

## Effects of soil amendments on the nutritional quality of okra (*Abelmoschus esculentus* [L.] Moench)

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### Abstract

This study examined the effects of different soil amendments [compost organic fertiliser (OR), NPK (IO), *Glomus mosseae* mycorrhiza (MY) or no soil amendment as the control (CT)] on the nutritional quality and nutrient uptake of okra during cultivation in a field contaminated with sewage sludge from the two oxidation ponds of the Obafemi Awolowo University, Ile-Ife, Nigeria. Okra (*Abelmoschus esculentus* [L.] Moench) belongs to the Malvacea family. The experiment consisted of a randomised complete block design with four replications. At full physiological maturity, the roots, shoots and pods samples of the okra plants were collected for analyses. The results showed that OR resulted in a significantly ( $p < 0.05$ ) higher nutrient uptake [N ( $0.0034 \text{ mg kg}^{-1}$ ), K ( $0.0160 \text{ mg kg}^{-1}$ ), Na ( $0.9753 \text{ mg kg}^{-1}$ ), Ca ( $0.0130 \text{ mg kg}^{-1}$ ) and Cu ( $0.01136 \text{ mg kg}^{-1}$ )] in the okra roots than in the other treatments, yet the significantly ( $p < 0.05$ ) highest uptake of P ( $0.0012 \text{ mg kg}^{-1}$ ) was obtained with the MY treatment. Lower values of these nutrient contents were obtained in the shoots. The control treatment gave the significantly highest values of crude fibre (27.33%) and total ash (14.05%), as compared to the other treatments, whereas the other nutritional properties obtained showed no significant difference among any of the treatments. The results indicated that high-quality okra pods could be effectively produced with no soil amendment when planted in soils with a high fertility, such as those treated with sewage sludge.

**Keywords:** Okra, sewage sludge, fertilisers, *Glomus mosseae*, nutrient uptake, nutritional quality.

## 1. Introduction

Okra [*Abelmoschus esculentus* (L.) Moench], a member of the Malvaceae family, is a widely cultivated vegetable crop and very important in the diet of Africans (Omotoso and Shittu, 2008). It is a valuable crop that provides an excellent income and generates other opportunities for small-scale farmers (Selleck and Opena, 1985). Indeed, it is one of the important nutritional vegetable crops cultivated in Nigeria, covering an estimated land area of 1-2 million hectares (FMAWR&RD, 1989). Okra grows in all types of soils and thrives best in a moist, friable, well-drained soil (Kochhar, 1986). The plant is tolerant to drought stress (Majanbu *et al.*, 1985); however, supplementary irrigation may be necessary during extended drought periods for a satisfactory production (Okunade *et al.*, 2009). In Nigeria, the widely cultivated okra is distributed and consumed either fresh (usually boiled, sliced or fried) or in a dried form (Fatokun and Chedda, 1983).

The approximate nutrient content of the edible okra pod is as follows: water, 88%; protein, 2.1%; fat, 0.2%; carbohydrate, 8.0%; fibre, 1.7% and ash, 0.2% (Tindall, 1983). However, the nutritional quality of okra can be influenced by the application of organic fertilisers, such as liquid seaweed, with the following composition, according to Zodape *et al.* (2008): carbohydrate, 7.39%; protein, 28.04%; and dietary fibre, 35.55%. The oil content in the seeds could be as high as that in poultry eggs and soybeans (Akinfasoye and Nwanguma, 2005). The young leaves from okra plants, when prepared together with perfectly ground melon, become a delicious delicacy called 'ilasa', a local soup mostly eaten with 'amala', another local food prepared from yam flour, which is very popular among the Yorubas in Nigeria.

