

Effect of different fertilizers on yield and grain composition of maize in the tropical rainforest zone

Efecto de diferentes fertilizantes sobre el rendimiento y composición del grano de maíz en la zona de selva tropical

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ABSTRACT

This study assessed the quality of selected organic-based fertilizers (OBF) (neem-fortified (NM) and cow dung compost (CD)) and compared them with an inorganic fertilizer (IF) NPK 20-10-10 to determine the growth response and grain composition of maize. The field study was conducted in the early and late cropping seasons of 2015 at the Teaching and Research Farm of the Obafemi Awolowo University, Ile-Ife, Nigeria. The experiment, laid out in a randomized complete block design, consisted of six treatments: 100% NM and 100% CD, each at the rate of 3 and 6 t ha⁻¹, IF at 0.3 t ha⁻¹ (inorganic fertilizer recommendation for local maize production), and zero fertilizer application as control. The highest grain yield of maize (1.87 ± 0.13 t ha⁻¹) was obtained with IF and the lowest one (1.01 ± 0.10 t ha⁻¹) with zero fertilizer application. Maize grain yield from the repeated experiment without treatments applications reduced by about 50 and 75% for OBFs and IF and control plots, respectively. Low crude fiber, 2.62-4.13% obtained using OBFs was a good indicator of maize quality. Organic-based fertilizers demonstrated superior effects on the quality of maize grains when compared to the inorganic fertilizer.

Key words: organic amendment, *Zea mays*, inorganic fertilizer, nutritional content, soil quality, composted manure.

RESUMEN

Este estudio evaluó la calidad de fertilizantes orgánicos seleccionados (FOS) (fortificados con neem (NM) y compost de estiércol de vaca (CD)) y los comparó con un fertilizante inorgánico NPK 20-10-10 (IF) para determinar la respuesta de crecimiento y composición del grano de maíz. El estudio de campo se realizó en las temporadas de cultivo tempranas y tardías de 2015 en la Granja de Enseñanza e Investigación de la Universidad Obafemi Awolowo, Ile-Ife, Nigeria. El experimento, establecido en un diseño de bloques completos al azar, constó de seis tratamientos: 100% NM y 100% CD, cada uno a razón de 3 y 6 t ha⁻¹, IF a 0.3 t ha⁻¹ (recomendación de fertilización inorgánica para la producción local de maíz), y cero aplicación de fertilizantes como control. El mayor rendimiento de grano de maíz (1.87 ± 0.13 t ha⁻¹) se obtuvo con IF y el menor (1.01 ± 0.10 t ha⁻¹) con cero aplicación de fertilizante. El rendimiento de grano de maíz del experimento repetido sin aplicaciones de tratamientos se redujo en aproximadamente un 50 y un 75% para las parcelas FOS e IF y de control, respectivamente. El bajo contenido de fibra cruda, 2.62-4.13% obtenido mediante FOS, fue un buen indicador de la calidad del maíz. Los fertilizantes orgánicos demostraron efectos superiores en la calidad de los granos de maíz en comparación con el fertilizante inorgánico.

Palabras clave: enmienda orgánica, *Zea mays*, fertilizante inorgánico, contenido nutricional, calidad del suelo, estiércol compostado.

Introduction

Soil nutrient depletion is a worldwide environmental challenge having serious negative impact on food security and soil quality. Soil nutrient depletion involves loss of organic matter and reduced soil nutrient levels (Osujieke *et al.*, 2020). This could lead to reduced crop yield and loss of agricultural lands. For sustainable long-term productivity and a better agricultural environment, maintenance of soil quality is necessary (Johnston & Poulton, 2018). Crop production in sub-Saharan Africa (SSA) has faced

various limitations. Among them are climate change and low soil fertility. However, low soil fertility status has been the most challenging in this region for many decades. It has been a major reason for increased food insecurity, which is evident in the declining food production per capita from smallholder farms (Mango *et al.*, 2017; Ayito *et al.*, 2018).

