Influence of Arbuscular Mycorrhizal Fungus Inoculation and Hedgerow Tree Pruning Regimes on Three Cassava Genotypes

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Abstract: Background: A field trial experiment was conducted with three cassava genotypes to evaluate their response to inoculation with arbuscular mycorrhizal fungus, *Glomus deserticola* and mulching under two- and three- monthly pruning regimes in a one- year old alley cropping system. **Result:** Inoculation with *G. deserticola* significantly increased the biomass accumulation and nutrient yield of alley cropped cassava. Mycorrhizal colonization of cassava roots was significantly higher in inoculated plants than uninoculated ones. Colonization was affected by pruning regimes as two monthly pruning regime favoured higher root colonization in all the genotypes in most cases. Higher fresh tuber yields were also obtained with two monthly pruning regime. Alley cropping cassava between *Senna siamea* hedgerows gave the lowest yield. On the whole, alley cropping with *Gliricidia sepium* and *S. siamea* mixed hedgerows in conjunction with *G. deserticola* inoculation gave the highest cassava root tuber yield followed by *G. sepium* alone. There were significant ($p < 0.05$) positive correlations between arbuscular mycorrhizal fungi colonization and root tuber yield of the three cassava genotypes. Root tuber nutrient yield for nitrogen, potassium and phosphorus was significantly increased by *G. deserticola* inoculation in all the genotypes. **Conclusion:** Cassava cv TMS 4 (2) 1425 root tuber yield responded most favourably to mycorrhizal inoculation under two - monthly pruning regime.

Key words: *Glomus deserticola*, alley cropping, cassava yield

INTRODUCTION

Cassava (*Manihot esculenta* Crantz) is an important staple food crop in tropical and subtropical Africa as well as Asia and Latin America. It plays a major role in efforts to alleviate the African food crisis taking advantage of its efficient food energy production, year round availability, tolerance to extreme stress conditions and relatively high productivity (Hahn and Keyser, 1985). Cassava root tubers are important carbohydrate source while the leaves are consumed as proteinaceous vegetable. It extracts large amounts of nutrients from the soil (Pellet and El-Sharkawy, 1993) making addition of nutrient to the soil after each harvest mandatory if the subsequent yield is to be worthwhile.

Cassava is highly mycorrhizal dependent for nutrient extraction (Howeler *et al.*, 1987). The use of organic matter to enrich the soil is fast gaining acceptance (Amaya-Carpio *et al.*, 2009). Among the advantages attributed to organic manure application to the soil include increase in soil water holding capacity and nutrient supply to the crops (Amanullah *et al.*, 2007). The source of this organic matter must be within easy reach of the farmers. This can be achieved by planting cassava in alley cropping plots where leguminous trees are planted in hedgerows. The hedgerow trees can then be

periodically pruned and used in mulching the soil. As a way of tackling nutrient depletion and soil erosion, some researches carried out with alley cropping as well as mulch application have yielded positive results (Cadavid *et al.*, 1998). The use of mulch in conjunction with arbuscular mycorrhiza has been shown to increase yield in cassava (Salami and Osonubi, 2003; Liasu *et al.*, 2006; Okon *et al.*, 2010). There is however, no data on the comparative response of some International Institute for Tropical Agriculture (IITA) improved cassava genotypes to mycorrhizal inoculation and hedgerow pruning or mulch application.

This study was carried out to investigate whether there is any variation in the responsiveness of three IITA cassava genotypes, most commonly planted by the local farmers in Ibadan, southwestern Nigeria, to mycorrhizal inoculation, hedgerow trees pruning regimes and mulching with a view to recommending the highest yielding genotype to them.

MATERIALS AND METHODS

This alley cropping experiment was conducted in a one year fallow alley cropped plot of the University of Ibadan Research farm, Nigeria located on 7° 45' N and 13° 9' E. The soil was a sandy clay loam Rhodic

Kandiustalf type of Balogun series. The total rainfall for the period of the experiment was 1298.4 mm while the total evaporation was 1499.0 mm. Soil properties at 0-27 cm depth revealed pH 6.50; organic carbon 0.98%; total N 0.036%; extractable P 1.17 mg kg⁻¹; Ca 3.12 mg kg⁻¹; 1.91, 0.11, 0.19 cmol kg⁻¹ of Mg, Na and K, respectively.

