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Effects of Different Kinds of Fertilizers on the Vegetative Growth, Antioxidative Defense System and Mineral Properties of Sunflower Plants

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Abstract: Long-term use of inorganic fertilizers can increase soil acidity, be harmful to the environment, and leaving bad effects on human health. Organic fertilizer application is one of the safer alternatives with numerous benefits, such as supplying nutrients for plant growth. Sunflower is one of the most important grown oilseed crops in the world. Sunflower plants need a supply of essential nutrients for their optimal growth. As a result, the aim of this research is to explore the effect and mechanism of two organic fertilizers from different sources (sugarcane bagasse ash (SBA), compost coupled with biofertilizer (CCB)) and NPK inorganic fertilizer as a control on enzyme activity, physiological traits, and the uptake of mineral contents and heavy metals in sunflower plant (*Helianthus annuus* L.). Fresh or dry mass (FM, DM), osmolytes and secondary metabolites, photosynthesis pigments, and enzymatic and non-enzymatic antioxidant molecules were all determined. Both sugarcane bagasse ash and compost coupled with biofertilizer resulted in a high value of fresh and dry mass, plant height, and chlorophyll content. The results revealed that the use of sugarcane bagasse ash (SBA) and compost coupled with biofertilizer increased osmolyte contents (soluble proteins and soluble sugars), antioxidants system enzyme/molecule (catalase, superoxide dismutase, peroxidase, and TGS) and secondary metabolites. However, the highest value of proline, total free amino acids, and phenolic compounds in sunflower plants was determined after NPK fertilizer application. On the contrary, it lowered Na, Na/K ratio, and Cd content. Data showed that organic fertilizers enhanced the accumulation of Cl, PO_4, and SO_4 content in sunflower plants. Generally, CCB and SBA treatments increased Cu, Zn, and Pb accumulation in sunflower plants. Using organic fertilizers with chemical NPK fertilizer can improve the chemical, physical, and biological soil properties.

Keywords: fertilizers; sunflower; vegetative growth; heavy metals; minerals; antioxidants

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1. Introduction

Sunflower plants (*Helianthus annuus* L.) are among the most essential oilseed crops, containing high-quality edible oil, and are considered a source of human food and raw materials for industry [1,2]. Sunflower growth declines are primarily caused by biotic and abiotic stress factors [3]. In order to optimize its growth, fertilization plays an important role in increasing sunflower yield. The demand to increase food production to feed an ever-increasing population has encouraged the application of synthetic mineral fertilizers capable of providing crops with the nutrients they require for growth [4]. However, farmers tend to add inorganic fertilizer excessively during plant cultivation based on the assumption that it may enhance the yield [5]. Using inorganic fertilizers alone increases crop yield in the first year but has a negative impact on long-term sustainability [6]. An imbalance in fertilizer use is one of the primary factors of sparse crop yield and reduction in soil fertility. As a result, organic fertilizers should be used to increase crop production. Organic fertilizers are naturally occurring products derived from plants or animals, such as green manure, crop residues, livestock manure, and compost [7]. The addition of organic fertilizers combined with inorganic fertilizers enhanced crop growth and yield by improving physical soil conditions, soil fertility, and nutrient availability [8–10].

Furthermore, organic fertilizers used in conjunction with chemical fertilizers produced higher yields than inorganic fertilizers alone, increased soil carbon storage, decreased emissions from mineral nitrogen fertilizer, and participated in high crop yields and quality [11–13].

Organic fertilizers are used to mitigate the toxicity of compounds (such as nitrates) produced by chemical fertilizers in plants, thereby improving crop quality and human health. As a result, farmers have initiated to utilize of organic fertilizers to improve soil chemical and physical structure. Crop productivity is increased by carefully adjusting agricultural methods such as fertilization, tillage, and removal of weeds [14–17]. There are several kinds of organic fertilizers, such as biofertilizers which are considered the most recent eco-friendly technique for promoting sustainable agriculture. Biofertilizers contain plant growth-promoting microbes such as nitrogen-fixing bacteria and phosphate-solubilizing microbes. Both of these microbes can stimulate the availability of nitrogen (N) and phosphate (P) enzymatically [18,19]. When these bacteria are used as seed or soil inoculants, they play an active role in nutrient cycling and enhance nutrient supply for plant growth, which has a significant effect on crop yield [20,21].

According to these studies, using of organic fertilizers and biofertilizers along with inorganic fertilizer, have a significant impact on soil nutrient availability, aggregate formation, and soil bacterial communities.