Many factors, both natural and artificial, have been identified as the reasons for such low soil quality. Some of them include soil erosion, overgrazing, and indiscriminate vegetation removal. Other factors that contribute to soil

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nutrient depletion through physico-climatic processes in tropical Africa and particularly SSA include loss of soil nitrogen and phosphorus through wind and water erosion as well as leaching of nitrogen and potassium (Slama *et al.*, 2020). Others are unsustainable tillage practices and continuous cropping with low or no fertilizer inputs. Continuous cropping without fertilizer inputs, or wrong or inadequate fertilizer applications have led to further reduction in soil quality (Choudhary *et al.*, 2018).

Fertilizer is any organic or inorganic material that is added to a soil to supply one or more plant nutrients essential to the growth of plants and also to improve soil condition (Cai *et al.*, 2019). Fertilizer application is an important farming practice. It has led to improved crop production and has increased the acceptance of other sustainable practices that enhance crop production. Hence, fertilizer application has been integrated into many agricultural programs and has become a key element for improving crop production in most countries.

Liu *et al.* (2019) observed that an improvement in soil fertility is needed to increase agricultural productivity. They further observed that enhanced soil fertility will lead to improvement in food security and increased income for many farmers. Therefore, to reduce nutrient depletion in soils, application of fertilizers (organic or inorganic) in required dosages and with appropriate methods are necessary (Adewole & Adeoye, 2008). However, sustaining soil quality depends on the ability to enhance nutrient recycling in soils (Schröder *et al.*, 2016).

Addition of different soil amendments in response to declining soil fertility has been studied by many scientists (Diacono & Montemurro, 2011; Syuhada *et al.*, 2016; Jjagwe *et al.*, 2020). An inorganic fertilizer improves crop yield, soil pH, total nutrient content and increased nutrient availability, while the use of an organic fertilizer, such as manure, leads to improved soil conditions for longer periods of time, particularly with continuous maize crop cultivation (Oladele *et al.*, 2019).

Maize (*Zea mays*) is an important agricultural crop in the SSA and ranks as the most important cereal crop, with Nigeria as the largest African producer (IITA, 2018). It is a staple food for more than 1.2 billion people in SSA and Latin America (IITA, 2018). Its importance could be attributed to its capability to be grown all through the year. Globally, the demand for maize sometimes surpasses supply as a result of the various domestic uses and importance (Ten Berge *et al.*, 2019). In most African countries, maize production

per capita has not been on the same level with the population growth over the past 60 years (Smale & Jayne, 2003). Therefore, with maize a strategic and important crop, its production must be maintained at adequate levels to ensure food security and self-sufficiency at both household and national levels (Santpoort, 2020). Maize is also a crop that requires a high amount of mineral nutrients for its growth and its productivity is largely dependent on soil nutrient management (Kannan *et al.*, 2013). Therefore, there is a need to take appropriate steps to ensure increased maize production by improving the physical and chemical properties of the soil. This can be achieved through appropriate farming practices such as the use of appropriate fertilizers and sound agronomic practices. This study, therefore, compared the efficacy of selected organic and inorganic fertilizers on the growth response and nutrient composition of maize on an Alfisol of a forest ecological zone in Southwestern Nigeria. A drought-tolerant maize variety was used as an improved seed technology to help flatten economic burden often associated with frequent droughts in the study area.

Materials and methods

The study was carried out at the Teaching and Research Farm, Obafemi Awolowo University (OAU), Ile-Ife, Osun State, Nigeria in the early (April - July) and late (August - November) seasons of 2015. The research farm was located at 07°30'0" N and 04°30'0" E, at an elevation of 268 m a.s.l. The study area falls within the lowland tropical rainforest (Adesina, 1989). The experimental sites had a total annual rainfall of 1165.2 mm and an average annual temperature of 35.2°C (Komolafe, 2015). The early seasons have an average temperature of 28.57°C and 141 mm of precipitation, the late seasons have average temperature of 26.49°C and 144.29 mm of precipitation (Climate Change Knowledge Portal, 2021).