Experimental layout and design: The experiment was laid out in a split-split-plot design with each treatment combination replicated three times. The main plots consisted of Arbuscular Mycorrhizal Fungus (AMF) inoculated (M⁺) and uninoculated (M⁻) arranged in blocks to reduce the possibility of mycorrhizal contamination of the uninoculated plots. Treatments were split on hedgerow tree species as subplots of *Gliricidia sepium*, *Senna siamea* and *G. sepium* interplanted with *S. siamea* while two- and three-monthly pruning regimes constituted the sub-subplots. Each subplot (12 × 12 m) consisted of three rows of each hedgerow tree species or their interplanted mixtures at 4 m inter row spacing and 0.5 m intra row spacing to give a population of 5,000 trees per hectare. Each sub-subplot was 6 × 12 m in size. The initial pruning of hedgerow trees was done at the beginning of the experiment and 0.5 t ha⁻¹ dry matter equivalent of their leaves was used to mulch their respective plots. Subsequent pruning at 2- and 3-month intervals were applied at the same rate to their designated sub-subplots.

Cassava (*Manihot esculenta* Crantz) genotypes cv TMS 30572; TMS 4 (2) 1425 and TMS 91934 stem cuttings (0.2 m long) were planted in a regular sequence at 1 × 1 m spacing to give a population of 10,000 cassava stands per hectare in the sole cassava (treeless control) subplots and 7,500 stands per hectare in the alley-cropped subplots. Mycorrhizal plots were inoculated with 20 g crude inoculum of *Glomus deserticola* Trappe, Bloss and Menge (INVAM, CA 113) applied directly in the planting holes. A basal fertilizer (N, P, K, 27:10:10) dressing of 25 kg ha⁻¹ was applied to the cassava. Weeding was done manually as necessary.

Harvesting was done at the end of twelve months. Dry matter yield for each cassava genotype was determined by oven drying the stems and leaves to constant weight at 70°C while the fresh tuber weight was determined at harvest. Subsamples of the root tubers were oven dried and ground for N, P, K analysis following the method of Juo (1979). Nutrients uptake was calculated as the product of percentage nutrient concentration and yield (Parmar and Sharma, 1996). Percentage AMF colonization of roots was determined for each cassava genotype by clearing and staining representative terminal feeder roots with trypan blue (Koske and Gemma, 1989) and evaluating by gridline intersect method of Giovannetti

and Mosse (1980). Cassava root tuber yield response to hedgerow trees by each genotype was calculated as the difference between AMF inoculated or uninoculated cassava alley-cropped with each hedgerow tree species and their respective sole-cropped cassava plants. Soil samples were taken from the different experimental plots to the for nutrient analysis following the method of Juo (1979).

All data were subjected to combined analyses of variance using SAS (SAS Institute, 1996) and Duncan multiple range test was used to separate the means.

RESULTS

Arbuscular Mycorrhizal Fungi (AMF) colonization of cassava roots: Inoculated cassava roots had significantly higher percentage of AMF colonization than those of uninoculated ones. Colonization generally was significantly lower in cassava roots planted under three-monthly pruning regime than under two-monthly pruning regime (Table 1).

Cassava biomass yield: Inoculation with AMF significantly increased all the yield components of the three cassava genotypes in all the hedgerow treatments and control except *S. siamea* where the leaf and stem dry matter were similar for cv TMS 91934 (Table 2). There was a significantly positive relationship between AMF percentage root colonization and root tuber yield (Fig. 1). Two-monthly pruning regime gave significantly higher tuber yield than three-monthly pruning regime in all the genotypes (Table 2). Interplanting *G. sepium* with *S. siamea* as hedgerows consistently gave the highest biomass yield followed by *G. sepium* plots while

Table 1: Effect of pruning regimes and AMF inoculation on the colonization of cassava enotypes

