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NUTRITIONAL VALUE AND MINERAL CONCENTRATIONS OF SWEET BASIL UNDER ORGANIC COMPARED TO CHEMICAL FERTILIZATION

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ABSTRACT

Safe production of leafy vegetables is more important than other horticultural crops due to their significant contribution in health promotion of consumers. In present study, the growth and nutritional quality of sweet basil plants (*Ocimum basilicum* L.) were evaluated under some organic compared to chemical NPK fertilizing treatments. A non cultivated soil was used in the experiment, and treatments were arranged in completely randomized design with four replications. Manure in 20 and 40% of pot volume, vermicompost in 20 and 40% of pot volume, biophosphate, NPK fertilizer and a no fertilizing control were applied to plants. The results showed that plant growth and biochemical characteristics were differently affected by organic and chemical fertilization treatments. Vermicompost particularly in 20% of pot volume resulted in highest growth and quality parameters, while the lowest values recorded in 40% manure treatment. The leaf content of ascorbic acid, essential oil, protein and minerals (P, K, Mn, Cu) were highest in 20% vermicompost, whereas leaf content of Mg, Fe and Zn was highest in 40% vermicompost treatment. Leaf nitrate content was significantly reduced in all organic treatments than NPK fertilization. Biophosphate also improved the yield and quality traits compared to unfertilized plants, while manure in both quantities reduced most of the traits (leaf vitamin C, oil yield and protein content) except the content of phenols. This indicates that processing as vermicompost can be a better way of manure application for basil production.

Key words: fertilizer, nutritional value, organic culture, plant nutrition, protein, vegetable

INTRODUCTION

During last century environmental pollutions and contamination of agricultural food commodities have been linked to unsustainable approaches to production policies [Tabosa et al. 1990, Citak and Sonmez 2010, Souri 2016]. Application of various fertilizers as well as uncontrolled use of pesticides has caused serious health challenges in ecosystem levels. In developing countries including Iran there is no control over application of chemicals in food produc-

tion processes, and still it has not a priority order [Naiji and Souri 2014, Souri 2016]. So, various public health and many unknown illnesses have arised in such countries. In many populated regions of developing countries, due to application of wastes and high use of low quality chemical fertilizers, the health conditions of soil, water, air and consequently agricultural food products are in question [Khalid et al. 2006, Citak and Sonmez 2010]. In many agricultural



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food products, the residues of chemicals and pesticides are over standard limits [Dahama 2007]. Leafy vegetable crops are more susceptible to such contaminations [Naiji and Souri 2014], whereas they are among the most important sources of beneficial nutrients in human diets, so their safe production with acceptable quality can significantly improve health and immune systems of human [Souri et al. 2016].

Organic production attitudes and sustainable practices inevitably are important measures to solve those raised challenges and to slow the rate of environmental degradation [Dahama 2007, Citak and Sonmez 2010]. Sustainable field management consisted of improving biodiversity, and soil-plant-microorganisms relationship, which is fully dependent on soil organic matter content and water supply.

In dry climates, due to rapid oxidation reactions, organic matter decline sharply after wetting periods. On the other hand, in many semi arid and arid parts of the world organic vegetable production is relatively more difficult than European wet regions [Tabosa et al. 1990, Singh and Guleria 2013, Souri 2016]. Problems associated with sufficient organic matter and manure supply, high rate of organic matter turnover, infertile and poor structured soils, inadequate water supply and water quality, hot weather and periodic and devastating droughts are some challenges raise to constrain organic production in arid regions.

Organic and biological fertilizers have been a part of agricultural production systems for centuries [Marschner 2011]. However, during last century chemical fertilizers due to immediate effects on plant growth and yield have replaced organic fertilizers application. Nowadays, organic farming tries to generalize the application of organic and biological fertilizers instead of unsustainable approach of chemical fertilizers [Dahama 2007]. In present study, some organic fertilizing practices were compared with chemical

fertilizer application on sweet basil nutritional value and mineral composition of plant shoots.

MATERIAL AND METHODS

This study was done in Faculty of Agriculture, Tarbiat Modares University, from June until August of 2014 using pot experiment and under greenhouse conditions. A soil with non cultivation background was used in the experiment. The physicochemical characteristics of soil were presented in Table 1. The soil had silty-loam texture with low levels of organic carbon and nitrogen. An amount of 120 mg N kg⁻¹ soil in form of urea was used to increase soil N status before experiment.

The experiment was arranged in completely randomized design with seven treatments and four replications (4 liter plastic pots). Treatments were: cow manure in 20% and 40% of pot volume, vermicompost in 20% and 40% of pot volume, biophosphate (Barvar 2), chemical NPK fertilizer and a no fertilized control. The chemical NPK fertilizer (N:20; P:15; K:20) was applied in an amount of 150 kg ha⁻¹.

The soil was disinfected for four weeks using solarization before applying treatments. For this purpose the soil was wetted and covered by thin layer plastic cover, while checked regularly. Black plastic pots with 4 kg volume were used and filled with respected soil treated. A local seed of sweet basil (the Karaj ecotype) was used. About 50 seeds were sown in each pot and then the pots were randomly arranged under open air condition with regular changing their position. After germination and in 3–4 leaves stage they were thinned to 7 plants per pot. The irrigation of pots was done daily by weighing the pots and based on 80% of soil water holding capacity. During July pots were partially protected from direct sunshine using green plastic nets.