Experimental site

The experimental site was cleared manually, and pre-cropped soil samples were collected at a depth of 0-15 cm for analysis. A drought-tolerant maize variety, DT-SYN-8W obtained from the Institute of Agricultural Research and Training, Ibadan, Nigeria was the test crop. Neem-fortified organic fertilizer sourced from Alesinloye Waste Recycling Complex of Ibadan (Nigeria) and fortified with neem leaves and NPK 20-10-10 inorganic fertilizer were procured from an open market in Ibadan. Fresh cow dung was obtained from the Beef Unit of the Teaching and Research Farm, OAU, Ile-Ife and composted aerobically. The experiment consisted of six treatments laid out in a

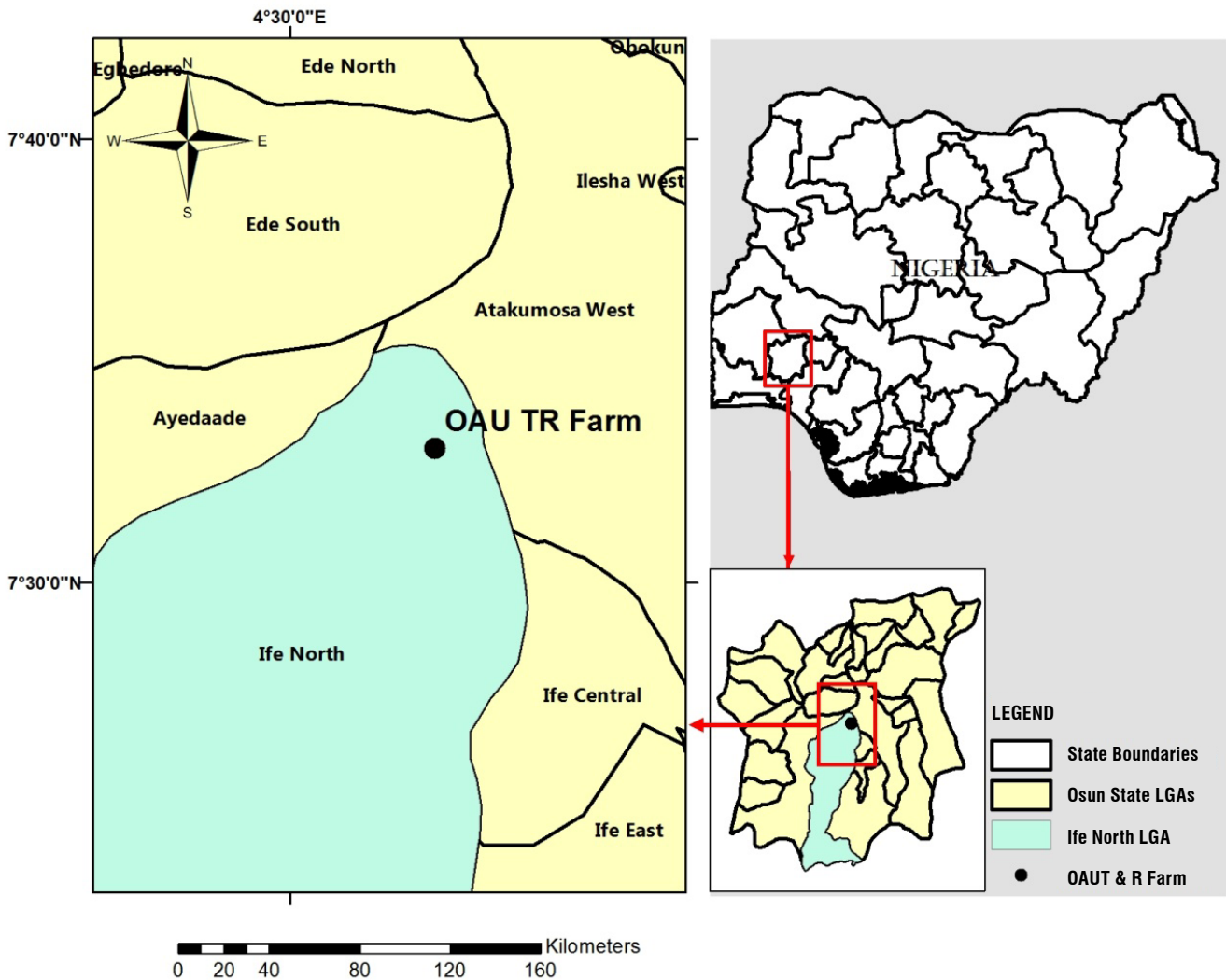


FIGURE 1. Map of Nigeria showing the study area.

randomized complete block design. The treatments were: 100% neem-fortified organic fertilizer (NM) and 100% cow dung compost (CD), each at the rate of 3 or 6 t ha⁻¹, NPK (20-10-10) at 0.3 t ha⁻¹ (inorganic fertilizer recommendation for local maize production) and zero fertilizer application as control.