Hedgerow	Pruning	TMS30572	TMS 4 (2)1425	TMS 91934
<i>S. siamea</i>	M ⁺ 2mo	55.5 ^f	53.3 ^e	51.8 ^{de}
	M ⁺ 3mo	45.6 ^e	36.8 ^{gh}	48.3 ^e
	M ⁻ 2mo	39.7 ^h	25.0 ⁱ	21.8 ^h
	M ⁻ 3mo	33.5 ⁱ	20.0 ^j	14.2 ⁱ
<i>G. sepium</i>	M ⁺ 2mo	84.1 ^b	81.5 ^b	76.4 ^b
	M ⁺ 3mo	75.6 ^d	67.5 ^c	64.0 ^c
	M ⁻ 2mo	54.9 ^f	33.7 ^h	51.6 ^{de}
	M ⁻ 3mo	53.0 ^f	39.5 ^g	36.9 ^f
Gs+Ss	M ⁺ 2mo	88.7 ^a	86.7 ^a	83.8 ^a
	M ⁺ 3mo	78.5 ^c	77.6 ^b	74.8 ^b
	M ⁻ 2mo	61.5 ^e	59.9 ^d	56.9 ^d
	M ⁻ 3mo	64.1 ^e	49.0 ^f	29.4 ^g
Control	M ⁺	64.0 ^e	60.4 ^d	69.1 ^c
	M ⁻	42.0 ^g	18.7 ^j	18.7 ^{hi}

M⁺: Mycorrhiza inoculated; M⁻: Mycorrhiza uninoculated; 2mo and 3mo: two- and three- monthly pruning regimes (Pr). Gs+Ss: *Gliricidia* and *senna* mixtures. Means in the same column followed by different letters are significantly different at p<0.05 according to Duncan's multiple range test

Table 2: Effect of hedgerow tree species pruning regimes and AMF inoculation on cassava biomass yield (tha⁻¹)

Hedgerow	Pruning	TMS 30572			TMS 4 (2)1425			TMS 91934		
		TFW	SDW	LDW	TFW	SDW	LDW	TFW	SDW	LDW
<i>S. siamea</i>	M*2mo	15.0 ^g	3.1 ^g	1.4 ^f	12.3 ^d	1.6 ^{fg}	1.4 ^g	10.8 ^f	0.9 ^d	0.7 ^g
	M*3mo	12.8 ^g	3.2 ^g	0.9 ^g	10.0 ^f	1.1 ^{gh}	0.8 ^h	8.5 ^g	0.4 ^d	0.3 ⁱ
	M2mo	8.8 ^h	7.0 ^d	3.3 ^e	5.0 ⁱ	0.4 ⁱ	0.4 ⁱ	4.3 ⁱ	0.2 ^d	0.2 ⁱ
<i>G. sepium</i>	M3mo	6.0 ^h	1.1 ^{hi}	0.6 ^h	3.5 ^m	0.3 ⁱ	0.3 ⁱ	2.5 ⁱ	0.3 ^d	0.2 ⁱ
	M*2mo	28.5 ^b	17.0 ^b	5.8 ^b	20.0 ^e	7.4 ^b	5.0 ^b	18.5 ^c	7.2 ^a	4.3 ^b
	M*3mo	25.8 ^c	6.1 ^d	2.5 ^d	11.0 ^g	5.5 ^c	4.4 ^c	12.5 ^e	4.2 ^b	2.4 ^f
<i>Gs+Ss</i>	M2mo	20.5 ^e	6.8 ^d	2.5 ^d	7.5 ^{hi}	2.6 ^e	2.2 ^e	9.3 ^g	4.5 ^b	2.8 ^d
	M3mo	13.5 ^h	3.7 ^g	2.0 ^f	6.3 ^{jk}	1.8 ^f	1.3 ^g	4.5 ⁱ	3.7 ^{bc}	1.9 ^f
	M*2mo	31.3 ^a	19.2 ^a	6.3 ^a	33.3 ^a	8.5 ^a	5.8 ^a	31.0 ^a	6.3 ^a	5.3 ^a
Control	M*3mo	28.7 ^b	8.4 ^c	3.4 ^e	25.5 ^b	4.3 ^d	3.9 ^d	24.0 ^b	4.4 ^b	3.4 ^e
	M2mo	21.8 ^d	2.0 ^h	0.9 ^g	8.3 ^{gh}	2.5 ^e	2.5 ^e	11.5 ^f	4.6 ^b	4.0 ^b
	M3mo	19.5 ^f	5.4 ^e	2.5 ^d	6.8 ^{ij}	1.6 ^{fg}	1.8 ^f	6.5 ^h	0.9 ^d	0.9 ^g
Control	M*	20.3 ^{ef}	4.2 ^f	2.4 ^d	8.8 ^g	1.5 ^{fg}	0.7 ^h	13.5 ^d	2.9 ^c	0.7 ^{gh}
	M	8.3 ⁱ	0.7 ⁱ	0.5 ^h	5.7 ^{kl}	0.6 ^{hi}	0.5 ^{hi}	2.8 ⁱ	0.9 ^d	0.3 ^{hi}