As a result, the current study was carried out to investigate the effects and mechanisms of fertilizers from various sources (sugarcane bagasse ash (SBA) compost coupled with biofertilizer (CCB) and NPK fertilizer) on vegetative growth, antioxidant system, and mineral and heavy metal accumulation in sunflower plants.

2. Materials and Methods

2.1. Experimental Setup

Under natural conditions of humidity, temperature, and light, a pot experiment was conducted in a wirehouse at the farm of the Botany and Microbiology Department, Faculty of Science, South Valley University, Egypt. The pot used in our experiment has specific dimensions (40 and 30 cm in height and diameter, respectively) and is filled with 10 kg soil. The experiment used a randomized complete design with 4 replications for each treatment. There were 3 kinds of fertilizers (NPK fertilizer (as a control) and compost coupled with biofertilizer (CCB). Organic treatment was used at a rate of 300 G/pot as a nitrogen source, with rock phosphate (31% P_2O_5) and feldspar (10% K_2O) as recommended. Organic fertilizer was mixed into the soil a month before seeding, and 3 days before planting, water was added (for saturation). The field capacity for each treatment was calculated, and the daily water loss was compensated. In each pot, 10 sunflower seeds were planted. Inorganic treatment was used as ammonium nitrate 33.5% N, superphosphate 15.5% P_2O_5, incorporated in 1 dose in each pot during preparation for planting, and potassium sulfate 48% K_2SO_4, added with the addition of ammonium nitrate fertilizer. All treatments received equal and necessary residual agricultural operations.

2.2. Plant Growth Parameters

After three weeks, plant samples from each of the treatments mentioned above were collected and divided into shoot and root samples. The shoot and root lengths of sunflower seedlings were then measured, and the shoot/root fresh mass (FM) of the seedlings. For DM determinations, the seedlings were dried in the oven at 70 °C for 48 h. For biochemical analysis, fresh plant tissue was also rapidly frozen and kept at −20 °C.

2.3. Soil and Organic Fertilizers Analysis

According to [22], the pipette technique was employed to estimate the particle size distribution of soil. For the determination of the organic material in soil and organic wastes, the method outlined in [23] was utilized, and soil organic material was then determined.

The total calcium carbonate content was measured based on a Collins volumetric calcimeter [22]. The electrical conductivity (EC) of soil samples was determined in a 1:10 water extract (soil:water ratio) using EC meters [22]. The soil texture used in the experiment was clay loam, with a pH of about 7.9 and an EC of 3 dS/m. The percentage of clay was 30%, silt 29%, and sand 41%. Soil chemical analyses indicated 8% $CaCO_3$, with 19 mg/kg available nitrogen, 11 mg/kg available P, 200 mg/kg available K, and 14 g/kg organic carbon. The chemical properties of organic fertilizers (Table 1) were calculated according to the method mentioned in [22].

Table 1. Chemical and physical properties of organic fertilizers.

Property	Compost Coupled with Biofertilizer	Sugarcane Bagasse Ash
pH (1:1) in water	7.32	6.7
EC (1:10; dS/m)	2.76	5.6
Organic carbon%	14.95	40.2
N%	0.85	2.5
P%	0.72	2.7
K%	1.62	0.45

2.4. Preparation of Organic Fertilizers Treatment

Organic waste sugarcane factories were used to produce SBA (sugarcane bagasse ash). CCB was made from experimental agricultural residues and also included nitrogen fixers (*Azospirillum lipoferum* and *Azotobacter chroococcum*) as well as phosphate-dissolving bacteria (*Bacillus polymyxa* and *Paenibacillus polymyxa*). *Azospirillum lipoferum* and *Azotobacter chroococcum* as nitrogen fixers were isolated, as mentioned by [24]. *Bacillus polymyxa and Paenibacillus polymyxa*, as phosphate-dissolving bacteria (PDB), were isolated, as mentioned in [25]. The studied microorganisms were obtained from the Microbiological Resources Center, Faculty of Agriculture. Ain Shams University, Cairo, Egypt.

As a carrier for preparing inoculants, dry pulverized, neutralized, and sterilized moss peat was used. Mixed biofertilizer inoculant was made up of an equal amount of chroococcum inoculant (4.4×10^6 cfu/gm), *Azospirillum lipoferum* inoculant (6×10^6 cfu/gm), and Phosphate dissolving bacteria inoculant (3×10^8 cfu/gm) just before use for seed inoculation at the percentage of 10% of the seed weight and was combined thoroughly till the seeds were homogeneously surface-covered using 20% Arabic gum solution as an adhesive.