The indiscriminate discharge of sewage sludge into open lands and flowing streams, with its con-

comitant degradation of the ecosystems, is a serious problem in many countries (Kakulu and Osibanjo, 1992). This practice is compounded in countries, such as Nigeria, where little or no treatment is performed on material prior to the discharge (Asia *et al.*, 2006). Furthermore, due to the scarcity, high cost and arbitrary use of inorganic fertilisers in Nigeria (Adewole and Adeoye, 2008), there is inadequate information on the proper application rate of inorganic fertilisers to achieve a high crop productivity of okra (Awe *et al.*, 2006). In addition, organically fertilised okra is more acceptable than the chemically cultivated counterparts (Taiwo *et al.*, 2002). Moyin-Jesu (2007) reported the superiority of an organic-based fertiliser over an inorganic fertiliser, both in quality and quantity, suggesting that the consumers will gain more minerals in their meals of okra and spend less money on purchasing vitamins and minerals to meet health requirements. Shaheen *et al.* (2007) also concluded that the application of bio-fertiliser, bacterial microorganisms in combination with chemical nitrogen fertiliser, in okra cultivation gave better nutritional values in the pod tissues compared with nitrogen fertiliser application alone. Another report (Ansari and Sukhraj, 2010) summarised the importance of the bio-crop cultivation of okra as helping to improve the health of the soil.

Despite the numerous advantages of okra and the influence of fertilisers on its productivity and quality, there is a dearth of information on the nutrient uptake and its nutritional quality when bio-fertilisers, such as compost organic fertiliser and *Glomus mosseae* (an arbuscular mycorrhiza), are applied to okra cultivation on sewage sludge-treated soils. This study, therefore, sought to assess the effects of different soil amendments on the nutrient uptake and nutritional quality of okra grown in a soil treated with sewage sludge.

## 2. Materials and methods

### *Location of the study and experimental design*

The experiment was conducted on a clay loam Alfisol soil during the wet season of 2010 on a manually cleared piece of land at latitude 07°30.362'N and longitude 004°30.747'E on the campus of Obafemi Awolowo University (OAU), Ile-Ife, Nigeria. Ile-Ife is located within the tropical rain forest of Southwestern Nigeria. The plot of land was believed to have been minimally contaminated with the heaps of sewage sludge evacuated from the two oxidation ponds on the campus of OAU, Ile-Ife. The sewage sludge was a product of domestic/kitchen wastes, predominantly from the students' hostels and the staff quarters of the OAU and channelled into the two oxidation ponds. These wastes were allowed to oxidise inside the ponds before dislodgement to the open land near the ponds by the university authorities. This dislodgement is performed once every ten years.

The experiment consisted of four 19 m x 2 m blocks; each block was divided into four plots of 4 m x 2 m, with an alley of 0.5 m between the blocks and 0.5 m within the plots. The experiment was laid out in a randomised complete block design (RCBD), with four treatments, and each treatment was replicated four times. The treatments consisted of a fertiliser application to the crop with (a) 6.0 t ha<sup>-1</sup> compost organic fertiliser (OR), (b) 0.20 t ha<sup>-1</sup> NPK 12-12-17 (IO), (c) 6.0 t ha<sup>-1</sup> *Glomus mosseae* mycorrhiza (MY) or (d) a control without any amendment (CT). Both the NPK and compost organic fertilisers were purchased at a local market. The OR had the following composition: N 6.4%, P 6.4%, K 4.6%, Ca 12.4% and Mg 5.9%. The okra was planted at four (4) seeds per hole using a 60 cm x 30 cm planting distance, and the soil amendments were applied at sowing. For the OR and IO treatments, a band fertiliser application method was

used, whereas for the MY treatment, an inoculum of soil that contained spores, hyphae and maize roots infected with *Glomus mosseae* was applied directly into the sowing holes. The seedlings were thinned to two stands per hole at 2 weeks after planting (WAP) to give 111,111 plants/ha. The plots were manually weeded at 2, 5 and 7 WAP using a hand-held hoe. The harvest of the okra pods began 45 days after planting and continued until 15 WAP, when the experiment was terminated. A repeat experiment was conducted on the same plot of land during the wet season of 2011.