M*: Mycorrhiza inoculated; M: Mycorrhiza uninoculated; 2mo and 3mo: two- and three- monthly pruning regimes (Pr). Gs+Ss: Gliricidia and senna mixtures. TFW: Tuber fresh weight; SDW: Stem dry weight; LDW: Leaf dry weight. Means in the same column followed by different letters are significantly different at $p < 0.05$ according to Duncan's multiple range test

Fig. 1: The relationship between cassava root tuber yield and arbuscular mycorrhizal fungi root colonization

S. siamea alone gave the lowest yield. Positive response of cassava tuber yield to hedgerow trees was favoured by AMF inoculation except in senna plots where negative response was observed (Fig. 2). Cassava cv TMS 4 (2) 1425 was found to be most dependent on mycorrhiza especially when mulched with *G. sepium* and *S. siamea* mulch mixture while to some extent, cv TMS 30572 was less dependent on mycorrhizal inoculation.

Cassava root tuber nutrient yield: Cassava root tuber N, P, K yield significantly increased by AMF inoculation in all the genotypes with those under two-monthly pruning regime giving higher nutrient yield than those of three months (Table 3).

Soil nutrient status: The soil nutrient contents were significantly increased ($p < 0.05$) compared to the treeless

Fig. 2: Response of cassava fresh tuber yield to hedgerow tree species pruning regimes

Table 3: Effect of hedgerow tree species pruning regimes and AMF inoculation on cassava tuber nutrient yield

