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(RESEARCH ARTICLE)

Enhancing nutritional quality of *Amaranthus cruentus* (L.) with fertilisers and salicylic acid

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# Abstract

Globally, deficiency of essential vitamins and minerals in major staple food crops is a great challenge to human health. The use of fertilisers and elicitors has been suggested as a sustainable approach to enhance food quality. Therefore, this study investigated the effects of compost at 5 and 10 t/ha, poultry manure at 5 and 10 t/ha, and NPK 15:15:15 fertiliser applied at rates of 50 and 100 kg N/ha, Salicylic Acid at 125 and 250  $\mu$ M/L, and their combinations on the growth and food quality of *Amaranthus cruentus*, a globally consumed vegetable. The experiment was conducted in a screenhouse and arranged in a completely randomized design with three replicates. Data were collected on marketable yield, dry matter, chlorophyll, folate, zinc, iron, crude fibre, ash, and moisture contents. SA<sub>250</sub>+NPK<sub>50</sub> enhanced the marketable yield by 88.0%, and SA<sub>125</sub>+NPK<sub>100</sub> enhanced dry matter accumulation by 92.6% compared to the control. The combination of SA<sub>250</sub>+NPK<sub>50</sub> had higher folate (285.00  $\mu$ g/100g) and iron (348.00 mg/kg) contents than control (125.00 mg/kg and 138.00 mg/kg, respectively), while zinc (47.00 mg/kg) was highest in *Amaranthus cruentus* treated with SA<sub>250</sub>+CP<sub>10</sub>. In addition, *Amaranthus cruentus* treated with SA<sub>125</sub>+PM<sub>5</sub> had significantly higher crude fibre (13.10%) and ash (12.35%) contents than the control. The combined application of 250  $\mu$ M/L salicylic acid and 50 kg N/ha NPK fertiliser improved the yield and quality of *Amaranthus cruentus*, especially in terms of folate, iron, and zinc, which are important micronutrients for human health.

Keywords: Amaranthus cruentus; Micronutrient malnutrition; Fertiliser; Salicylic acid; Food Security; Human health

# 1. Introduction

The majority of commonly consumed agricultural food crops are deficient in essential vitamins and minerals, which can adversely affect human health [1]. Micronutrient deficiency is a global health problem that affects people of all socioeconomic statuses. This occurs when the diet does not provide adequate amounts of essential vitamins and minerals for optimal health and development. According to [2], approximately one-third of the world's population is malnourished, with Africa having high prevalence of micronutrient deficiency. This deficiency can lead to increased mortality, reduced cognitive function, and increased vulnerability to infectious diseases. In Nigeria, micronutrient deficiency is a major cause of stunting and underweight among children under five years old, as well as acute malnutrition among women of reproductive age. Nigeria has the second highest rate of stunting in the world, affecting 32% of children under five years of age, where 20% are underweight [3]. Micronutrient deficiency poses a serious threat to human health [4]. Therefore, the quality of food crops is important for ensuring food security.

Food security is not only about producing enough food, but also ensuring that the food is nutritious and meets the dietary needs of people. The agricultural sector is more concerned with increasing food quantity than food quality [5, 6]. The gap between quantity and quality must be bridged to improve global food security. However, agronomic practices, environmental factors, and crop species determine the quality of the crops produced. For example, the use of

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different fertilisers affects the types of nutrients, availability, and uptake by crops. All these, in turn, determine the nutrient content of the crop produced. The application of organic fertilisers has been reported to make all required nutrients available to crops in the right proportion [7]. Organic and inorganic fertilisers can provide plants with all necessary nutrients, including micronutrients, such as iron and zinc [8]. For example, the micronutrient content of maize treated with compost was higher than that of plants treated with inorganic fertilisers [9].

Similarly, application of elicitors such as salicylic acids and jasmonates have been shown to increase the production of folate and other vitamins in crops [10, 11]. Elicitors are substances released from plants by the action of pathogens and can stimulate plant defense. Elicitors can be used to boost plant secondary-metabolite synthesis in crops, which serves as an alternative method where the high cost of research associated with metabolic engineering can be avoided.

Leafy vegetables are rich in nutrients [12] and are vital to human health. One of the most popular leafy vegetables in Nigeria is *Amaranthus cruentus* [13]. However, information on folate and mineral contents is limited and has not been adequately studied. To address the health challenges caused by micronutrient deficiency, it is essential to conduct more research on the micronutrient content and nutritional quality of *Amaranthus cruentus* and to devise sustainable ways to increase its folate and mineral levels. This could help to eliminate micronutrient malnutrition in Nigeria. Therefore, this study compared the effects of different types of fertilisers and salicylic acid on the micronutrient content and nutritional quality of *Amaranthus cruentus*.

