

Combine effect of bio-fertilizer and poultry manure on growth, nutrients uptake and microbial population associated with sesame (*Sesamum indicum* L) in North-eastern Nigeria

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Abstract: The combine effect of bio-fertilizer (*Azospirillum* and arbuscular mycorrhiza fungi, AMF; *Glomus mossea*) and poultry manure (PM) on nutrients uptake, plant growth and soil microbial population associated with sesame under field condition was studied. The experiment comprised of four treatments; T1 (*Azospirillum* + AMF), T2 (*Azospirillum* + AMF + 5 ton ha⁻¹ poultry manure), T3 (10 ton ha⁻¹ poultry manure), and T0 (Control) laid out in randomized complete block design with three replications. Plant growth parameters viz., plant height, numbers of leaves/plant, numbers of branches/plant, leaf area, shoots and root dry biomass increased significantly due to the application of bio-fertilizer and poultry manure singly or in combination over control. Combined application of bio-fertilizer and poultry manure @ 5 ton ha⁻¹ (bio-organic) significantly produced the plants with desirable growth parameters, nutrients content and uptakes for N, P, and K. Root colonization by AM fungi was recorded in inoculated and un-inoculated plants. Colonization % ranges from 6% in control to 62.8% in Bio-organic. There was no significant difference in % root colonization of inoculated plants. Populations of *Azospirillum* and AM spores have increased in all treatments over the initial population prior to experiment in all treatments. Bio-organic recorded the highest *Azospirillum* population (28.56×10^6 CFU g⁻¹ soil) and 69.3 AM spores g⁻¹ soil and values were significantly higher to all the treatments. The overall findings of this research indicated that Bio-organic (bio-fertilizer and poultry manure @ 5 ton ha⁻¹) produced plants with highest growth parameters, nutrients uptake, increased soil *Azospirillum* population, AM spore density and mycorrhization compared to exclusive application of bio-fertilizer or poultry manure @ 10 ton ha⁻¹. Combination of bio-fertilizer with organic amendments could be recommended for successful production of sesame.

Key words: Bio-fertilizer, Poultry manure, Bio-organic, Plant growth, Nutrients uptake, Microbial population

I. Introduction

Sesame (*Sesamum indicum* L.) or beniseed is a high value cash crop commonly cultivated by small holder farmers in Nigeria (Uwala, 1998; Alegbejo, 2003) for oil seed. The seed is nutritious: it contains 50-52% oil, 19-25% protein and 16-18% carbohydrates (Uzoh, 1998; Weiss, 2000). Sesame is adapted to many soil types, drought-tolerant and grows on poor fertile soil. Virtually, poor-resource holder farmers in Nigeria rarely apply fertilizer, but, best yields are obtained in properly fertilized farms (Papari Moghaddamfard and Bahrani, 2005; Saeidi, 2008; Anon, 2006; Malik et al., 2003; Rao et al., 1994). Poor soil nutrients concentration especially, low N, P and soil organic matter, coupled with unscientific method of cultivation are major constraints to its production in Nigeria. Yields are as low as 300 kg ha⁻¹ compared to 1,960 kg ha⁻¹ in Venezuela, 1,083 kg ha⁻¹ in Saudi Arabia (Abubakar et al., 1998), 1323 kg ha⁻¹ in Egypt and 825 kg ha⁻¹ in Ethiopia. Chemical fertilizers have been used for decades to increase crop yield. However, current trends in agriculture are focused on search for alternative to chemical fertilizer due to, huge cost of procurement, contamination of environment, and couple with improper application leading to the degradation of soil quality (Tilman et al., 2002). Furthermore, the world demands for the production of quality food and in a most sustainable way maintaining soil biodiversity. Moreover, food produced organically fetch premium price in global market.

Bio-fertilizer (microbial inoculants) and organic amendments are cheap nutrient source that could serve as alternative to chemical fertilizers and improve crop production in low-input agriculture. Application of organic amendments, increases soil organic carbon and stimulate microbial activity which provides N and P to soil. Soil microorganisms on the other hand play a vital role in their ability to provide and recycle nutrient for plant growth (Weil and Magdoff, 2004). Their population and activities may not only reflect the quality of soil, but also reflect soil environmental conditions (Leangvutiviroj et al., 2010). They are involved in interactions with plant roots, either symbiotically or as free-living, improving plant nutrients uptake, bolster crop production and improve soil quality (Wu et al., 2005; Okon and Itzisoehn, 1995; Shah et al., 1992). Sesame production using organic amendments (Haruna and Abimiku, 2012; Ogbonna and Umar-Shaba, 2012) and chemical fertilizers

(Umar et al., 2012; Shehu et al., 2010; Jakusko and Usman, 2013) have well been documented in Nigeria. Babajide et al. (2012) conducted pot experiment on bio-fertilizer (mycorrhizal inoculum) with organic and inorganic fertilizer on performance of sesame, in South-western region of Nigeria. However, there is little or no similar such work under field conditions. In this context, the combine effect of bio-fertilizer (*Azospirillum* and arbuscularmycorrhizal fungi, AMF) and poultry manure (PM) on nutrients uptake, plant growth and soil microbial population associated with sesame was studied under field conditions.