Hedgerow	Pruning	TMS 30572			TMS 4(2)1425			TMS91934		
		N	P	K	N	P	K	N	P	K
<i>S. siamea</i>	M ⁺ 2mo	22.1 ^f	3.9 ^g	23.3 ^e	11.3 ^{gh}	3.3 ^c	22.3 ^d	15.1 ^d	2.3 ^{ef}	15.4 ^{de}
	M ⁺ 3mo	17.3 ^g	3.4 ^h	16.3 ^f	12.6 ^{fg}	2.2 ^f	14.2 ^{ef}	9.0 ^g	2.1 ^{fg}	11.3 ^f
	M ⁻ 2mo	11.3 ^h	1.9 ⁱ	11.5 ^g	5.4 ⁱ	1.2 ^j	6.8 ^{gh}	4.3 ^{hj}	1.5 ^{hi}	7.1 ^{gh}
<i>G. sepium</i>	M ⁺ 3mo	7.7 ⁱ	1.5 ^j	8.9 ^g	2.9 ^k	0.9 ^j	4.9 ^h	2.8 ^j	0.7 ⁱ	4.3 ^h
	M ⁺ 2mo	54.2 ^a	7.9 ^e	29.6 ^c	38.7 ^b	5.0 ^b	29.5 ^c	31.4 ^{ab}	4.6 ^c	23.4 ^b
	M ⁻ 3mo	40.0 ^f	4.8 ^f	25.0 ^{de}	29.0 ^d	3.0 ^d	20.9 ^d	17.4 ^d	3.4 ^d	18.1 ^{cd}
<i>Gs+Ss</i>	M ⁻ 2mo	26.9 ^g	7.0 ^d	24.4 ^{de}	8.9 ^j	1.4 ^{hi}	10.5 ^{fg}	10.8 ^{ef}	2.0 ^g	13.4 ^{ef}
	M ⁺ 3mo	17.3 ^g	3.3 ^h	11.3 ^g	9.6 ^{hi}	1.8 ^g	10.4 ^{fg}	5.9 ^{hi}	1.1 ^{ij}	7.0 ^{gh}
	M ⁺ 2mo	45.3 ^b	9.8 ^a	56.4 ^b	58.2 ^a	8.1 ^a	114.5 ^a	33.6 ^a	7.8 ^a	54.7 ^a
Control	M ⁻ 3mo	43.6 ^b	8.9 ^b	62.0 ^a	35.8 ^c	5.2 ^b	36.2 ^b	30.6 ^b	6.3 ^b	54.0 ^a
	M ⁻ 2mo	35.5 ^d	5.5 ^e	27.3 ^{cd}	15.3 ^e	2.7 ^e	15.5 ^e	12.0 ^f	3.5 ^d	20.2 ^{bc}
	M ⁺ 3mo	19.3 ^{fg}	3.9 ^g	18.1 ^f	8.6 ^f	1.6 ^h	10.4 ^{fg}	6.7 ^{gh}	1.8 ^{gh}	10.1 ^{fg}
Control	M ⁺	25.4 ^e	4.1 ^g	22.1 ^e	14.2 ^{ef}	1.9 ^g	11.2 ^{ef}	21.2 ^e	2.8 ^c	21.8 ^{bc}
	M ⁻	8.1 ⁱ	1.5 ^j	10.4 ^g	8.2 ⁱ	1.3 ⁱ	12.3 ^{ef}	3.8 ^{ij}	0.6 ^j	3.1 ^h

M⁺: Mycorrhiza inoculated; M⁻: Mycorrhiza uninoculated; 2mo and 3mo: two- and three-month pruning regimes (Pr). Gs+Ss: Gliricidia and senna mixtures. Means in the same column followed by different letters are significantly different at p<0.05 according to Duncan's multiple range test

control plots except for nitrogen (Table 4). Alley cropping with senna hedgerows resulted in significantly lower values of soil nitrogen, phosphorus and calcium compared to gliricidia or gliricidia interplanted with senna hedgerow plots.

Gliricidia hedgerows and its mixtures with senna gave a similar soil nutrient enrichment except for K where the soil from sole gliricidia hedgerows gave significantly higher soil nutrient content than the mixtures.

Table 4: Effect of alley cropping on the soil nutrient status

Hedgerow		Total N (%)	P (mg kg ⁻¹)	Ca	Mg (cmol kg ⁻¹)	K
<i>S. siamea</i>	M ⁺	0.15 ^b	8.28 ^e	8.29 ^e	1.63 ^e	0.38 ^e
	M ⁻	0.14 ^b	16.26 ^d	9.16 ^b	1.82 ^a	0.44 ^b
<i>G. sepium</i>	M ⁺	0.16 ^b	23.34 ^a	9.71 ^a	1.69 ^{bc}	0.58 ^a
	M ⁻	0.17 ^a	21.12 ^b	9.03 ^b	1.73 ^b	0.41 ^{bc}
Gs+Ss	M ⁺	0.17 ^a	17.94 ^c	9.66 ^a	1.72 ^b	0.38 ^e
	M ⁻	0.18 ^a	24.30 ^a	9.57 ^a	1.40 ^d	0.39 ^e
Control	M ⁺	0.15 ^b	3.42 ^f	5.42 ^e	1.26 ^e	0.15 ^e
	M ⁻	0.16 ^b	4.32 ^f	7.42 ^d	1.72 ^b	0.25 ^d