2.5. Photosynthetic Pigments

Chlorophyll a and b, as well as carotenoids, were determined by spectrophotometry using equations recommended in [26]. A 250 mg fresh leaf sample was extracted in a pre-chilled mortar with 10 mL acetone (80%, *v*/*v*), and the extract was transferred to a 15 mL centrifuge tube and centrifuged at 10,000 rpm for 10 min. The pellet was discarded, and the absorbance of the resulting supernatant was determined with a spectrophotometer at 663, 645, and 470 nm to determine chlorophylls and carotenoids. For the blank, acetone (80%, *v*/*v*) was used.

2.6. Metabolites Content

The method of [27] was used to determine the soluble protein content. For 2 h, powdered tissue samples (50 mg) were boiled in 10 mL of distilled water. The water extract was centrifuged at 6000 rpm after cooling; the supernatant was taken out and made up to a specific volume with distilled water.

Proline was performed according to the method outlined by [28]. The plant sample (0.2 g) was homogenized in 5 mL of 3% sulfosalicylic acid and reacted in a test tube for

1 hour at 100 °C with 2 mL of acid ninhydrin and 2 mL of glacial acetic acid. The reaction was stopped with an ice bath, and the mixture was extracted with 4 mL of toluene. At room temperature, the chromophore containing toluene was aspirated, and absorbance at 520 nm was estimated using toluene as a blank. Total free amino acids were elicited as described by [29].

The anthrone sulfuric acid method [30] was used to quantify the water-soluble sugars. A known weight of dried tissue material was boiled in 10 mL of H_2O for 2 h to estimate water-soluble sugars; after cooling, the extracts were filtered and made up to 50 mL. 4.5 mL of anthrone reagent was mixed with 0.5 mL of the prepared solution, and the soluble sugar content was determined as mg g^{-1} DM.

2.7. Enzymatic and Non-Enzymatic Antioxidants

After homogenizing, the plant tissue in the K-phosphate buffer contained ethylenediaminetetraacetic acid (EDTA) and polyvinylpyrrolidone (PVP). The supernatant was used for the enzyme assay. Catalase (CAT) activity was assessed using the protocol outlined in [31]. The rate of H_2O_2 decomposition was measured spectrophotometrically at 240 nm. The reaction mixture contained 2 mL of 0.05 M K-phosphate buffer pH 7.0, 1 mL of 0.1 mM H_2O_2, and 100 l of supernatant. At 30 °C, 1 unit of catalase activity (U) was defined as the amount of enzyme that decomposed 1 mol H_2O_2 mg^{-1} soluble protein min^{-1}. Peroxidase (POD) activity was detected by the protocol of [32]. Superoxide dismutase (SOD) activity was determined as described by [33]. A unit of SOD activity was expressed as the amount of enzyme needed to inhibit NBT photoreduction by 50%.

Glutathione content was estimated using 150 µL of a solution including 0.5 mL buffer (50 mM HEPES-KOH pH 7.6 and 330 mM betaine), 0.3 mL of 10% (*w*/*v*) sulfosalicylic acid and 0.1 mL of extract was mixed with 700 µL of 0.3 mM NADPH, 100 µL of 6 mM 5,5-O-dithiobis-(2-nitrobenzoic acid) and 50 µL of glutathione reductase (10 units ml^{-1}). The formation of 2-nitro-5-mercaptobenzoic acid was accompanied by an increase in absorbance at 412 nm, as described by [34].

2.8. Phenolic Compounds, Tocopherol Contents

The content of phenolic compounds was calculated based on the protocol outlined in [35]. A 0.1 g sample of plant tissue was mixed in 10 mL ethanol (80%) and then centrifuged at 10,000 rpm for 10 min. The supernatant was taken out and centrifuged once more. An alcoholic aliquot (1 mL) was added to 1 mL of 20% sodium carbonate before the addition of 0.5 mL of the Folin-phenol reagent. It was boiled in a water bath for 10 min at 100 °C. The final volume was 20 mL of distilled water, and the sample's absorbance was measured at 660 nm using a UV spectrophotometer. Tocopherol was determined in the supernatant of fresh leaves grounded in chloroform and was used for measuring tocopherol according to the protocol outlined in [35].

2.9. Determination of Ionic Status

Plant tissues were weighed, oven-dried, and crushed to fine powder at the end of the experiment for extraction and chemical analysis. According to [36], plant extracts were prepared for mineral analysis (Cl, SO_4, PO_4, Na, K, Ca, and Mg). The mixed acids digestion procedure described by [37] is used for heavy metals analysis. The total contents of Pb, Zn, Cu, and Cd were determined using an atomic absorption spectrophotometer.