### *2.1. Soil sampling and sample analysis*

Four composite surface soil samples (0-15 cm) were collected using a simple random technique before sowing; the samples were air-dried and sieved through a 2-mm mesh prior to analysis. The soil pH was potentiometrically determined in a 1:1 soil:water ratio (McLean, 1982). The soil organic carbon was determined using the Walkley-Black method (Nelson and Sommers, 1982), and the total nitrogen of the soil was determined using the macro-Kjeldahl method (Bremner and Mulvaney, 1982). The available phosphorus in the soil was determined using the Bray P1 method (Olsen and Sommers, 1982), and the Ca<sup>2+</sup>, Mg<sup>2+</sup>, Na<sup>+</sup> and K<sup>+</sup> were determined using 1 M NH<sub>4</sub>OAc (ammonium acetate) buffered at pH 7.0 as the extractant (Thomas, 1982). The Ca<sup>2+</sup> and Mg<sup>2+</sup> concentrations in soil extracts were measured using a Perkin-Elmer Model 403 (Shelton, Connecticut, USA) Atomic Absorption Spectrophotometer (AAS) and the Na<sup>+</sup> and K<sup>+</sup> concentrations were measured using a Gallenkamp Flame photometer. The exchangeable acidity (H<sup>+</sup> + Al<sup>3+</sup>) in the soil was extracted with 1 M KCl (Thomas, 1982), and the extract was titrated with 0.05 M NaOH using phenolphthalein as the indicator (Odu *et al.*, 1986).

The main soil properties are summarised in Table 1. The mean soil pH (1:1 soil:water) was 5.50, indicating an acidic soil condition. Some of the other mean values obtained were as follows: total nitrogen, 3.85 g kg<sup>-1</sup>; organic carbon, 9.92 g kg<sup>-1</sup> and available phosphorus, 74.25 mg kg<sup>-1</sup>. These values rated high in the classes of soil fertility in Nigeria (Singh, 2002). The values obtained for Cd, Pb and Cu were 0.01, 0.45 and 0.45 mg kg<sup>-1</sup>, respectively.

**Table 1.** Mean values of selected soil properties of the experimental site before planting (2010 and 2011).

Property	Value
pH (1:1 soil-water)	5.50
Organic carbon (g kg <sup>-1</sup> )	9.92
Total nitrogen (g kg <sup>-1</sup> )	3.85
Available phosphorus (mg kg <sup>-1</sup> )	74.25
Exchangeable cations (cmol kg <sup>-1</sup> )	
Na <sup>+</sup>	3.34
K <sup>+</sup>	7.50
Ca <sup>++</sup>	0.50
Mg <sup>++</sup>	0.25
Exchangeable acidity (cmol kg <sup>-1</sup> )	2.90
Heavy metals (mg kg <sup>-1</sup> )	
Cd	0.01
Pb	0.45
Cu	0.45
Textural class	Clay loam

## 2.2. Shoots and roots analyses

Fifteen weeks after sowing, when the experiment was terminated, shoot samples were randomly collected per treatment plot (from all the replicates) using a clean knife, and the roots were carefully dug up with a hand-held hoe. The plant tissues were separated into root and shoot, thoroughly washed with distilled wa-

ter, oven-dried at 70 °C for 48 hours and ground using a Thomas stainless-steel milling machine.

The total nitrogen in the root and shoot samples of the okra plants were determined by the micro-Kjeldahl method and phosphorus by the vanadomolybdate method (Juo, 1979). Concentrated HNO<sub>3</sub> and HClO<sub>4</sub> (5 ml) in a ratio of 2:1 were used to digest 0.5 g of each sample for 2 hours at 150 °C using a Tecator block digester. These samples were allowed to cool, and each was diluted with distilled water to bring the volume to 25 ml. The concentrations of Ca and Mg in each plant digestion sample were measured using an AAS, whereas the concentrations of Na and K were measured using a flame photometer. The heavy metal (Cu, Pb and Cd) concentrations in the roots and shoots were determined using 5 ml of a mixture of concentrated HNO<sub>3</sub> and HClO<sub>4</sub> in a ratio of 2:1 and 5 ml of concentrated H<sub>2</sub>SO<sub>4</sub> to digest 0.5 g of each sample for 2 hours at 150 °C using a Tecator block digester. These samples were allowed to cool, and each was diluted with distilled water to bring the volume to 25 ml. The concentrations of the Cu, Pb and Cd in the extracts were measured using an AAS. The nutrient concentrations obtained were multiplied by the mean dry weights of the roots and shoots to obtain the nutrient uptake per treatment.