M⁺: Mycorrhiza inoculated; M⁻: Mycorrhiza uninoculated; Gs+Ss: Gliricidia and senna mixtures. Means in the same column followed by different letters are significantly different at $p < 0.05$ according to Duncan's multiple range test

DISCUSSION

The low percentage of AMF colonization recorded in cassava roots from three-month pruning regime treatments could have been due to shading by the hedgerow trees. Studies have shown that shading or reduction in photo-irradiance can reduce AMF growth (Smith and Gianinazzi-Pearson, 1990; Thomson *et al.*, 1990). A possible explanation for this could have been a lower carbon production by the shaded cassava leaves resulting in lower AMF colonization compared to those under the two-monthly pruning regime treatment. The increased root tuber and other components yields in inoculated cassava genotypes are similar to results obtained by Oyetunji and Osonubi (2007). Cassava is known to be highly mycorrhizal dependent (Howeler *et al.*, 1987). Therefore, the increased yield in inoculated cassava could have been due to improved nutrient and water uptake (Douds *et al.*, 2000; Amaya-Carpio *et al.*, 2009; Okon *et al.*, 2010). Increased nutrient uptake especially nitrogen and phosphorus would have then enhanced chlorophyll synthesis leading to enhanced photosynthesis and subsequent biomass accumulation. The higher biomass yield in inoculated mulched cassava could have resulted from not only the additional nutrients to the soil but also the enhanced mineralization by the presence of the mycorrhizal fungus. Feng *et al.* (2003) have reported mineralization and utilization of organic phosphorus by AMF. The positive relationship between AMF colonization and root tuber yield indicates the usefulness of the introduced AMF in enhancing growth and yield in cassava. The steeper slope in cv TMS 30572 than cv TMS 4 (2) 1425 and cv TMS 91934 is an indication that the indigenous AMF contributed more to its root tuber yield or that it is less mycorrhizal dependent than the other genotypes.

The higher cassava root tuber yields from the two-monthly pruning regime treatment than their three-monthly pruning regime counterparts could possibly be explained as resulting from: (a) the reduced amount of

shading on the cassava thereby giving it an advantage of adequate sunlight for photosynthesis; (b) frequent mulch application keeping the soil surface covered and supplied with nutrients while suppressing weeds for most part of the growing season; (c) more frequent addition of nutrient to the soil resulting from dying back of fine roots at each pruning (Haggar and Beer, 1993). The negative response of the three cassava genotypes to senna hedgerow plots and mulch could have resulted from (i) competition between its roots and cassava for nutrient and water as they both extracted them from within the same soil depth. Ruhigwa *et al.* (1992) have reported that up to 76% of senna feeder roots occupy the top 20 cm of the soil profile, a depth normally explored by most cassava roots; (ii) the initial immobilization of nitrogen by senna mulch (Tian *et al.*, 1992). The significantly higher root tuber yield in the gliricidia-senna mixed plots must have been due to advantage being taken of the slow decomposition and gradual release of nutrients from the mulch mixtures compared to gliricidia mulch alone. Besides increased nitrogen fixation to the soil caused by the close proximity of the nodulating gliricidia to the non-nodulating senna could have also been an added advantage. Van Noordwijk and Dommergues (1990) have explained the possibility of faster nitrogen depletion in this vicinity resulting in stimulated nitrogen fixation. This must have allowed the slow growing cassava time to absorb the nutrients without much leaching beyond the rooting zone.

The higher nutrient yields in all inoculated cassava genotypes than uninoculated ones must have resulted from enhanced nutrient uptake by the AMF hyphae while that of mulched plants apparently resulted from additional nutrients from the prunings.

CONCLUSION

The findings from this study have shown that integration of mycorrhizal application with mulch application from appropriate hedgerow trees in an alley cropping system can boost cassava production in marginal soil such as found in many parts of the tropics. Cassava cv TMS 4 (2) 1425 gave the highest yield with this technology and it is recommended that this genotype which is low in cyanide content should be cultivated by the local farmers followed by cv TMS 30572 in gliricidia-senna alley cropping system with appropriate mycorrhizal inoculation for maximum profitability.

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