2.10. Statistical Analysis

The SPSS software version 23 (SPSS Inc., Chicago, IL, USA) was applied for statistics. A general 1-way model was used for the analysis of variance (ANOVA), and Duncan's test was used to compare means. The data and the significant differences between the treatments were evaluated using a 1-way analysis of variance (ANOVA) and Duncan's test at $p \leq 0.05$ and shown as mean ± standard error (n = 4) for 4 replicates.

3. Results

The data in Table 2 indicate that fertilizing sunflower plants with organic fertilizer (sugarcane bagasse ash SBA) were noticed to be the most influential for obtaining the highest values of the examined vegetative growth parameters (shoot/root fresh and dry mass and shoot/root length) as compared with NPK inorganic fertilizer. In contrast, fertilizing the plants with inorganic fertilizer resulted in the lowest values of vegetative growth parameters.

Table 2. Effects of different kinds of fertilizers (inorganic fertilizer, compost coupled with biofertilizer, sugarcane bagasse ash) on some vegetative growth parameters of sunflower plants.

Treatments	Shoot Fresh Mass (g/Plant)	Shoot Dry Mass (g/Plant)	Root Fresh Mass (g/Plant)	Root Dry Mass (g/Plant)	Shoot Length (cm)	Root Length (cm)
Inorganic fertilizer	6.2 ± 0.30 c	0.587 ± 0.01 b	2.2 ± 0.2 c	0.19 ± 0.01 c	21 ± 1 b	10 ± 1 c
Compost coupled with biofertilizer	7.2 ± 0.30 b	0.737 ± 0.02 c	3.2 ± 0.2 b	0.300 ± 0.01 b	23 ± 1 b	12 ± 1 b
Sugarcane bagasse ash	9.1 ± 0.30 a	0.920 ± 0.02 a	4.6 ± 0.2 a	0.446 ± 0.01 a	26 ± 2 a	14 ± 1 a

Different lowercase letters within the same parameters show significant differences. ($p < 0.05$). Data are represented as means ± SD (n = 4). Different letters indicate significant differences between treatments at $p < 0.05$ according to Duncan test.

The effects of two organic fertilizers from different sources (CCB and SBA) on the photosynthetic pigments are explained in Table 3. Compost coupled with biofertilizer (CCB) reduced chlorophyll a (Chl a) and chlorophyll b (Chl b) levels while increasing carotenoids. In contrast, there was a significant increase in Chl a, Chl b, and carotenoids after fertilizing with sugarcane bagasse ash as compared with NPK inorganic fertilizer.

Table 3. Effects of different kinds of fertilizers (inorganic fertilizer, compost coupled with biofertilizer, and sugarcane bagasse ash) on photosynthetic pigments of sunflower plants.

Treatments	Chlorophyll a (mg g^{-1} FM)	Chlorophyll b (mg g^{-1} FM)	Carotenoids (mg g^{-1} FM)	Total	Chl a/b
Inorganic fertilizer	0.526 ± 0.027 b	0.116 ± 0.010 b	0.159 ± 0.011 b	0.802 ± 0.039 b	4.555 ± 0.371 b
Compost coupled with biofertilizer	0.415 ± 0.015 c	0.065 ± 0.006 c	0.172 ± 0.009 b	0.654 ± 0.021 c	6.360 ± 0.766 a
Sugarcane bagasse ash	0.702 ± 0.020 a	0.158 ± 0.018 a	0.206 ± 0.025 a	1.064 ± 0.028 a	4.472 ± 0.670 b

Different lowercase letters within the same parameters show significant differences. ($p < 0.05$). Data are represented as means ± SD (n = 4). Different letters indicate significant differences between treatments at $p < 0.05$ according to Duncan test.

Figure 1 depicts that both organic treatments (CCB and SBA) significantly increased soluble proteins (SP) and soluble sugars (SS) in *Helianthus annus* plants as compared with the inorganic treatment. The increase of soluble proteins in sunflower plants under the effect of CCB and SBA was 43% and 73%, respectively, as compared with the control. Furthermore, the increase of soluble sugars under the effect of CCB and SBA was 43% and 103%, respectively, as compared with inorganic fertilizer. On the other hand, CCB and SBA reduced total free amino acids content by about 41% with SBA application and 10% in the case of CCB application as compared with the control (NPK treatment). Proline content was only significantly reduced (by 38%) due to the application of sugarcane bagasse ash.