# 2. Material and methods

The study was conducted in the screenhouse of the Department of Crop Protection and Environmental Biology at the University of Ibadan, Nigeria. The fertilisers used were: poultry manure (PM), compost (CP), and inorganic fertiliser (NPK). Salicylic acid was used as the elicitor and applied at concentrations of 125 and 250  $\mu$ M/L. Cured poultry manure was obtained from the Teaching and Research Farm of the University of Ibadan, Nigeria. NPK (15:15:15) was obtained from the Department of Agronomy, University of Ibadan, Nigeria. Composting was performed from the shoot of Tithonia diversifolia (L) and fresh poultry manure mixed at a ratio of 3:1 using the heap method. It was left to decompose for three months with continuous turning and watering every week. The matured compost was air-dried and ground to ensure uniformity [14]. Compost and poultry manure were applied at 5 and 10 t/ha, respectively, whereas NPK was applied at 50,100 kg N/ha. The experiment comprised of 21 treatments; 2 sole salicylic acid; Salicylic acid at 125  $\mu$ M (SA125), Salicylic acid at 250 µM (SA250); 6 sole fertilisers treatments; NPK at 50 kg N/ha (NPK50), NPK at 100 kg N/ha (NPK100), Poultry manure (PM) at 5 t/ha (PM5), Poultry manure (PM) at 10 t/ha (PM10), Compost (CP) at 5 t/ha (CP5), Compost (CP) at 10 t/ha (CP<sub>10</sub>): 6 combinations of SA<sub>125</sub> with fertilisers; SA<sub>125</sub> + CP<sub>5</sub> (125  $\mu$ M SA + 5 t/ha CP), SA<sub>125</sub> + CP<sub>10</sub> (125 μM SA + 10 t/ha CP), SA<sub>125</sub> + NPK<sub>50</sub> (125 μM SA + 50 kg N/ha NPK), SA<sub>125</sub> + NPK<sub>100</sub> (125 μM SA + 100 kg N/ha NPK), SA125 + PM5 (125 µM SA + 5 t/ha PM) and SA125 + PM10 (125 µM SA + 10 t/ha PM), and 6 combinations of SA250 with fertilisers; SA250 + CP5 (250 µM SA + 5 t/ha CP), SA250 + CP10 (250 µM SA + 10 t/ha CP), SA250 + NPK50 (250 µM SA + 50 kg N/ha NPK), SA<sub>250</sub> + NPK<sub>100</sub> (250 μM SA + 100 kg N/ha NPK), SA<sub>250</sub> + PM<sub>5</sub> (250 μM SA + 5 t/ha PM) and SA<sub>250</sub> + PM<sub>10</sub> (250 µM SA + 10 t/ha PM) and a control (No treatment). The treatments were arranged in a completely randomized design (CRD) with three replicates. Amaranthus cruentus seeds obtained from the National Institute of Horticultural Research (NIHORT) Ibadan, Nigeria was used in this study.

## 2.1. Experimental procedures

The experimental pots (5- litre capacity) were filled with 5 kg of dry soil. Compost and poultry manure were applied to the soil receiving organic fertiliser treatments one week before sowing, whereas NPK was applied to the pots receiving NPK treatments two weeks after sowing. *Amaranthus cruentus* seeds were sown directly into pots. Two plants per bag were thinned 2 weeks after sowing (WAS).

## 2.2. Preparation of salicylic acid

Salicylic acid was first dissolved according to the procedure described in [15] in 1 ml of absolute ethanol and added dropwise to 1 L of distilled water (ethanol/water, 1/1000 v/v), and three drops of Tween 20 were added as a surfactant. The *Amaranthus cruentus* plants were sprayed at 3, 4, and 5 WAS according to the treatment layout using the foliar method. Harvesting was done six weeks after sowing, and yield parameters, such as shoot fresh weight and dry matter, were determined following standard procedures. Chlorophyll content was measured using a SPAD-502 meter. Folate, nitrogen, phosphorus, potassium, zinc, iron, and proximate contents were determined in *A. cruentus* leaves after harvesting.

### 2.3. Chemical analysis

Folate was determined by weighing 20 mg of plant sample into a beaker, and 10 ml of 0.1 M HCL was added. The mixture was shaken on a mechanical shaker for 15 minutes and filtered using 2 mm filter paper. Two (2) ml of the sample solution was placed in marked test tubes. In each test tube, 2 ml of 0.02% potassium permanganate solution, 2 ml of 2% sodium nitrate solution, 2 ml of 4 M hydrochloric acid solution, 1 ml of 5% ammonium sulphamate solution and 1 ml of dye solution (0.1 % N, N diethyl aniline dye solution in iso propyl alcohol) were added and mixed thoroughly. The absorbance was recorded at 535 nm [16, 17, 18]. Mineral elements were determined by weighing the milled plant sample (0.5 g) into a beaker and 5 ml of an acid mixture of nitric perchloride acid in a ratio 2:1 was added. The beaker was then placed on a hot plate at 70°C for 45 minutes on a fume cupboard. The ash was boiled in 20% HCL in a beaker. The content was filtered into a 100 ml volumetric flask. This was marked with distilled water. Na and K were read on a flame emission photometer, and P, Mg, Ca, and Fe were read spectrophotometrically at 470 nm [19, 20]. Total nitrogen was determined using a spectrophotometer [21, 22], and percentage moisture, crude fibre, and ash contents were determined using standard procedures [20]. Data on soil and plants were analyzed by analysis of variance (ANOVA) using the SAS code. Means were separated using Duncan's multiple range test (P=0.05).