The experimental site consisted of three 23.0 x 2.5 m blocks; each block was in turn divided into six plots of 3.0 x 2.5 m with an alley of 1.0 m between blocks and 1.0 m within plots. Each treatment plot was replicated thrice to give a total of 18 plots. The test crop was sown at three seeds per hill using 75 x 50 cm planting distance. All the treatments, except NPK (20-10-10), were applied at sowing. The NPK fertilizer was applied two weeks after planting. Maize seedlings were later thinned to two seeds per hole at two weeks after sowing (WAS) to give a total of 53,333 maize

plants per ha. Manual weeding using a handheld hoe was carried out at 2 and 5 weeks after sowing. Maize ears from each treatment plot were harvested, threshed, and stored for grain yield weight determination. The experiment was repeated during the late maize cropping season to identify any improvements or changes in the performance of the crops over time due to the earlier treatments. Post-cropped soil samples were collected immediately after maize harvesting in the late cropping season.

Analysis of soil properties

The following chemical properties of the sampled soil, neem-fortified organic fertilizer and cow dung compost were determined using standard methods (Page *et al.*, 1982). Soil pH was determined in a 1:1 soil to water suspension using the Dwyer model WPH1 waterproof pH

tester. Particle size distribution was determined using the hydrometer method. Soil organic carbon was determined using the Walkley and Black method. The exchangeable cations were determined using 1 M Ammonium acetate buffered at pH 7.0 as extractant. The K⁺ and Na⁺ concentrations in the soil were read on a Gallenkamp flame photometer while Ca²⁺ and Mg²⁺ concentrations in the soil extracts were read on a Perkin-Elmer Model 403 atomic absorption spectrophotometer (AAS). Exchangeable acidity was determined by the titration method after extraction with KCl. Total nitrogen was determined using macro-Kjedahl method. Available phosphorus was determined by the ascorbic acid molybdate blue method as described by Murphy and Riley. Micronutrients (Zn, Mn, Cu, B, and Fe) and As were extracted using 0.1 M HCl (Juo, 1982) and their concentrations in the soil extracts were read on the AAS. Dried maize grains (at 12% percentage moisture) were subjected to proximate composition using the methods from AOAC (1990).

Statistical analysis

Collected data were analyzed using ANOVA (analysis of variance) and their treatment means were calculated by Duncan's Multiple Range Test (DMRT) method using GraphPad Prism 5 and SAS. Descriptive statistics was used to determine the reduction in the grain yield of maize between the two cropping seasons.

Result and discussion

Experimental site and the organic fertilizers used

The soil texture was sandy loam with 792.00, 114.00, and 94.00 g kg⁻¹ of sand, clay, and silt, respectively (Tab. 1). The soil pH (1:1 soil-H₂O) of the experimental site was 7.86, indicating an alkaline condition. Other soil properties included: 22.51 g kg⁻¹ of organic carbon, 2.12 g kg⁻¹ of total nitrogen, 1.63 mg kg⁻¹ of available phosphorus and 21.01 cmol kg⁻¹ of cation exchange capacity, but mostly dominated by Ca²⁺. The micronutrient contents of Zn, Mn, Cu, and Fe were 1.28, 30.80, 1.65, and 141.00 mg kg⁻¹, respectively. The neem-fortified organic fertilizer had 148.88 and 16.50 g kg⁻¹ organic carbon and total nitrogen, respectively, and a carbon-nitrogen ratio of 9.02. Other values of the neem-fortified organic fertilizer were: 31.37 mg kg⁻¹ of available phosphorus, and 13.30, 16.97 and 0.33 cmol kg⁻¹ of available K, Ca and Mg, respectively. The Zn, As and B in neem-fortified organic fertilizer had values 1.73, 2.10 and 1.30 mg kg⁻¹, respectively. Except for the carbon-nitrogen ratio, available P and Mg, all other parameters were lower in the cow dung compost than in the neem-fortified organic fertilizer. In this study, the soil organic carbon and

total nitrogen are considered moderate, according to the ratings of the Developing Agri-input Markets in Nigeria (DAIMINA) (Singh, 2002). These, however, may not be adequate for optimum production of maize, as low grain yield of maize is the current realizable scenario by most resource-poor Nigerian farmers (IITA, 2018).