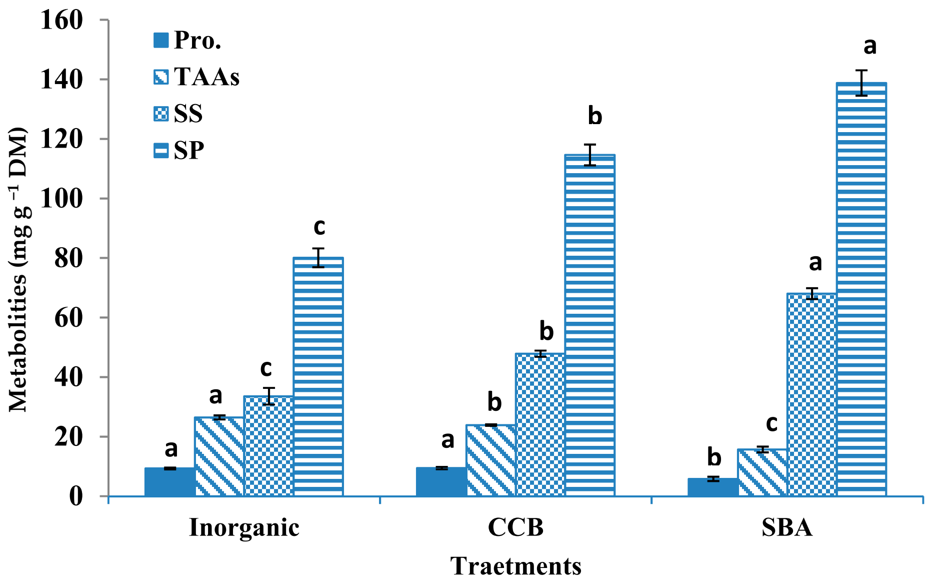

Figure 1. Metabolites (proline (pro.), total free amino acids (TAAs), soluble sugar (SS), soluble protein (SP)) of sunflower plants under the effects of different kinds of fertilizers (inorganic fertilizer, compost coupled with biofertilizer (CCB), sugarcane bagasse ash (SBA)). Data are represented as means ± SD (n = 4). Different letters indicate significant differences between treatments at $p < 0.05$ according to Duncan test.

The activities of peroxidase (POD), superoxide dismutase (SOD), and catalase (CAT) improved under the effect of both organic applications (CCB and SBA). In comparison to inorganic treatment, SBA application resulted in a significant increase in catalase activity (155%), peroxidase activity (94%), and superoxide dismutase (88%). In the case of compost combined with biofertilizer, the increase in CAT, POD, and SOD activities was approximately 43%, 38%, and 31%, respectively, when compared to the control (NPK inorganic treatment; Figure 2).

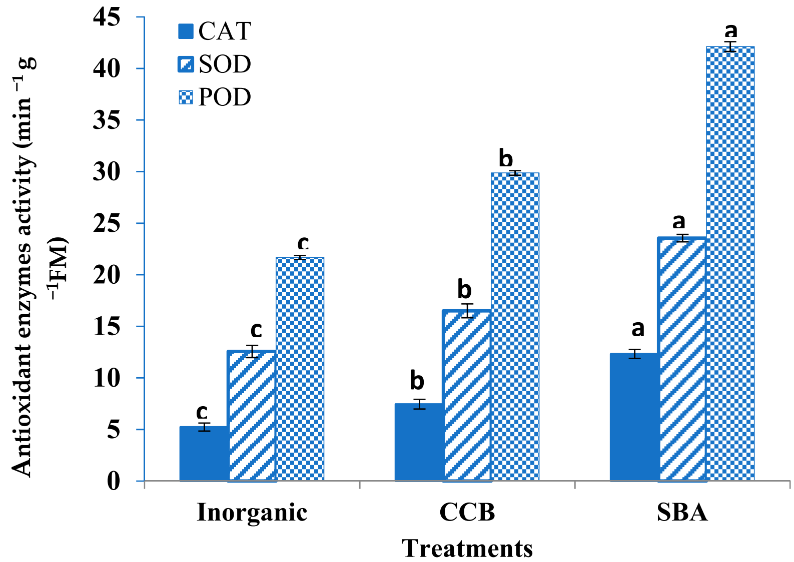

Figure 2. Antioxidant enzymes activity (peroxidase (POD), superoxide dismutase (SOD) and catalase (CAT)) of sunflower plants under the effects of different kinds of fertilizers (inorganic fertilizer, compost coupled with biofertilizer (CCB), sugarcane bagasse ash (SBA). Data are represented as means ± SD (n = 4). Different letters indicate significant differences between treatments at $p < 0.05$ according to Duncan test.