# 3. Results

# 3.1. Effects of fertilisers and salicylic acid on fresh weight, dry matter and chlorophyl content of Amaranthus cruentus

#### 3.1.1. Fresh weight (g/plant)

Generally, the fresh weight of *Amaranthus cruentus* were significantly (P<0.05) enhanced by fertiliser application, higher fresh weight (55.63 g), when compared to control (40.00 g) was obtained in PM<sub>10</sub>, though across all the sole treatment applications it was not significantly different from all fertiliser treatments except in CP<sub>5</sub>. Combined application of fertiliser and salicylic acid shows the highest fresh weight (75.20 g) of *Amaranthus cruentus* was obtained in SA<sub>250</sub> +NPK<sub>50</sub> and was similar to 74.68 g fresh weight obtained in *Amaranthus cruentus* treated with SA<sub>125</sub> +NPK<sub>50</sub>, while the lowest fresh weight (40.0 g) was obtained in control (Table 1)

#### 3.1.2. Dry matter (g/plant)

NPK<sub>100</sub> enhanced dry matter (5.96 g) more than the control (3.12 g). This was closely followed by the 5.27 g obtained from *Amaranthus cruentus* treated with PM<sub>10</sub>. In the combined application, SA<sub>125</sub> +NPK<sub>100</sub> had the highest dry matter (6.01 g), which was similar to 5.52 g obtained in plants treated with SA<sub>250</sub> +CP<sub>10</sub>, whereas the control had the lowest dry matter yield (3.12 g).

#### 3.1.3. Chlorophyll (SPAD)

SPAD chlorophyll content in *Amaranthus cruentus* was also enhanced. NPK<sub>50</sub> had a higher chlorophyll content (43.30 SPAD) than the control (29.50 SPAD) and was similar to all other treatment rates except SA<sub>125</sub> and CP<sub>5</sub>, which were not significantly higher than the control. The best effect on chlorophyll content (49.30 SPAD) in combined fertilisers and salicylic acid was obtained in SA<sub>250</sub> +NPK<sub>50</sub> treated plants, followed by (46.85 SPAD) in *Amaranthus cruentus* treated with SA<sub>250</sub> + CP<sub>10</sub> while the control had the lowest chlorophyll content (29.50 SPAD) (Table 1)

# 3.2. Effects of fertilisers and salicylic acid on Folate, Nitrogen, Phosphorous, Potassium content of Amaranthus cruentus

#### 3.2.1. Folate concentration (µg/100g)

The results in Table 2 show that the application of fertiliser and salicylic acid significantly (P<0.05) enhanced folate concentrations in *Amaranthus cruentus*. In the sole treatments, the accumulation of folate in *Amaranthus cruentus* was significantly enhanced (Table 2). Highest folate concentration (245.40 µg/100 g) was obtained in SA<sub>250</sub> which was similar to 230.00 µg/100 g obtained in SA<sub>125</sub>, however, folate concentrations (123.60 µg/100 g, 101.35 µg/100 g, 140.70 µg/100 g) obtained in sole fertiliser (PM<sub>5</sub>, PM<sub>10</sub>, CP<sub>5</sub>, respectively) were not significantly (p>0.05) different from control (125.40 µg/100 g) (Table 2). In combined fertiliser and salicylic acid treatments, the highest folate concentration (285.00 µg/100 g) was enhanced by combination of SA<sub>250</sub>+NPK<sub>50</sub> and was not significantly different from that of SA<sub>250</sub>+CP<sub>10</sub>, SA<sub>250</sub>+NPK<sub>100</sub>, SA<sub>250</sub>+PM<sub>10</sub>, and SA<sub>125</sub>+NPK<sub>50</sub>. The lowest folate concentration (125.40 µg/100 g) was recorded in the control group (Table 2).