TABLE 1. Properties of pre-cropped soil and two organic fertilizers used.

Property	Soil	NM	CD
pH (1:1 soil-water)	7.86	-	-
Organic carbon (g kg ⁻¹)	22.51	148.88	135.37
Total N (g kg ⁻¹)	2.12	16.50	11.60
C/N	-	9.02	11.67
Available P (mg kg ⁻¹)	1.63	31.37	35.15
Exchangeable cations (cmol kg⁻¹)			
Na ⁺	0.27	-	-
K ⁺	0.20	13.30	1.45
Ca ²⁺	20.15	16.97	1.11
Mg ²⁺	0.39	0.33	0.58
CEC	21.01	-	-
Exchangeable acidity (cmol kg ⁻¹)	0.40	-	-
Zn (mg kg ⁻¹)	1.28	1.73	0.05
Mn (mg kg ⁻¹)	30.80	-	-
Cu (mg kg ⁻¹)	1.65	-	-
As (mg kg ⁻¹)	-	2.10	1.30
B (mg kg ⁻¹)	-	1.30	0.80
Fe(mg kg ⁻¹)	141.00	-	-
Sand (g kg ⁻¹)	792.00	-	-
Clay (g kg ⁻¹)	114.00	-	-
Silt (g kg ⁻¹)	94.00	-	-
Textural class	Sandy loam	-	-

NM = Neem-fortified organic fertilizer, CD = Cow dung compost.

Shehu *et al.* (2018) and Lucas *et al.* (2019) worked extensively on nutrient requirements for enhanced grain yield of maize in Nigeria and Brazil, respectively. These authors observed that near moderate soil organic matter can be related to inherently high sandy nature of the parent material and low capacity to store carbon. This could also be the reason for low total N and cation exchangeable capacity, as soil organic carbon plays a vital role in soil fertility maintenance. The neem-fortified organic fertilizer with low C/N ratio had better opportunity to mineralize and release its essential nutrients for maize use faster than cow dung compost with higher C/N ratio. Syuhada *et al.* (2016) observed a similar scenario in their study, where biochar with small fractions of carbon mineralized later than synthetic fertilizers with fast release of nutrients. Seman-Varner *et al.* (2019) and Jjagwe *et al.* (2020) also observed better response from low

C/N ratio amendments of plant- and animal-based manure than those with high C/N ratio.

Effects of organic and inorganic fertilizer applications on grain yield

The effects of organic and inorganic fertilizer applications on the grain yield and percentage reduction of maize during the two cropping seasons are presented in Table 2. The application of NPK 20-10-10 gave significantly ($P < 0.05$) highest grain yield, 1.87 t ha^{-1} ; while the control plot had the least significant grain yield (1.01 t ha^{-1}) of maize from the early cropping season. Except for NPK 20-10-10 and control plots that had reduced grain yield of maize by over 75%, other plots with organic-based fertilizers had about 50% reduction of the grain yield of maize in the late cropping season.

The inorganic fertilizer NPK 20-10-10 gave significantly highest grain yield of maize during the early cropping season because of its ability to release nutrient elements faster than most organic-based fertilizers, resulting in high maize grain yield in the early cropping season. The neem-fortified organic fertilizer that had higher maize grain yield than cow dung compost could be due to the faster mineralization and releasing tendency in neem-fortified organic fertilizer. This agrees with Slomon *et al.* (2018), who stated that breakdown and mineralization of the neem amendment result in the release of nutrients to the soil, thus, improving the soil nutrients and subsequently improving the crop yield. The higher the quantity of organic-based fertilizer used, the higher the grain yield of maize. During the repeat experiment in the late cropping season, about 50% reduction was observed with organic-based treatment applications, while over 75% reduction was observed with NPK 20-10-10 and zero treatment applications. Lower percentage reduction obtained with organic-based fertilizers was due to their nutrient slow-releasing effect in maize crop production.