Contents of glutathione, tocopherol, and phenolic compounds were determined in sunflower plants (Table 4). The phenolic content result in Table 4 shows that the fertilizer source had a significant impact on the total phenolic content of sunflower plants. The content of phenolic compounds under the application of inorganic samples was 22.05, while compost (CCB) and SBA samples had values of 19.45 and 17.34 mg g FM, respectively. It was noticed that using organic fertilizer reduced total phenolic production in sunflower plants. According to the data presented in Table 4, compost (CCB) had the highest tocopherol content with 512.95 μg g^{-1} FM, followed by SBA with 452.16 μg g^{-1} FM, and inorganic samples had the lowest tocopherol content (432.56 μg g^{-1} FM). The data show that compost coupled with biofertilizer as a source of nitrogen produced more tocopherol than inorganic and SBA fertilizers. Glutathione content was enhanced significantly under the effect of organic fertilizers application by 56% and 28% in the case of SBA and CCB application, respectively, as compared with NPK inorganic application.

Table 4. Effects of different kinds of fertilizers (inorganic fertilizer, compost coupled with biofertilizer, sugarcane bagasse ash) on glutathione, tocopherol and phenolic compounds of sunflower plants.

Treatments	GSH (μg g^{-1} FM)	Tocopherol (μg g^{-1} FM)	Phenolic Compounds (mg g^{-1} FM)
Inorganic fertilizer	12.35 ± 0.11 c	432.56 ± 3.982 b	22.05 ± 0.241 a
Compost coupled with biofertilizer	15.75 ± 0.07 b	512.95 ± 4.341 a	19.45 ± 0.723 b
Sugarcane bagasse ash	19.21 ± 0.14 a	452.16 ± 2.786 b	17.34 ± 0.123 b

Different lowercase letters within the same parameters show significant differences ($p < 0.05$). Data are represented as means ± SD (n = 4). Different letters indicate significant differences between treatments at $p < 0.05$ according to Duncan test.

The data presented in Table 5 showed that there was a noticeable enhancement in K, Ca, and Mg contents in sunflower plants under the effect of both organic applications as compared to the control (inorganic treatment). For instance, K content reached the highest level (about 120% higher than the control) under fertilizing *H. annus* plants with SBA. Furthermore, the results presented revealed that the highest contents of Ca and Mg in sunflower plants were recorded in soil with SBA, followed by soil amended with CCB. The results in the same table show that, in comparison to the other minerals, Na content decreased significantly, reaching 50% less than the control under SBA application and 39% less than the control under CCB fertilization. Both organic applications showed a remarkable reduction in Na/K ratio in sunflower plants, which was maximum (about 77% and lesser than the control) under the application of SBA and (59% lesser than the control) in the case of compost coupled with biofertilizer (CCB; Table 5). Generally, the K, Ca, and Mg contents in the control plant were lower than the plant fertilized with organic fertilizer.

Table 5. Effects of different kinds of fertilizers (inorganic fertilizer, compost coupled with biofertilizer, sugarcane bagasse ash) on cations content and Na/K ratio of sunflower plants.

Treatments	Na (mg g^{-1} DM)	K (mg g^{-1} DM)	Ca (mg g^{-1} DM)	Mg (mg g^{-1} DM)	Na/K
Inorganic fertilizer	8.72 ± 0.344 a	11.76 ± 0.265 c	14.62 ± 0.564 c	22.65 ± 1.03 c	0.741 ± 0.07 a
Compost coupled with biofertilizer	5.34 ± 0.232 b	17.45 ± 0.824 b	21.43 ± 0.123 b	28.22 ± 2.54 b	0.306 ± 0.05 b
Sugarcane bagasse ash	4.38 ± 0.114 c	25.65 ± 0.512 a	34.98 ± 0.166 a	41.73 ± 1.43 a	0.171 ± 0.02 c

Different lowercase letters within the same parameters show significant differences ($p < 0.05$). Data are represented as means ± SD (n = 4). Different letters indicate significant differences between treatments at $p < 0.05$ according to Duncan test.

SBA and CCB enhanced the accumulation of Cl, PO_4, and SO_4 content in the tested plant as compared with the control (NPK inorganic treatment; Table 6). The greatest PO_4 and SO_4 contents were recorded in soil with CCB, followed by soil amended with SBA.

At the same time, Cl content reached a maximum value when fertilizing *H. annus* plants with SBA.

Table 6. Effects of different kinds of fertilizers (inorganic fertilizer, compost coupled with biofertilizer, sugarcane bagasse ash) on heavy metals content and anions of sunflower plants.