### 3.2.2. Nitrogen (%)

In the sole fertiliser treatment, a higher nitrogen concentration (2.86%) compared to the control (1.55%) was obtained in NPK<sub>100</sub>, which was similar to 2.63% under PM<sub>10</sub>, whereas NPK<sub>50</sub>, CP<sub>5</sub>, and CP<sub>10</sub> had values that were similar to those of the control (Table 2). In combined salicylic acid and fertiliser treatment, SA<sub>250</sub>+CP<sub>5</sub> gave the highest N (2.90%) which was similar to 2.70% obtained in SA<sub>250</sub>+CP<sub>10</sub> treated plants and were all significantly more than control (Table 2)

### 3.2.3. Phosphorous (%)

Phosphorous concentrations in *Amaranthus cruentus* varied across the treatments. In the sole fertiliser treatments, the highest phosphorous concentration (0.88%) was obtained in NPK<sub>50</sub> and was not better than 0.76% in SA<sub>250</sub>, whereas PM<sub>5</sub>, CP<sub>5</sub>, and CP<sub>10</sub> had values (0.47, 0.29, and 0.31%, respectively) which were similar to those of the control (Table 2). In combination with salicylic acid and fertiliser, a higher phosphorous concentration (0.85%) compared to the control (0.31%) was obtained in SA<sub>250</sub>+NPK<sub>50</sub> treated plants and was not different from the 0.76% obtained under SA<sub>125</sub> + CP<sub>5</sub> and SA<sub>250</sub>+CP<sub>10</sub> (Table 2).

#### 3.2.4. Potassium (%)

Potassium concentrations in *Amaranthus cruentus* also varied across treatments compared to the control. In the sole fertiliser treatment, the highest potassium concentration (4.41%) was obtained in NPK<sub>100</sub>, which was not better than CP<sub>10</sub>; but significantly higher than 2.08% under SA<sub>125</sub> and 2.50% under SA<sub>250</sub>. A significantly (P<0.05) higher potassium concentration was obtained in the combined salicylic acid and fertilisers than in the control (2.46%). A higher potassium concentration (5.40%) compared to the control (2.46%) was obtained in SA<sub>125</sub>+ CP<sub>5</sub> and was not significantly different from 5.39% obtained under SA<sub>250</sub>+NPK<sub>50</sub> and 5.06% under SA<sub>250</sub>+CP<sub>5</sub> (Table 2).

#### 3.3. Effects of fertilisers and salicylic acid on Iron and zinc content of Amaranthus cruentus (mg/kg)

#### 3.3.1. Iron content (mg/kg)

In sole treatments,  $CP_5$  gave higher iron concentration (170.20 mg/kg) when compared to control (138.10 mg/kg) and was similar to 158.50 mg/kg obtained in  $CP_{10}$ . In combined fertilisers and salicylic acid, the highest iron concentration (348.00 mg/kg) in *Amaranthus cruentus* was obtained under  $SA_{250}$  +  $NPK_{50}$  and was not significantly higher than iron content (342.50 mg/kg) obtained in  $SA_{250}$  +  $CP_5$  and (320.00 mg/kg) under  $SA_{250}$  +  $CP_{10}$  (Table 2).

## 3.3.2. Zinc content (mg/kg)

In sole treatments,  $CP_{10}$  had higher zinc concentration (37.50 mg/kg) when compared to control (23.00 mg/kg) and was similar to 30.00 mg/kg obtained in plant treated with  $CP_5$  (Table 2). The highest value for zinc content (47.00 mg/kg) in combined treatments of salicylic acid and fertiliser was obtained in  $SA_{250}+CP_{10}$  followed by 42.45 mg/kg obtained under  $SA_{250}+CP_5$  (Table 2).

Treatments	Fresh Weight(g)	Dry matter (g)	Chlorophyll (SPAD)
SA125	35.35e	4.72bc	34.66cde
SA250	45.60cde	4.46c	41.00abc
PM <sub>5</sub>	47.50bc	3.76cd	39.07cd
PM10	55.63b	5.27ab	40.10bc
CP <sub>5</sub>	44.00cde	4.58bc	33.80de
CP10	52.30bc	4.66c	37.52cde
NPK50	49.61c	4.23c	43.30abc
NPK100	54.70c	5.96a	40.15bc
SA <sub>125</sub> + CP <sub>5</sub>	55.00c	4.64cd	39.43cd
SA <sub>125</sub> + CP <sub>10</sub>	69.40ab	4.72bcd	43.70abc

**Table 1** Effects of fertilisers and Salicylic acid ( $\mu$ M) on fresh weight, dry matter and chlorophyll contents of *Amaranthus cruentus* in the screenhouse