Effects of organic and inorganic fertilizer applications on the grain composition and grain yield of maize

The compositions of the harvested maize grain in the early and late cropping seasons are presented in Table 3. Crude protein 8.12-11.99%, crude fat 2.16-4.13%, reducing sugar 3.27-7.08%, vitamin C 2.86-3.85%, and crude fiber 2.62-4.13% were enhanced with the addition of organic-based fertilizers, compared to the inorganic fertilizer or zero treatment application. Except for crude fiber, lower values were obtained with inorganic fertilizer and zero treatment applications.

In this study, the early maize cropping season produced better grain quality than the late cropping season. The early maize cropping season generally occurs during the rainy period, when more soil moisture will be available for nutrient element mobility and their uptake by maize plants than in the late cropping season. Maize grains from early cropping season had higher crude fiber and vitamin C than maize grains from the late cropping season, and these might have been influenced by higher nutrient element mobility and their uptake by the maize plant. Low crude fiber is useful in quality assessment of maize grain as this enhances the utilization of nitrogen and accumulation of micronutrients useful for human nutrition (Obinna-Echem *et al.*, 2018).

Effects of organic and inorganic fertilizer applications on post-cropped soil properties of the maize field are presented in Table 4. Most of the soil parameters, such as organic carbon, total nitrogen, available phosphorus, and CEC, increased after maize harvesting of the late cropping season when organic-based fertilizers were applied to soil. These soil parameters, however, decreased with NPK 20-10-10 and zero treatment applications. Also, the soil parameters increased with increase in treatment applications. The soils became more acidic after the late maize cropping season.

TABLE 2. Mean (\pm SD) grain yield (t ha^{-1}) and percentage reduction of maize during two cropping seasons.

Treatment	Early cropping season	Late cropping season	Percentage reduction
NM3	$1.27 \pm 0.13 \text{ ab}$	$0.64 \pm 0.15 \text{ a}$	49.6
CD3	$1.10 \pm 0.15 \text{ ab}$	$0.50 \pm 0.14 \text{ a}$	54.5
NM6	$1.53 \pm 0.17 \text{ ab}$	$0.77 \pm 0.13 \text{ a}$	49.7
CD6	$1.42 \pm 0.23 \text{ ab}$	$0.72 \pm 0.13 \text{ a}$	49.3
NPK 20-10-10	$1.87 \pm 0.13 \text{ a}$	$0.40 \pm 0.15 \text{ ab}$	78.6
Control	$1.01 \pm 0.10 \text{ b}$	$0.25 \pm 0.10 \text{ b}$	75.2

NM3 = Neem-fortified organic fertilizer at 3 t ha^{-1} , CD3 = Cow dung compost at 3 t ha^{-1} , NM6 = Neem-fortified organic fertilizer at 6 t ha^{-1} , and CD6 = Cow dung compost at 6 t ha^{-1} . Mean values with the same letter(s) down the column are not significantly different by Duncan's Multiple Range test at $P < 0.05$.

TABLE 3. Grain composition of maize harvested in the early and late cropping seasons.

Treatments	Crude protein (%)	Crude fat (%)	Crude fiber (%)	Ash (%)	Moisture (%)	Total sugar (%)	Reducing sugars (%)	Vitamin C (%)
Early cropping season								
NM3	9.74 a	4.10 a	2.92 b	4.24 a	13.06 a	10.40 a	7.08 a	3.85 a
CD3	8.12 b	3.42 a	3.93 b	3.54 a	10.89 c	8.67 bc	5.90 bc	3.21 ab
NM6	10.45 a	4.13 a	4.10 a	4.72 a	13.30 a	11.87 a	8.00 a	4.05 a
CD6	8.24 b	3.48 a	4.13 a	3.63 a	10.23 c	9.13 ab	6.16 b	3.12 b
NPK 20-10-10	5.96 d	2.45 c	5.11 a	2.61 c	7.55 d	6.59 c	4.45 c	2.28 c
Control	8.86 c	3.21 b	4.95 a	3.82 b	11.53 b	9.61 b	6.28 b	3.01 b
Late cropping season								
NM3	10.81 a	2.59 a	2.62 b	2.51 a	10.81 b	11.78 a	3.92 c	3.43 a
CD3	9.01 b	2.16 a	3.24 a	2.09 a	9.01 bc	9.82 b	3.27 d	2.86 b
NM6	11.99 a	3.03 a	3.28 a	2.74 a	13.61 a	12.87 a	6.11 a	3.81 a
CD6	9.23 b	2.33 a	3.36 a	2.11 a	10.12 b	9.90 b	4.70 b	2.93 b
NPK 20-10-10	7.38 d	1.86 c	4.26 a	1.69 b	8.10 c	7.92 c	3.76 cd	2.34 c
Control	8.05 c	2.20 ab	4.03 a	2.08 a	10.86 b	8.21 bc	3.94 c	3.21 ab

