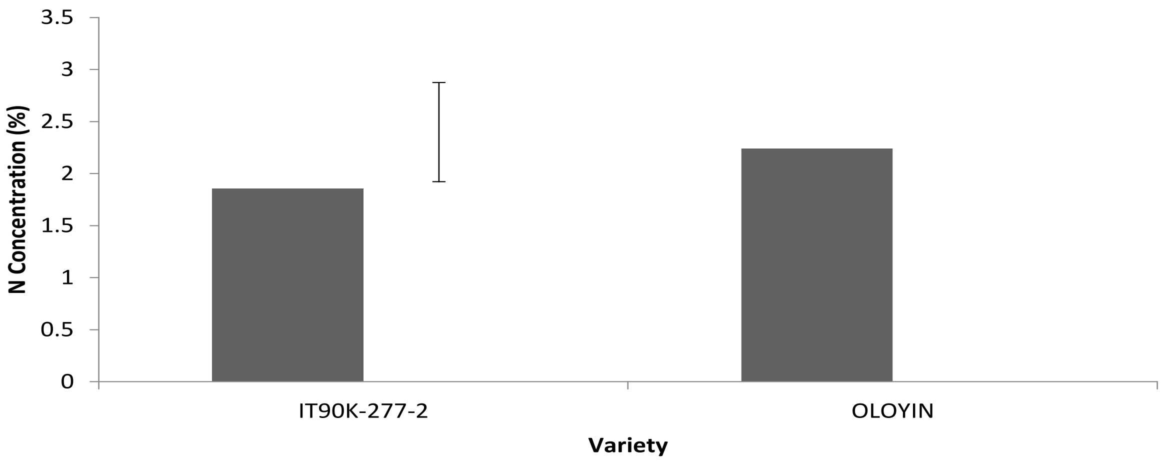

Figure 5. Effect of cropping sequence on nitrogen concentration. The vertical bar represents L.S.D at P (5%).

The percent nitrogen fixed and amount of total nitrogen fixed from the atmosphere was also not significantly different (Figure 6 and 7).

Figure 8 explains the relationship between the nitrogen fixed from the atmosphere and the grain yield of cowpea which is best represented by a polynomial relationship. This explains that about 51.68% of the grain yield of cowpea can be accounted for by the percentage of the nitrogen fixed from the atmosphere.

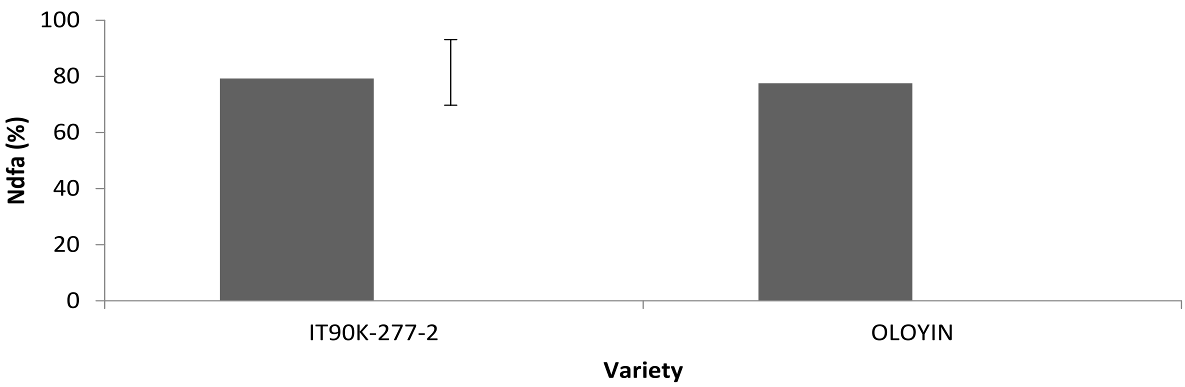

Figure 6. Effect of cropping sequence on % nitrogen fixed. The vertical bar represents L.S.D at P (5%) .