## 2.3. Mycorrhizal infection determination

Portions of the roots were collected, using a clean knife from all of the replicates, in McCartney bottles for the mycorrhizal infection determination before the grinding and storage in 50% ethanol. The initiation of the mycorrhizal staining in the roots was achieved by heating the root samples in 10% KOH, rinsing with distilled water and soaking in 1% HCl for 10 minutes. The Trypan blue solution used to stain the roots was prepared by mixing 600 ml of glycerol, 450 ml of distilled water, 50 ml of 1% HCl and 0.05% of Trypan blue in a water bath at 90 °C for 1 hour. The roots were

soaked in the Trypan blue solution for 2 hours, and the stained roots were destained with 50% glycerol. The grid-intersect method of Giovanetti and Mosse (1980) was used to evaluate the percentage of root infection.

#### 2.4. Fruit analysis

Manual harvesting of the okra fruits began from 45 days after planting until 15 WAP, when the experiment was terminated. Each harvest of okra fruits per plot (at every five days after the first set of matured fruits) were weighed, oven-dried at 70 °C for 48 hours and ground using a stainless-steel milling machine. The nutritional content of the okra (crude protein, fat, carbohydrate, total sugars, reducing sugars and vitamin C) and the fruit moisture, crude fibre, dry matter, total ash contents and juice pH were then determined (AOAC, 1990).

#### 2.5. Statistical analysis

One-way analysis of variance (ANOVA) was carried out at a 0.05 level of significance on the data, following the procedure of Gomez and Gomez (1984), and the significant means were compared using the Duncan's Multiple Range Test ( $p < 0.05$ ).

### 3. Results and discussions

#### 3.1. Root nutrients and heavy metal content

The effect of the different treatments on the root nutrients and heavy metal content is presented in Table 2. All nutrient concentrations were significantly ( $p < 0.05$ ) influenced by the application of the soil amendments. The organic fertilisation (OR) resulted in significantly ( $p < 0.05$ ) higher Ca, K, Na and N concentrations than the inorganic (IO) or mycorrhizal (MY) applications. The control treatment (sewage sludge only) gave the lowest concentrations of Ca, Mg, Na, P and Cu compared to the other treatments. The increase in the nutrient contents by the application of organic fertiliser (OR) might be due to their influence in enhancing the reactive surface of the soil (Woomer, 1993). Hence, the highest concentrations of mineralised soil nutrients, such as Ca, K and Na, were taken up by the roots in the plots treated with OR during the growing period. However, the highest concentration of P (0.21 mg P kg<sup>-1</sup>), obtained with the MY application, was significantly ( $p < 0.05$ ) higher than the P concentrations obtained with OR, IO or no soil amendment. The Cd and Pb values were below the detection limits of the technique that was used.

**Table 2.** Effect of soil amendments on the nutrient concentrations in the roots of okra plants (2010 and 2011).

Treatment	Property								
	Ca	Mg	K	Na	N	P	Cu	Cd	Pb
	mg kg <sup>-1</sup>			g kg <sup>-1</sup>		mg kg <sup>-1</sup>			
Mycorrhiza	2.10a	0.58a	1.43c	146.45b	0.32b	0.21a	18.59ab	-	-
Inorganic fertiliser	1.50b	0.44ab	1.53b	154.32c	0.50a	0.13b	18.00b	-	-
Organic fertiliser	2.20a	0.54ab	2.71a	165.30a	0.58a	0.16b	19.25a	-	-
Control	1.45b	0.42b	1.63c	135.55d	0.36b	0.13b	14.54c	-	-

Means followed by the same letter(s) within a column are not significantly different at  $p < 0.05$  by Duncan's Multiple Range Test.

Legend: - = not detected

### 3.2. Shoot nutrients and heavy metal content

The effect of the different treatments on the root nutrients and heavy metal content is presented in Table 3. The data obtained showed that all of the nutrient concentrations in the shoot were significantly affected by the treatments, as compared with the control. The organic fertilisation (OR) and the mycorrhizal appli-

cation (MY) resulted in significantly similar K, P and Na contents, which were higher than the inorganic fertilisation (IO) and the control. In addition, OR significantly ( $p < 0.05$ ) increased the concentration of total N and Cu. However, the Cd and Pb amounts were below the detection limits of the technique. In general, most of the measured nutrients in the shoots were lower than those in the roots.