NM3 = Neem-fortified organic fertilizer at 3 t ha⁻¹, CD3 = Cow dung compost at 3 t ha⁻¹, NM6 = Neem-fortified organic fertilizer at 6 t ha⁻¹, CD6 = Cow dung compost at 6 t ha⁻¹. Mean values with the same letter(s) in the same column are not significantly different by Duncan's Multiple Range Test at $P < 0.05$.

TABLE 4. Effects of organic and inorganic fertilizers on post-cropped soil properties.

Property	NM3	CD3	NM6	CD6	NPK 20-10-10	Control
pH	7.56±0.12	7.60±0.15	7.63±0.17	7.65±0.05	7.33±0.17	7.03±0.07
Organic C (g kg ⁻¹)	42.63±1.04	33.81±0.85	49.46±0.64	43.16±1.00	19.43±0.27	13.59±0.11
Total N (g kg ⁻¹)	4.12±0.27	3.25±0.20	4.61±0.19	3.72±0.27	1.85±0.25	1.22±0.15
Available P (mg kg ⁻¹)	4.84±0.23	2.99±0.18	5.57±0.25	4.64±0.15	3.13±0.17	2.98±0.13
Exchangeable bases (cmol kg⁻¹)						
Na ⁺	0.54±0.06	0.41±0.05	0.65±0.05	0.54±0.06	0.36±0.04	0.33±0.07
K ⁺	0.44±0.04	0.34±0.04	0.34±0.05	0.29±0.01	0.34±0.06	0.23±0.07
Ca ²⁺	6.02±0.15	4.63±0.14	5.99±0.11	4.99±0.11	3.32±0.10	3.15±0.10
Mg ²⁺	0.68±0.15	0.52±0.11	0.67±0.13	0.56±0.14	0.36±0.04	0.30±0.05
Micronutrients (mg kg⁻¹)						
Zn	1.82±0.28	1.40±0.20	1.56±0.22	1.30±0.10	1.20±0.05	1.15±0.05
Mn	30.11±2.25	23.16±1.45	29.64±0.45	24.70±0.24	17.28±0.12	16.20±0.17
Cu	1.50±0.05	1.16±0.02	1.36±0.04	1.13±0.07	0.95±0.05	0.95±0.04
Fe	132.90±3.25	102.20±2.80	107.60±3.20	89.80±2.20	90.67±1.13	86.50±1.77

NM3 = Neem-fortified organic fertilizer at 3 t ha⁻¹, CD3 = Cow dung compost at 3 t ha⁻¹, NM6 = Neem-fortified organic fertilizer at 6 t ha⁻¹, and CD6 = Cow dung compost at 6 t ha⁻¹.

Conclusions

Organic-based fertilizers vary in nutrient composition and mineralization rate depending on their organic materials make-up. The two organic-based (neem-fortified and cow dung compost) fertilizers have longer-lasting and more positive effects on soil properties, particularly soil organic carbon, total nitrogen, and CEC than the

inorganic fertilizer (NPK 20-10-10). Also, neem-fortified organic fertilizer demonstrated superior influence on soil properties and grain yield of maize, and there was a direct relationship on the quantity of added fertilizer and grain yield of maize. Organic-based fertilizers demonstrated superior effects on the quantity and quality of maize grains than the inorganic fertilizer.

Conflict of interest statement

The authors declare that there is no conflict of interests regarding the publication of this article.

Author's contributions

OK and MA designed the experiments. OK conducted the research process, specifically performing the experiments and data collection, applied statistical techniques to analyze study data, and wrote the original draft. MA oversaw and led the research activity planning and execution and verified the overall replication/reproducibility of results/experiments and other research outputs. MA also revised the manuscript. All authors reviewed the final version of the manuscript.

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