II. Materials And Methods

The experiment was conducted at 2007/2008 rainy season in the Teaching and Research Farm, Agricultural Technology, Mohamet Lawan College of Agriculture, Maiduguri. Maiduguri lies in the semi-arid region of northeastern Nigeria, (11°15'N, 13°15'E latitude). It is characterized by a short rainy season of 3-4 months (June – September) with an annual rainfall varying from 300mm to 650mm and a long term mean of 503mm (Grema et al., 1995). The basic physico-chemical and biological properties of soil indicated that the soil was sandy loam (57% sand, 23.4% silt and 19.7% clay), neutral (pH 6.8), with EC value, 0.76dS m⁻¹. The soil had low nitrogen (0.17%), organic carbon (0.66%), available soil P (5.8 mg kg⁻¹), and exchangeable K (0.32 meq/100 g soil), with initial *Azospirillum brasilense* population as few as 0.34 × 10⁵, and *Glomus mossea* (AMF) 5 spores/g soil.

The treatments were; T1 (*Azospirillum* + AMF), T2 (*Azospirillum* + AMF + 5 ton ha⁻¹ poultry manure), T3 (10 ton ha⁻¹ poultry manure), and T0 (Control) laid out in randomized complete block design with three replications. *Azospirillum brasilense* was isolated from rhizosphere of maize, grown in nutrient broth for 48 h at 32°C in rotary shaker at the microbiological unit, Lake Chad Research Institute, Maiduguri. Number of *Azospirillum* in suspension was 1 × 10⁹ cfu ml⁻¹. Sand: soil mixture (1:2) containing extramatrical hyphae, spores, and root fragments of maize infected with *Glomus mossea* grown for 3 months was used as soil inoculums for AMF. *Azospirillum* was applied as seed treatments and sprayed in soil 1 week after seedling emergence at 500 ml ha⁻¹ in 2000 l of water. Viable sesame seeds (Ex-Sudan) were surface-sterilized and soaked in *Azospirillum* culture for 1 h which was sown immediately while 10 g of AMF inoculums (25 spores g⁻¹ soils with ≥ 80% root infection) was placed within inoculated seeds 3 cm below soil surface. The control plots were not inoculated.

Experimental site was ploughed and harrowed. Composted poultry manure (2.1% N, 1.3% P, and 1.5% K) was incorporated to individual plot 1 week prior to seed sowing according to treatments. Six sterilized and viable seeds were sown at 75 × 15 cm between and within rows in 2m × 2m and later thinned to one. Inoculated and un-inoculated plots were separated by 2 m unplanted boarder. Seeds were sown on 16th July and plants harvested 6 weeks after sowing (WAS). At harvest, observations on plant height, number of branches/plant, leaf area, dry biomass of roots and shoots (constant weight after oven drying @ 65°C), shoot content of N, P, and K were analyzed using Kjeldahl apparatus for nitrogen, while phosphorous and potassium were determined, by acetic acid extraction and measured with spectrophotometer and flame photometer as outlined by Johnson and Ulrich, (1959) and Knudsen *et al.*, (1982) for P and K respectively. *Azospirillum* in rhizosphere were detected by most probable method, mycorrhizal root colonization assessed by root clearing and staining (Philips and Hayman, 1970), and spore enumeration using wet-sieving and decanting (Gerdemann and Nicolson, 1963). Data were subjected to analysis of variance and differences among treatment means were separated using Fisher's least significant difference (LSD) at P ≤ 0.05.

III. Results

Plant growth response

Plant growth parameters *viz.*, plant height, numbers of leaves/plant, numbers of branches/plant, leaf area, shoots and root dry biomass (Fig.1 & Table 1) increased significantly due to the application of bio-fertilizer and poultry manure singly or in combination over control. Combined application of bio-fertilizer and poultry manure @ 5 ton ha⁻¹ (bio-organic) significantly produced the plants with desirable growth parameters; plant height (68.6 cm), numbers of leaves/plant (98.1), leaf area (6.2 dm²), shoot and root dry biomass (6.18, 3.88 g plant⁻¹), followed by bio-fertilizer and poultry manure (PM).