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SA125 + NPK50	74.68a	5.36abc	46.50ab
SA <sub>125</sub> + NPK <sub>100</sub>	71.33ab	6.01a	37.00cd
SA125 + PM5	55.26cd	4.85bcd	46.10a
SA <sub>125</sub> + PM <sub>10</sub>	65.30bc	5.40abc	37.05cd
SA <sub>250</sub> + CP <sub>5</sub>	65.70bc	4.90bcd	45.30ab
SA <sub>250</sub> + CP <sub>10</sub>	71.05a	5.52ab	46.85a
SA <sub>250</sub> + NPK <sub>50</sub>	75.20a	5.64ab	49.30a
SA250 + NPK100	69.43ab	5.51abc	43.60abc
SA250 + PM5	69.00ab	4.97bcd	39.30cd
SA <sub>250</sub> + PM <sub>10</sub>	64.00bc	5.38abc	43.74ab
Control	40.00d	3.12e	29.50e
ans with identical lett <sub>25</sub> = Salicylic acid at	ers in each column are sta 125 μM NPK <sub>50</sub> = 5	i atistically significant (F 60 kg Nha <sup>.1</sup>	? =.05). Control = No treat

 $\begin{array}{l} \text{SA}_{125} &= \text{Salicylic acid at } 125 \ \mu\text{M} \\ \text{SA}_{250} &= \text{Salicylic acid at } 250 \ \mu\text{M} \end{array}$ 

 $NPK_{50} = 50 \text{ kg Nha}^{-1}$  $NPK_{100} = 100 \text{ kg Nha}^{-1}$ 

 $CP_5 = 5 \text{ t-ha}^{-1} \text{ Compost}$ 

 $CP_{10} = 10 \text{ t-ha}^{-1} \text{ Compost}$ 

 $PM_5 = 5 \text{ t-ha}^{-1}$  Poultry manure  $PM_{10} = 10 \text{ t-ha}^{-1}$  Poultry manure

Table 2 Effects of fertilisers and Salicylic acid (µM) on folate, zinc, iron, nitrogen, phosphorus and potassium content of Amaranthus cruentus in the screenhouse

Treatments	Folate (µg/100g)	Zn (mg/kg)	Fe (mg/kg)	N (%)	P (%)	k (%)
SA125	230.00b	27.00d	155.50de	2.06bc	0.42cde	2.08de
SA <sub>250</sub>	245.40b	30.10cd	146.30de	2.25b	0.76abc	2.50de
PM <sub>5</sub>	123.60ef	17.20e	105.80ef	2.29bc	0.47def	2.75de
PM10	101.35f	25.10d	143.00bc	2.63ab	0.62abc	2.39de
CP <sub>5</sub>	140.70def	30.00cd	170.20d	1.90c	0.29f	2.20de
CP <sub>10</sub>	152.80cde	37.50c	158.50de	1.85cd	0.31f	3.85bc
N50	180.12cd	33.30c	95.90f	1.71cd	0.80a	3.97bc
N100	138.22ef	18.90e	130.00def	2.86a	0.75ab	4.41ab
SA <sub>125</sub> + CP <sub>5</sub>	182.00c	31.80c	165.00e	2.22b	0.76ab	5.40a
SA <sub>125</sub> + CP <sub>10</sub>	242.01b	42.45b	312.20ab	1.96bc	0.54d	2.10e
SA125 + NPK50	250.00ab	44.50ab	244.6c	2.52a	0.65cd	2.56de
SA125 + NPK100	241.64b	41.60bc	210.50c	2.50a	0.40ef	3.01cd
SA <sub>125</sub> + PM <sub>5</sub>	199.30de	40.00bc	194.00cd	2.28b	0.83a	3.22c
SA <sub>125</sub> + PM <sub>10</sub>	250.45ab	25.20de	139.50e	1.95b	0.34f	4.17bc
SA <sub>250</sub> + CP <sub>5</sub>	276.13a	42.45ab	342.50a	2.90a	0.50d	5.06a
SA <sub>250</sub> + CP <sub>10</sub>	260.45ab	47.00a	320.00ab	2.70a	0.76ab	3.34cd
SA250 + NPK50	285.00a	40.50ab	348.00a	2.38bc	0.85a	5.39a
SA <sub>250</sub> + NPK <sub>100</sub>	261.50ab	34.70c	257.30c	2.01bc	0.48ef	4.41ab
SA <sub>250</sub> + PM <sub>5</sub>	184.90e	43.50ab	215.00c	2.55b	0.44ef	3.62bc
SA <sub>250</sub> + PM <sub>10</sub>	256.33ab	45.80ab	320.00ab	1.60c	0.68c	3.20cd

Control	125.40f	23.00de	138.10f	1.55c	0.31ef	2.46de
Means	Means with identical letters in each column are statistically significant (P = 05).					
SA125	= Salicylic acid at 125 μM	NPK <sub>50</sub> = 50 kg Nha	a <sup>-1</sup>	Control	= No treatme	nt
SA250	= Salicylic acid at 250 μM	NPK100 = 100 kg N	ha-1			
CP <sub>5</sub>	= 5 t-ha-1 Compost	$PM_5 = 5 t - ha^{-1}P$	oultry manure			
CP10	= 10 t-ha <sup>-1</sup> Compost	$PM_{10} = 10 \text{ t-ha}^{-1}$	Poultry manure			