**Table 3.** Effect of soil amendments on the nutrient concentrations in the shoots of okra plants (2010 and 2011).

Treatment	Property								
	Ca	Mg	K	Na	N	P	Cu	Cd	Pb
	----- mg kg <sup>-1</sup> -----			g kg <sup>-1</sup>		----- mg kg <sup>-1</sup> -----			
Mycorrhiza	1.22a	0.36b	1.05a	100.83a	0.23b	0.12a	10.14b	-	-
Inorganic fertiliser	1.15b	0.48a	0.90a	100.05a	0.46a	0.07b	9.15c	-	-
Organic fertiliser	1.45a	0.47a	1.08a	101.65a	0.56a	0.11a	15.12a	-	-
Control	1.04b	0.32b	0.66b	84.36b	0.32b	0.07b	9.20c	-	-

Means followed by the same letter(s) within a column are not significantly different at  $p < 0.05$  by Duncan's Multiple Range Test.

Legend: - = not detected

### 3.3. The percentage of mycorrhizal infection

The effect of the different treatments on the mycorrhizal root infection is presented in Table 4. The percentage of mycorrhizal colonisation increased significantly under all of the treatments compared to the control. The highest root infection (87.5%) was obtained by mycorrhizal inoculation (MY), whereas the control treatment (CT) gave the lowest colonisation percentage (30.6%). The result of our study agreed with the previous findings of Osonubi *et al.* (1991), who stated that inoculated leguminous woody seedlings had higher mycorrhizal root infections than the non-inoculated seedlings. Moreover, Adewole *et al.* (2010) reported the superiority of inoculated sunflower plants in a higher fungal colonisation and enhanced phyto-remediating potential, as compared to the un-inoculated plants. In the present study, the native fungi may have accounted for

the mycorrhizal colonisation in the un-inoculated plots. However, in both the organic and inorganic fertiliser plots, the added amendments may have enhanced the colonisation of the resident soil fungi on the roots of the okra plants because a low percentage of colonisation was obtained in the plots with zero soil amendment.

**Table 4.** Arbuscular mycorrhiza infection in the roots of okra plants (2010 and 2011).

Treatment	% colonisation
Mycorrhizae	87.5a
Inorganic fertiliser	51.5c
Organic fertiliser	66.4b
Control	30.6d

Means followed by different letter down the column are significantly different at  $p < 0.05$  by Duncan's Multiple Range Test.

### 3.4. Nutrient uptake in the roots and shoots of okra during cultivation

The effect of the different treatments on the nutrient uptake in the roots and shoots of the okra plants 15 WAP is presented in Table 5. The organic fertiliser enhanced the uptake of all of the nutrients evaluated. The organic fertiliser may have enhanced the availability, mobility and uptake of these nutrients. This agreed with the earlier work of Shaheen *et al.* (2007), where organic-based fertiliser was found to enhance the availability and uptake of metals by two cultivars of okra (Balady and Eskandrany) during the spring growing season.

The highest P uptake in the roots of the okra plants may have been enhanced with the *G. mosseae* fungal application. This also agreed with the findings of Lambert *et al.* (1979) and El-Shaikh and Mohammed (2009) that arbuscular mycorrhizae enhanced the uptake of P and several micronutrients, such as Zn, Fe, Mn and Cu. After nutrients are absorbed by the roots, they are transported to other parts of the plants through the xylem (Adewole *et al.*, 2010); however, lower values, except for N and Mg, of these nutrients were obtained in the shoots. The order of nutrient (N, K, Na, Ca, Mg and Cu) uptake in the roots and shoots of okra, as influenced by the soil amendments used, was the following: OR > MY > IO > CT. However, for P uptake, the order was MY > OR > IO > CT.

**Table 5.** Mean dry weight (kg plant<sup>-1</sup>) and nutrient uptake (mg kg<sup>-1</sup>) by the roots and shoots of okra plants during cultivation (2010 and 2011).