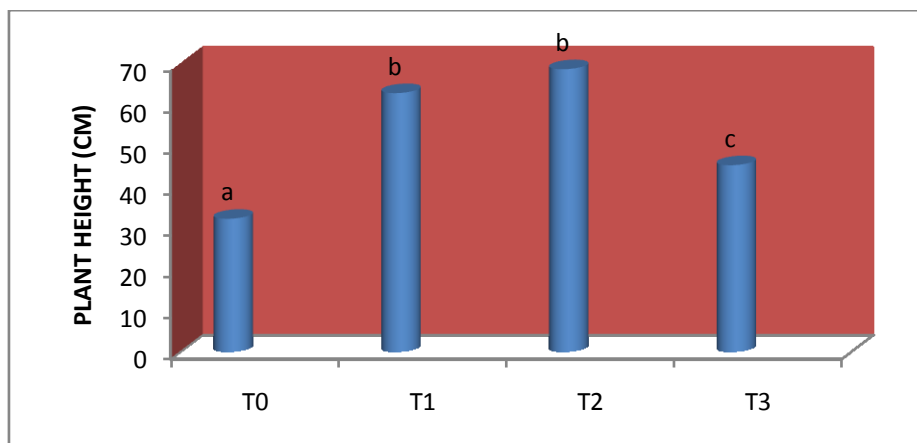


Fig. 1: Effect of bio-fertilizer and poultry manure on plant height of sesame 6 WAS

Legend: T0: (00) Absolute control, T1: (Bio-fertilizer), T2: (Bio-organic), T3: (PM).

Table 1: Growth response of sesame as affected by bio-fertilizer and poultry manure 6 WAS

Treatments	No. of branches /plant	No. of leaves /plant	Leaf (dm ²)	Dry biomass (g/plant)	
				Shoot	Root
T0 Control	4.5 ^a	42.3 ^a	3.8 ^a	4.45 ^a	2.72 ^a
T1 Bio-fertilizer	7.3 ^b	92.8 ^b	5.3 ^b	5.40 ^b	3.70 ^b
T2 Bio-organic	8.3 ^b	98.1 ^b	6.2 ^c	6.18 ^c	3.88 ^{bc}
T3 PM	7.3 ^b	75.3 ^c	5.1 ^b	5.35 ^b	3.62 ^b
LSD (5%)	1.92	7.08	0.67	0.62	0.22

Values followed by the same superscript are not significantly ($P < 0.05$) different according to Fischer's LSD test

Nutrients concentration and uptake

Nutrients concentration and uptakes (Table 2) differs significantly amongst all the treatments. Plants inoculation with bio-fertilizer singly or in combination with PM increased nutrients contents and uptakes compared to the control. Bio-organic recorded the highest nutrients content and uptakes for N, P, and K, and values were significantly higher to plants with single application of either bio-fertilizer or PM.

Table 2: Shoot nutrient contents and uptakes of N, P, and K as affected by bio-fertilizer and poultry manure on sesame 6 WAS

Treatments	Concentration (%)			Uptakes (kg ha ⁻¹)		
	N	P	K	N	P	K
T0 Control	1.82 ^a	0.32 ^a	0.98 ^a	12.47 ^a	1.34 ^a	34.56 ^a
T1 Biofertilizer	3.54 ^b	0.51 ^b	1.43 ^b	36.38 ^b	5.62 ^b	50.41 ^b
T2 Bio-organic	3.93 ^c	0.56 ^c	1.62 ^c	40.96 ^c	6.79 ^c	51.44 ^b
T3 PM	3.20 ^d	0.46 ^d	1.30 ^d	31.42 ^d	3.02 ^d	45.47 ^c
LSD (5%)	0.11	0.02	0.09	3.76	0.54	3.98

Values followed by the same superscript are not significantly ($P < 0.05$) different according to Fischer's LSD test

Root colonization

Root colonization by AM fungi was recorded in inoculated and un-inoculated plants (Fig. 2). Colonization % ranges from 6% in control to 62.8% in Bio-organic. There was no significant difference in % root colonization of inoculated plants. However, un-inoculated plants treated with 10 ton ha⁻¹ PM recorded significant colonization level of 11.8% over control (6.0%).