# *3.4.* Effects of salicylic acid and fertilisers on Crude fibre, ash and moisture content (%) of *Amaranthus cruentus*

#### 3.4.1. Crude fibre (%)

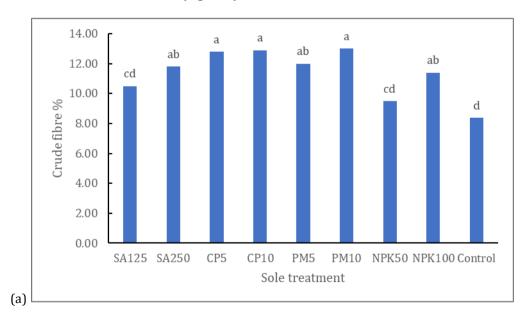
Crude fibre, ash, and moisture contents in *Amaranthus cruentus* were significantly influenced by fertilisers and salicylic acid. In the sole treatments, as shown in Figure 1, when compared to the control (8.39%), higher crude fibre content (13.00%) was obtained in  $PM_{10}$  and was not significantly higher than values obtained in the sole fertiliser and sole elicitor treatments, except in  $SA_{125}$  and  $MejA_{500}$ . The results showed that the application of  $SA_{125} + PM_5$  resulted in the highest crude fibre content (13.10%) among all the treatments, and it was not significantly different from  $SA_{125} + NPK_{100}$ ,  $SA_{250} + NPK_{50}$  and  $SA_{250} + PM_5$  (Figure 1).

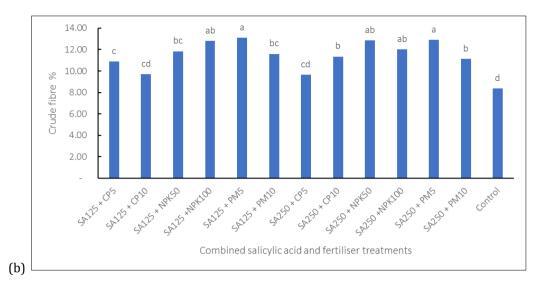
#### 3.4.2. Ash (%)

In the sole treatments,  $PM_{10}$  had the highest ash content (10.50%), which was not different from the ash content obtained in all organic fertilizer treatments,  $NPK_{100}$ , and  $SA_{250}$  (Figure 2). Higher ash content (12.35%) of *Amaranthus cruentus* in the combined treatments when compared to the control was obtained in  $SA_{125}+PM_5$  and was similar to  $SA_{250}+PM_5$  and  $SA_{250}+PM_{10}$  (Figure 2).

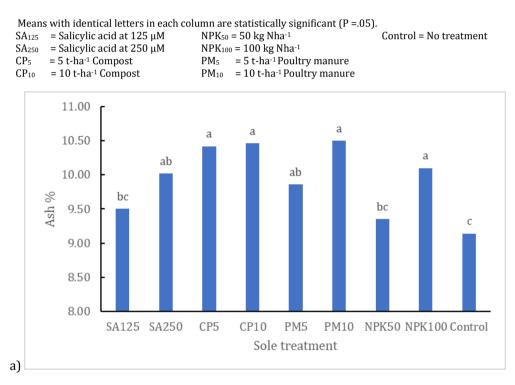
#### 3.4.3. Moisture (%)

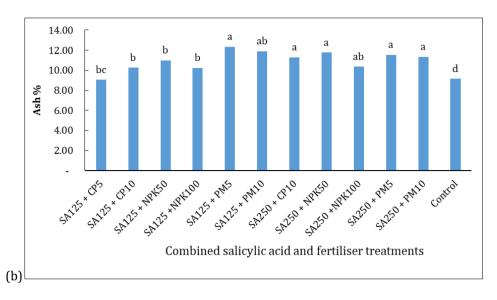
In terms of moisture content of *Amaranthus cruentus* as indicated in Figure 3, sole organic fertilisers enhanced moisture content when compared to the control with  $CP_{10}$  having the highest value (11.70%) and was comparable to values obtained in all organic fertiliser treatments and  $SA_{250}$ . In the combined fertilizer and salicylic acid treatments, higher moisture content (12.25%) was obtained in  $SA_{250}$ +PM<sub>10</sub> when compared to the control and was similar to all the rates in the combined SA and fertilizer treatments (Figure 3).