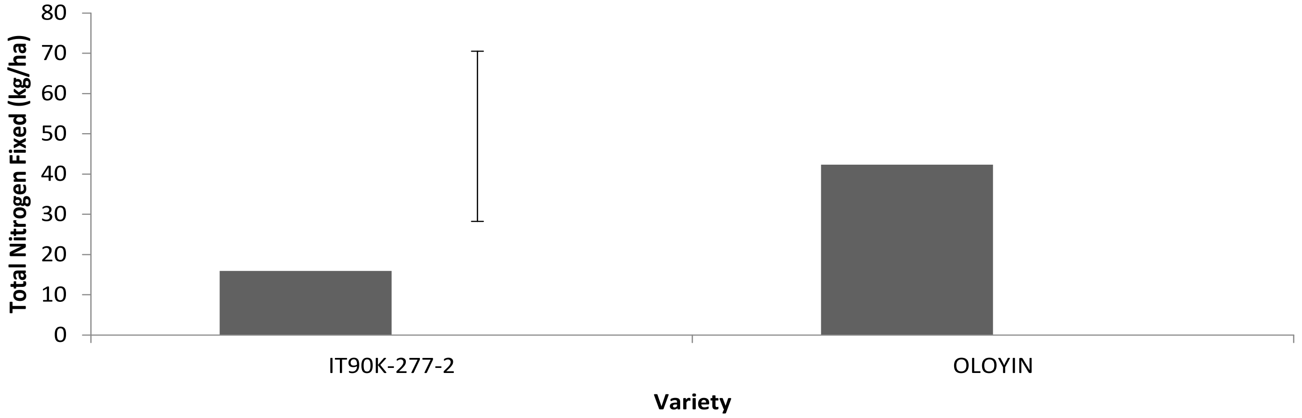

Figure 7. Effect of cropping sequence on total nitrogen fixed. The vertical bar represents L.S.D at P (5%) .

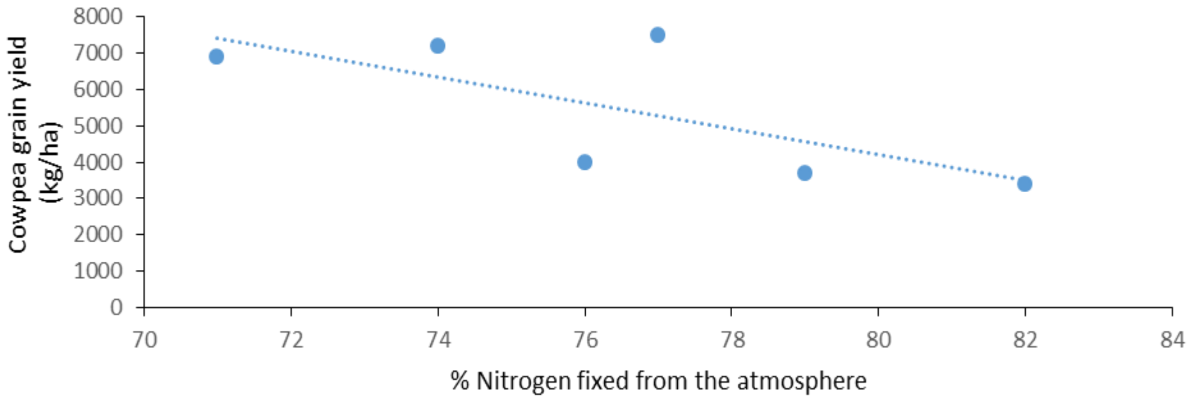

Figure 8. Relationship between the Nitrogen fixed from the atmosphere and the grain yield of cowpea.

The relationship between the nitrogen fixed in the atmosphere and percentage of nitrogen in the plant tissue of cowpea is best explained by a polynomial relationship in Figure 9. This shows that 65.19 % of the percentage of nitrogen in the plant tissue can be explained by the atmospheric nitrogen trapped by the plant.