Treatments	µg/g.DM				µg/DM	mmol/g DM	mmol/g DM
	Pb	Zn	Cu	Cd	Cl	SO_4	PO_4
Inorganic fertilizer	24.12 ± 0.42 c	121.68 ± 2.12 c	79.29 ± 0.63 c	26.58 ± 0.21 a	35.67 ± 0.45 c	120.69 ± 1.55 c	85.48 ± 1.34 c
Compost coupled with biofertilizer	34.28 ± 1.21 b	260.04 ± 3.23 b	150.63 ± 0.95 b	14.87 ± 0.11 c	49.65 ± 1.12 b	232.34 ± 3.45 a	150.95 ± 2.12 a
Sugarcane bagasse ash	42.72 ± 1.65 a	385.64 ± 4.76 a	230.53 ± 2.76 a	20.19 ± 0.09 b	58.78 ± 1.62 a	174.56 ± 1.42 b	123.45 ± 1.11 b

Different lowercase letters within the same parameters show significant differences ($p < 0.05$). Data are represented as means ± SD (n = 4). Different letters indicate significant differences between treatments at $p < 0.05$ according to Duncan test.

Some of the heavy metal contents, including Pb, Zn, Cu, and Cd, were measured in sunflower plants as affected by inorganic and organic fertilizers (Table 6). The results in this table show that sugarcane bagasse ash (SBA) treatment scored the highest value of Pb, Zn, and Cu (42.72, 385.64, and 230.53, respectively), followed by compost (34.28, 260.04, and 150.63 respectively), while the inorganic application had the lowest values (24.12, 121.68 and 79.29 respectively). Cd, in contrast to Pb, Zn, and Cu, exhibited a significant reduction when treated the sunflower plant with organic treatments. The highest decrease was 44% lesser than the control in sunflower plants under CCB application.

4. Discussion

The vegetative growth period is a sensitive stage to nutrition and water conditions for any plant. Plant growth and yield are directly proportional to the sufficiency of its nutrient supply. The findings of this study show that different types of fertilizers have a positive effect on the growth of sunflower plants. Rotkittikhun et al. [38] demonstrated the ability to use compost in conjunction with lower doses of inorganic fertilizer to boost biomass production in field crops such as lemongrass and sunflower. The use of inorganic and organic fertilizers, either alone or in combination, significantly influenced chickpea grain and biomass yield [39].

Naik et al. [40] showed that application of compost coupled with biofertilizer increase plant growth by releasing trace elements, antioxidants, bioactive substances, and exopolysaccharides.

Organic fertilizers can stimulate substrate nutrients, improve both the chemical and physical characteristics of the soil, stimulate nutrient absorption by plants, increase the amount of nutrients, and promote vegetative and reproductive growth [41,42]. The results of this experiment revealed that organic fertilizer applications have a positive effect on the vegetative growth of sunflower plants, and there is a high correlation between the type of fertilizer used and the growth rate. Application of NPK fertilizers may adversely affect soil chemical, physical, and biological properties, in addition to soil health. The negative effects of chemical fertilizers, combined with rising prices, have resulted in a massive increase in interest in the use of organic fertilizers as a source of nutrients. [43]. Organic and inorganic fertilizers both supply plants with the nutrients they require to grow healthy and strong. However, each contains different ingredients and provides these ingredients in different ways. Organic fertilizers produce an appropriate growing environment gradually, whereas inorganic fertilizer provides immediate nutrition [1].

Organic fertilizers stimulate the rate of metabolism inside the plant and increase metabolite movement from the roots to the leaves, potentially enhancing plants' mineral content [44,45]. Organic fertilizers improve the physical properties of soil and lower pH, which affect the availability of soil nutrients for plant uptake, as well as enhance plant growth [1]. Concerning the studied vegetative growth parameters of sunflower plants (shoot/root fresh and dry mass and shoot/root length), there was a significant difference between the fertilizers treatments used. In all the vegetative growth parameters measured, organic fertilizers showed the highest value, and NPK inorganic fertilizer had the lowest value. These findings are consistent with those of [46], which found that the performance of tomato growth in vermicompost-amended soil was better as compared to inorganic fertilizer-amended soil. Furthermore, [47] investigated the dry mass content of various vegetables and realized that organically grown vegetable crops had higher dry mass content than inorganically grown vegetable crops.

Also, an increase in photosynthetic pigment content (PPC; Table 3) indicates a significant increase in fresh and dry mass as well as plant growth parameters. The obtained results are consistent with those reported by [48–50].

According to [51], organic fertilizer increases the chlorophyll level of wheat leaves, and the SPAD value can reach 60.1, which is 58% higher than CK. Furthermore, the obtained data are consistent with those reported by [52], who discovered that organic fertilizers derived from various plant sources increased the Pn of crisp jujube leaves to varying degrees and altered the transpiration rate of the leaves.