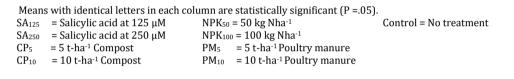


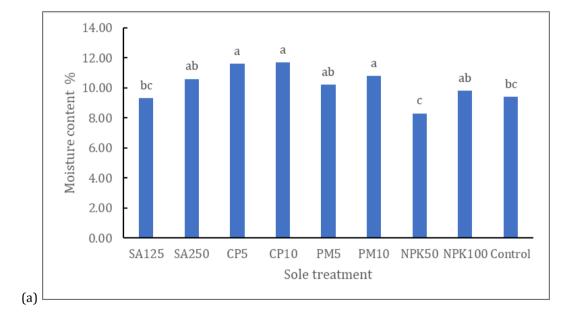
**Figure 1** Effects of (a) sole fertiliser and elicitor, (b) combined salicylic acid (μM) and fertiliser crude fibre content of *Amaranthus cruentus* in the screenhouse

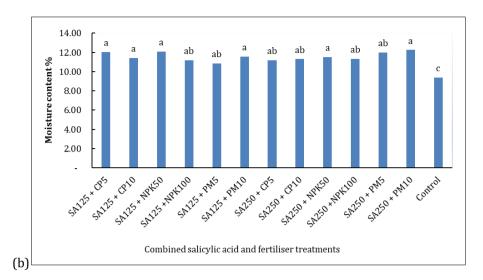




**Figure 2** Effects of (a) sole fertiliser and elicitor, (b) combined salicylic acid (μM) and fertiliser on ash content of *Amaranthus cruentus* in the screenhouse







**Figure 3** Effects of (a) sole fertiliser and elicitor, (b) combined fertilisers and salicylic acid (μM) on moisture content of *Amaranthus cruentus* in the screenhouse

Means with identical letters in each column are statistically significant ( $P = .05$ ).					
$SA_{125}$	= Salicylic acid at 125 μM	$NPK_{50} = 50 \text{ kg Nha}^{-1}$	Control = No treatment		
SA250	= Salicylic acid at 250 μM	$NPK_{100} = 100 \text{ kg Nha}^{-1}$			
CP <sub>5</sub>	= 5 t-ha <sup>-1</sup> Compost	PM <sub>5</sub> = 5 t-ha <sup>-1</sup> Poultry manure			
$CP_{10}$	= 10 t-ha-1 Compost	PM <sub>10</sub> = 10 t-ha <sup>-1</sup> Poultry manure			

## 4. Discussion

Soil nutrients can be enhanced by the application of fertilisers (organic and inorganic), which leads to improved crop production and nutrient quality. Moreover, organic manure is environmentally friendly and can improve soil organic mineral content, although the required amount varies depending on the intensity of the planting, the combination of plants, and the types of crops grown [23]. In contrast, inorganic fertilisers provide nutrients to crops in readily available forms [24]. Understanding the amount of nutrients required for optimum plant performance is key to mitigating the effects of micronutrient malnutrition.

Furthermore, elicitation, which involves the use of abiotic or biotic agents to increase the content of a desired metabolite in plants, is also a good option for enhancing the nutrient quality of crops, because elicitors such as salicylic acid can be used to boost plant secondary metabolite synthesis in crops, which serves as an alternative method in which the high cost of research is associated with metabolic engineering. In this study, the positive effects of foliar spray of salicylic acid alone or in combination with either organic or inorganic fertilisers on the marketable fresh weight and dry matter accumulation in *Amaranthus cruentus* might be linked to the bioregulatory effects of salicylic acid on enzyme activity as well as regulation of translocation process of enzymes across plant parts, thus making foliar application effective in improving *Amaranthus cruentus* performance.

Moreover, the observed effects in improving marketable yield and dry matter accumulation in *Amaranthus cruentus* could be because salicylic acid is involved in cell membrane protection and in binding transporter proteins, thus maintaining their structure and function against the destructive effects of reactive oxygen species (ROS), which in turn can enhance absorption and translocation of minerals [25]. Furthermore, the yield and dry matter accumulation were found to generally improve with sole fertiliser application of poultry manure at 10 t/ha and NPK at 100 kg N/ha, respectively, having profound effects on *Amaranthus cruentus*. The values obtained were similar to most of the fertiliser rates, suggesting that any of the fertilisers used in this study have the ability to enhance marketable yield of *Amaranthus cruentus*. These effects of fertilisers in enhancing plant yield and dry matter accumulation were reported by Singh [26], and Makinde [27] also reported that organic fertilisers alone or in combination with NPK significantly improved the proximate composition of *Amaranthus*.

Salicylic acid (SA) at either 125 or 250  $\mu$ M/L combined with fertilisers did not show significant differences in their effects on plant performance across different treatments. However, SA<sub>250</sub> had higher values, suggesting that SA<sub>250</sub> might be more effective in enhancing plant performance in leafy vegetables. Moreover, SA<sub>125</sub> combined with NPK<sub>100</sub> had the highest dry matter yield, but it was not significantly different from that of SA<sub>250</sub> and compost at 10 t/ha. These results

indicate that both organic and inorganic fertilisers can enhance the dry matter yield when combined with SA at either 125 or 250  $\mu$ M/L. This assertion was corroborated by Ibrahim [28], who reported positive effects of salicylic acid and fertilisers on the yield of wheat plants.