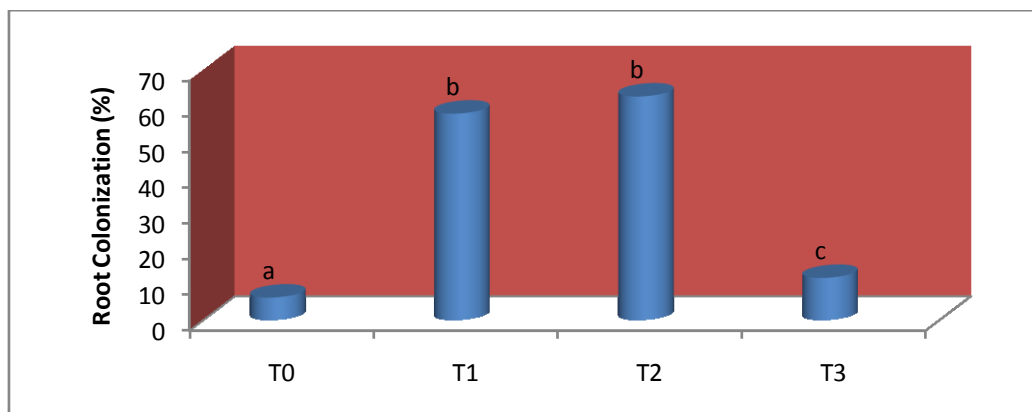


Fig. 2: Effect of bio-fertilizer and poultry manure on root colonization of sesame by AM fungi 6 WAS
Legend: T0: (00) Absolute control, T1: (Bio-fertilizer), T2: (Bio-organic), T3: (PM).

Azospirillum population and soil spore density of AMF

Populations of *Azospirillum* and AM spores have increased in all treatments over the initial population prior to experiment. Bio-organic recorded the highest *Azospirillum* population (28.56×10^{-6} CFU g^{-1} soil) and 69.3 AM spores g^{-1} soil and values were significantly higher to all the treatments.

Table 3: *Azospirillum* population and AM spores as affected by different treatments 6 WAS of sesame

Treatments	<i>A. brasilenses</i> (CFU $\times 10^{-6} g^{-1}$ soil)	<i>G. mossea</i> (spores g^{-1} soil)
T0 Control	4.50 ^a	8 ^a
T1 Bio-fertilizer	23.12 ^b	62.5 ^b
T2 Bio-organic	28.56 ^c	69.3 ^c
T3 PM	13.16 ^d	10.3 ^a
LSD (5%)	0.06	4.61

Values followed by the same superscript are not significantly ($P < 0.05$) different according to Fischer's LSD test

IV. Discussion

Response in plant growth characteristic and nutrients uptakes in sesame plants was observed under inoculated plants compared to un-inoculated ones. Bio-organic produced plants with highest growth parameters, nutrients uptakes, colonization % and increased microbial population followed by single application of bio-fertilizer then PM @ 10 ton ha^{-1} , while control recorded the least. Poor growth characteristics in control plants could be due to poor nutrients concentration and low soil organic matter in the study site. The positive effects of bio-fertilizer and poultry manure on plant growth as observed in this study have also been reported by some workers (Ahiabor and Hirata, 1994; Nwagburuka et al., 2012) using various organic amendments. They observed that inoculated plants grown with organic amendments produced plants with higher growth characteristics than un-inoculated ones. Several workers have reported similar effect of bio-fertilizer on crop nutrients availability and yield (Sridevi and Ramakrishnan, 2010; Geeta et al., 2013). However, results were better obtained when bio-fertilizers were combined with organic amendments (Babajide et al., 2012; Worthington, 2001; Asaf et al., 2009). Positive growth response of inoculated plants could be due to provision of nutrients especially nitrogen and growth promoting hormones by *Azospirillum* and enhanced uptake of phosphorus and other nutrients due to mycorrhizal colonization (Bama and Ramakrishnan, 2010; Zaidi et al., 2004). Enhanced nutrients availability could also be attributed to the decomposition of organic manure or transforming of inorganic substances to available form by microorganisms. Increase in *Azospirillum* population, AM spore density and % colonization could be motivated by the application of poultry manure, which became a source of carbon to the microbes thereby motivating their activities. Gianinazzi-Pearson (1982) reported that free living rhizobacteria such as *Azotobacter* and *Azospirillum* spp. can motivate spore production, bacterial community and colonization in rhizosphere of mycorrhizal plants. The result also confirms previous work on microbial properties in relationship to organic amendments (Groaker and Sreenivasa, 1994; Johansson et al., 2004; Crecchio et al., 2001). They concluded that addition of organic amendments could increase bacterial population and enhanced AMF colonization.

V. Conclusion

The overall findings of this research indicated that combined effect of bio-fertilizer and poultry manure @ 5 ton ha⁻¹ produced plants with highest growth parameters and increased soil *Azospirillum* population, AM spore density and mycorrhization compared to exclusive application of bio-fertilizer or poultry manure @ 10 ton ha⁻¹. For successful production of sesame especially, on sandy soil with low nutrients concentration, and poor soil organic carbon, bio-organic fertilization could be recommended.

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