Treatment	Mean dry weight	N	P	K	Na	Ca	Mg	Cu
MY	<u>0.57</u>	<u>0.18c</u>	<u>0.12a</u>	<u>0.82b</u>	<u>83.48b</u>	<u>1.20b</u>	<u>0.33a</u>	<u>10.60b</u>
	0.45	0.10c	0.05ab	0.47b	45.37c	0.55b	0.16c	4.56b
IO	<u>0.41</u>	<u>0.21b</u>	<u>0.05c</u>	<u>0.63c</u>	<u>63.27c</u>	<u>0.62c</u>	<u>0.18b</u>	<u>7.38c</u>
	0.47	0.22b	0.03b	0.42b	47.02b	0.54b	0.23b	4.30c
OR	<u>0.59</u>	<u>0.34a</u>	<u>0.09b</u>	<u>1.60a</u>	<u>97.53a</u>	<u>1.30a</u>	<u>0.32a</u>	<u>11.36a</u>
	0.64	0.36a	0.07a	0.69a	65.06a	0.93a	0.30a	9.68a
CT	<u>0.31</u>	<u>0.11d</u>	<u>0.04c</u>	<u>0.51d</u>	<u>42.02d</u>	<u>0.45d</u>	<u>0.13b</u>	<u>4.51d</u>
	0.39	0.12c	0.03b	0.26c	32.90d	0.41c	0.12c	3.59d

Means followed by different letter in the same column (numerators and denominators are separately compared) are significantly different at  $p < 0.05$  by Duncan's Multiple Range Test.

Legend: MY = mycorrhiza, IO = inorganic fertiliser, OR = organic fertiliser, CT = control.

p = Nutrient uptake or mean dry weight in the roots

q = Nutrient uptake or mean dry weight in the shoots

### 3.5. Fruit quality and nutritional value

The effects of the different treatments on the fruit quality characters and nutritional value are presented in Table 6. The fruit dry matter, ether extract, vitamin C, carbohydrate and total sugar contents were not affected by any of the applied treatments, as compared with the control. The mycorrhizal application resulted in lower crude fibre and ash contents than the control,

but the values were similar to the organic and inorganic fertilisation treatments. The inorganic fertilisation treatment produced the lowest crude protein content, when compared with the other treatments. Thus, the cultivation of good-quality okra is achievable in soils rated high in the classes of soil fertility (Singh, 2002); highly fertile soils produced by treatment with sewage sludge may not require additional soil amendment for good-quality okra production.

**Table 6.** The effect of soil amendments on the nutritional quality of okra (2010 and 2011).

Treatment	Property								
	DM	EE	CHO	VIT C	TS	RS	CP	CF	Ash
	----- % -----			mg kg <sup>-1</sup>		----- % -----			
MY	73.87a	15.12a	85.45a	8.54a	7.08a	4.20a	6.24b	22.69c	10.25c
IO	73.61a	15.96a	84.84a	8.76a	7.32a	4.92a	5.28c	25.33b	11.55b
OR	73.70a	15.71a	83.28a	8.54a	7.25a	4.32a	7.35a	25.73b	11.26b
CT	73.61a	15.72a	83.42a	8.88a	7.43a	4.82a	6.86b	27.33a	14.05a

Means followed by the same letter within a column are not significantly different at  $p < 0.05$  by Duncan's Multiple Range Test (DMRT).

Legend:

MY = Mycorrhiza, IO = Inorganic fertiliser, OR = Organic fertiliser, CT = Control, DM = Dry matter, EE = Ether extract, CHO = Carbohydrate, VIT C = Vitamin C, TS = Total sugar, RS = Reducing sugar, CP = Crude protein, CF = Crude fibre.

## 4. Conclusion

We conclude that organic-based fertilisers enhanced the bioavailability and mobility of plant nutrients and, thus, improved the uptake of nutrients by the okra plant roots. Specifically, the compost organic fertiliser enhanced the uptake of nutrients (N, K, Na, Ca and Cu) more than any of the other soil amendments considered. From the present study, it may be deduced that *Glomus mosseae*, an arbuscular mycorrhiza, enhanced the uptake of many of the nutrients, including P, which often remains fixed in the soil. However,

high-quality okra pods were effectively produced when no soil amendment was applied to the soil used because it had a high fertility status produced by the sewage sludge.

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