Chlorophyll is a major pigment necessary for photosynthesis, and the amount present in plants indicates photosynthetic efficiency and plant health status. Plant chlorophyll levels have been shown to have significant effects on plant performance in terms of yield and nutrient content. Chlorophyl has also been linked to enhanced synthesis of some macro-and micronutrients in plants. Thus, chlorophyll is essential for plant growth, yield, and the synthesis of nutrients such as vitamins and minerals. The SPAD values for chlorophyll in this study in combined application, were higher in plants treated with either SA at 125 and 250  $\mu$ M/L, as well as in NPK fertilisers at both levels (50 and 100 kg N/ha). This is also corroborated by Ghassemi-Golezani and Lotfi [29], who reported a positive influence of salicylic acid on photosynthesis relative vitality in mung beans, while Khandaker [30] also reported increased chlorophyll content in red amaranths sprayed with salicylic acid.

Furthermore, as observed in this study, the sole or combined use of fertiliser and salicylic acid (SA) treatments influenced the nutrient content of *Amaranthus cruentus*. Fertilisers may have increased the availability of nutrients to *Amaranthus cruentus*, resulting in higher yield and nutrient content. This suggests that fertiliser application is important for production of quality foods that contribute to a healthy diet. However, the highest dose of NPK (100 kg N/ha) combined with salicylic acid did not produce a significantly higher yield than the lower dose of NPK (50 kg/ha) combined with salicylic acid, indicating that NPK<sub>50</sub> is adequate for *Amaranthus cruentus* production because excessive application should be avoided to prevent the negative environmental impacts of inorganic fertilisers. Moreover, the effects of NPK fertilisers were similar to those of compost and poultry manure, demonstrating that organic fertilisers can also enhance the yield and nutrient content of *Amaranthus cruentus* by improving soil properties and crop quality.

Folates are highly unstable molecules whose turnover rate is higher than that of ATP [31] and are susceptible to oxidative damage by cleaving to form their derivatives [32]; thus, the overall performance and management practices of plants during the growth period are essential for folate synthesis. The present study shows that foliar application of salicylic acid increases folate concentration in *Amaranthus cruentus*. Generally, folates in plants are highly unstable owing to oxidative cleavage, which results in the degradation of folates into Pteridine and PABA- glutamyl fragments. The effects of SA on folate enhancement might be due to its ability to improve folate stability by reducing oxidative stress in *Amaranthus cruentus* [32]. SA also reduces cellular free radical levels, thus enabling folate-binding proteins to be tightly bound to its matrices, thus enhancing folate stability and detection in *Amaranthus cruentus* [33, 34, 35].

SA<sub>250</sub> + NPK<sub>50</sub> appeared to be the most suitable combination for folate enhancement in *Amaranthus cruentus*. It is noteworthy to state that these effects were also comparable to compost effects, while the combination of compost with SA<sub>250</sub> enhanced zinc and iron levels. This further shows that organic fertilisers affected the apportioning quality of *Amaranthus cruentus*. In terms of crude fibre content, SA<sub>125</sub> combined with PM<sub>5</sub> showed higher effects on the crude fibre content of *Amaranthus cruentus*. SA<sub>125</sub> combined with PM<sub>5</sub> showed superior effects on ash contents of *Amaranthus cruentus*, similar trend was observed in moisture content with combination of SA<sub>250</sub> and PM<sub>10</sub> improving moisture content and was also similar to combined fertilisers and salicylic acid rates. This shows that the combined application of salicylic acid and fertilisers improved the crude fibre, ash, and moisture content of *Amaranthus cruentus*, suggesting that the combined application enhanced *A. cruentus* overall nutritional value. This is corroborated by Mofunanya [36], who reported a high crude fibre content in *Amaranthus spinosus* L. treated with poultry manure.

# 5. Conclusion

Salicylic acid and fertilisers enhanced the marketable yield, dry matter, chlorophyll, folate, iron, zinc, and proximate composition of *Amaranthus cruentus* (L.) compared to the control. Foliar application of SA at 250  $\mu$ M/L significantly increased the folate concentration in *Amaranthus cruentus*. The optimal treatment with a combination of salicylic acid at 250  $\mu$ M and NPK fertiliser at 50 kg N/ha resulted in higher marketable yield, dry matter, and folate. Additionally, salicylic acid at 250  $\mu$ M combined with compost at 10 t/ha enhanced iron and zinc. In conclusion, the application of combined salicylic acid and fertiliser could improve the nutritional quality of *Amaranthus cruentus* (L.) and is a promising option for the production of nutritious diets that can help ameliorate folate, iron, and zinc deficiency in humans.

#### **Compliance with ethical standards**

#### Disclosure of Conflict of interest

There is no conflict of interest to be disclosed.

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