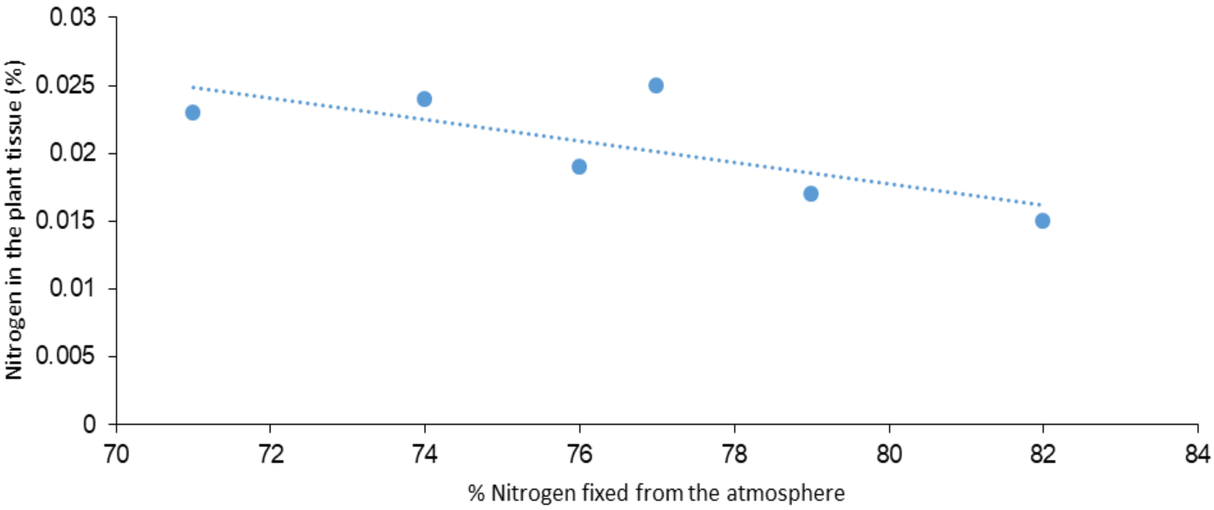

Figure 9. Relationship between the Nitrogen fixed in the atmosphere and percentage of Nitrogen in the plant tissue of cowpea.

DISCUSSION

There was no significant difference in the stem girth of the two varieties. This response may be a result of genetic differences existing between the two varieties and is in agreement with Mellek et al. (2010).

The non-significant correlation among the chlorophyll content is possibly a genotypic response. The response may have happened because the amount of nitrogen supplied through nitrogen fixation was higher and met the needs of this nutrient by plants, favoring the higher chlorophyll content. According to Taiz and Zeiger (2013), plants with a high concentration of chlorophyll are potentially capable of achieving higher photosynthetic rates due to its light energy capture value per time unit. The nitrogen use efficiency explains the potential of nitrogen that could be gained from the two varieties when incorporated into the soil as a nitrogen source as it has been used to enhance soil fertility status for cereal production (Galindo et al., 2021).

There was no significant difference in N uptake, shoot dry weight yield, grain yield and N concentration of the two varieties (Figures 2 to 5). The amount of nitrogen fixed from the atmosphere was also not significantly different (Figures 6 and 7). This observation indicates that the two varieties of cowpea were effective in soil that had sufficient levels of P and K, as suggested by Stamford et al. (2008). The relationship between the nitrogen fixed from the atmosphere and the grain yield of cowpea supports the essentiality of nitrogen as a limiting nutrient for cowpea production and it also points to the importance of the high Phosphorus content in the soil (Table 1) as responsible for enhancing nitrogen fixation, nodulation and high cowpea productivity (Nielsen et al., 2001: Nziguhela et al., 2016). The relationship between the nitrogen fixed in the atmosphere and percentage of nitrogen in the plant tissue of cowpea supports the nitrogen fixing ability of cowpea as sufficient nitrogen is made available in the soil to enhance soil fertility status when the grain is harvested and the residue incorporated as green manure after harvest to boost crop productivity as sufficient nitrogen is available in the soil and plant tissue (Okonji et al., 2011).

CONCLUSIONS AND RECOMMENDATIONS

Cowpea variety Oloyin has higher yield and nitrogen fixing ability in the soil as compared to IT90K-277-2. The variety Oloyin can be adopted by farmers who engage in the legume-cereal cropping sequence as its potential to boost food and livelihood security is more prominent among the people.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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