Analogous to these findings, Dineshkumar et al. [53] showed that increased chlorophyll content under the application of organic fertilizers, even at a lower level, could be due to the consortium cooperative impacts, which improve plant N, P, and K uptake, resulting in higher chlorophyll content.

Organic fertilizers applications had an additive effect on enzymatic antioxidant defense systems, including the CAT, SOD, and POD activities, as well as non-enzymatic antioxidant defense systems such as carotenoids, glutathione, soluble protein, and soluble sugar when compared to the NPK inorganic treatment. Plant cells generate reactive oxygen species as a result of normal metabolic processes or under environmental stress conditions such as nutrient deficiency. Aina et al. [54] noticed that using organic fertilizer instead of inorganic fertilizer raised the level of secondary metabolites such as phenolics and flavonoids and the activity of plant antioxidants. According to Hosseinzadeh et al. [55], organic matter increases the uptake of nutrients, total enzyme activity, cation exchange capacity, and water holding.

Organically grown sunflowers (CCB and SBA) had lower phenolic compound content than inorganic fertilizer-grown plants (inorganic). This strongly suggests that the source of nitrogen influences the level of phenolic compounds formed in sunflowers; when nitrogen is abundant, plants will primarily produce nitrogen-rich compounds such as proteins, according to the C/N balance hypothesis. When nitrogen is limited, metabolism changes toward non-N-containing secondary compounds like terpenoids and phenolics [56].

The findings corroborate the idea the dispersal of nutrients from different kinds of fertilizers depends on the type of nitrogen applied. The proportional variations in the nutrient release from different fertilizers may result in different C/N ratios in plants, which in turn may result in differences in secondary metabolite production [56]. These findings imply that using chemical fertilizers can boost the synthesis of secondary metabolites in sunflowers. Organic fertilizers contain nitrogen that is linked to organic matter and released gradually.

Organic fertilizers significantly enhanced the minerals content and heavy metal content of sunflower plants in this study. Our finding showed that organic fertilizers significantly increased potassium (K), Calcium (Ca), and magnesium (Mg) content in sunflower plants when compared to inorganic fertilizer as a control. This increase in minerals could be attributed to an increase in the activity of microbes, which increases the availability and uptake of nutrients, as described by [57]. Improvement of soil physical properties and microbial action contribute to enhanced nutrient availability from organic sources.

The current findings contradict those of Asante et al. [58], who found that high nitrogen application had no effect on the nutrient accumulation (N, P, K, Ca, and Mg) in the leaves of *M. oleifera*.

Hanchimani [59] maintained that *M. oleifera* responded well to fertilizer application, resulting in increased availability of nutrients in the pods and, as a result, raised yield.

Mohamed et al. [60] found that organic fertilizer treatments enhanced macro and micronutrient availability over inorganic fertilization, which is in line with our results. Furthermore, Azad et al. [61] observed that the prescribed amount of inorganic fertilizer enhanced the content of N, P, K, and S in wheat grains significantly.

Banerjee [62] revealed that excessive chemical fertilizers application boosts the agricultural sector's influence on the ecosystem and reduces the sustainability and maintenance of agricultural practices.

5. Conclusions

Fertile soils are essential for crop production, and soil amendment with organic and inorganic fertilizers contributes to boosting crop yields. The study revealed that different kinds of fertilizers (inorganic fertilizer, compost coupled with biofertilizer, sugarcane bagasse ash) had significant effects on physiological and metabolic alterations in sunflower plants by enhancing plant growth, metabolites, activities of enzymatic antioxidants and minerals and heavy metals content of sunflower plant. As a whole, organic and inorganic fertilization improved the physio-biochemical characteristics of the sunflower plant. Furthermore, using organic and inorganic fertilizers would be highly beneficial not only in terms of boosting crop productivity but also in terms of enhancing soil fertility and crop quality. At the same time, organic wastes will not become a source of environmental pollution. Organic wastes can be effectively used in the production of high-quality, low-cost organic fertilizers that can easily be used to boost the yield of one of the most important oilseed crops in the world, consequently contributing to the solution of the problem of manufactured oil shortage as well as the problem of safe agronomic waste disposal. Organic fertilizers are undeniably beneficial, but when used in excess, they can harm the environment just as much as inorganic fertilizers. Using organic fertilizer in conjunction with inorganic fertilizer increases the activity of microbes, nutrient absorption efficiency, and plant accessibility to essential nutrients. To avoid environmental problems, more research should be conducted to find new application techniques and a suitable amount of fertilizer application that takes into account crop type, soil properties, and region conditions.

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