Table 1. Physicochemical characteristics of soil used in experiment

Soil texture	Sand (%)	Silt (%)	Clay (%)	EC (dS m ⁻¹)	pН	Organic arbon (%)	N (mg kg ⁻¹)	P (mg kg ⁻¹)	K (mg kg ⁻¹)
Silty-loam	31	49	20	0.415	7.17	0.42	5.1	15.2	256

	pН	EC (dS m ⁻¹)	Organic carbon (%)	Nitrogen (%)	Phosphorus (%)	Potassium (%)
Cow manure	9.01	4.16	52	1.07	0.24	2.18
Vermicompost	4.69	8.19	37	1.11	0.9	1.28

Table 2. Chemical properties of cow manure and vermicompost used in study

The vermicompost was purchased from Zist Salem Kimia Company, Tehran-Iran. The cow manure was used after 8 months composting. The physiochemical properties of cow manure and vermicompost are presented in Table 2.

The biophosphate fertilizer (Barvar 2) contained two species of phosphorus solubilizing bacteria namely *Pseudomonas putida* (Strain 13) and *Pantoea agglomerans* (Strain P5) which were prepared from National Genetic Institute, Tehran-Iran. The population of each bacterium was 10⁸ active bacteria per g inoculants. Biophosphate was applied as seed inoculation before sowing under dark conditions. Seeds were inoculated for 30 min in a solution of 1g biophosphate to10 ml distilled water. The seeds were then dried in shadow and after dryness they were sown in pot soil.

Plants were grown for thirteen weeks and during growth period and/or at harvest various growth and biochemical traits were measured. At harvest, leaf SPAD value was measured using a portable SPAD meter (model 502 Plus, Illinois, USA), and total shoot fresh weight was determined using a precise balance and then calculated per single plant.

Leaf biochemical characteristics were also measured with different methods. Leaf L-ascorbic acid was determined using dichlorophenol indophenols method in which 10 g of leaf materials were manually crushed in mortar with 10 ml metaphosphoric acid 6%, thereafter the mixture was centrifuged at 9000 rpm for 10 min at 4°C. Five ml of supernatant was diluted to 10 ml using 5 ml metaphosphoric acid 2% and the solution was titrated using dichlorophenol indophenols 0.2 mM. The amount of L-ascorbic acid

content of plants was calculated from dilution factors and the amount of consumed reagent and the reagent equivalent, after calibrating the method using different L-ascorbic acid concentrations.

Leaf carotenoid contents were determined following method used by Lewis et al. [1998]. Shoot essential oil was extracted by Clevenger apparatus and water distillation method. For this purpose, plants were harvested at a marketable quality for fresh vegetables, in which 30 g of dried samples was used for determination of essential oil concentration. The essential oil yield was calculated using pot dry matter of each replicate and respected oil percentage. Leaf protein content was determined following Bradford method, and phenolic compound were determined by method used by Ghazghazi et al. [2010].

Leaf nutrient concentrations were measured by various methods. Nitrogen was determined using kjeldahl method, potassium using flame photometry, phosphorus using spectrophotometric molibdate-vanadium method, calcium, magnesium, iron, zinc, manganese and copper using atomic absorption spectroscopy method. Nitrate was determined potentiometrically by which 10 g of plant leaves were homogenated in 50 ml deionized water for two minutes using a pestle and mortar. The homogenates were transferred to a 100 ml centrifugal tube, and after centrifugation the supernatant was used for determination of nitrate concentration using flow-injection-analysis after Cd-catalysed reduction to N₂O.

The data were analyzed using SPSS (16) and comparison of means was done using Duncan' multiple range test at 5% level.

RESULTS

The results showed that fertilization treatments had significant effects on many morphophysiological traits measured in this study. Vermicompost treatments of 20 and 40%, as well as NPK fertilizer had the highest leaf SPAD value (tab. 3). The significant lowest leaf SPAD was in plants which were treated with 40% manure. Plant shoot fresh weight which is the actual yield was significantly highest in vermicompost 20% which showed no significant difference to vermicompost 40% and NPK fertilizer (tab. 3). Biophosphate and manure 20% were the next which also showed significant higher records compared to control. The lowest shoot fresh weight was in manure 40% which showed no significant difference compared to unfertilized control (tab. 3). There was no significant difference among treatments regarding carotenoids content (tab. 3). Plants which were treated with vermicompost 20%, NPK and vermicompost 40% treatments had the highest ascorbic acid content (tab. 3). The lowest ascorbic acid was in those plants which were treated with 40% manure that showed no significant difference with unfertilized control. Vermicompost 20% resulted in highest percentage of essential oil, despite NPK treatment and vermicompost also increased this trait compared to unfertilized plants (tab. 3). Oil yield was highest in vermicompost treatements. The NPK and biophosphate also resulted in higher oil yield compared to unfertilized control. The lowest oil yield was in 40% manure which showed no significant difference to unfertilized control and manure 20% (tab. 3). Determination of nutrient elements in leaf of sweet basil showed that the highest leaf N concentration was in NPK treatment which showed no difference with vermicompost 40% (tab. 4). Plants in unfertilized control and manure treatments showed the lowest N concentration. All fertilizer treatments resulted in significant higher leaf P concentration compared to unfertilized control plants (tab. 4). The highest P concentration was in vermicompost 20% which showed no difference to NPK treatment. Vermicompost 20 and 40% and NPK treatments showed the significant higher leaf K compared to unfertilized control (tab. 4). Vermicompost 40 and 20%, manure 20%, and NPK treatments resulted in higher leaf magnesium concentration, whereas leaf calcium concentration was significantly higher in NPK and vermicompost 20% treatments (tab. 4).

Table 3. Physiochemical characteristics of sweet basil plants under different organic fertilization treatments. Data are average of four replications ±SD

Treatments	SPAD	Shoot FW (g plant ⁻¹)	Carotenoids (mg 100 g ⁻¹ FW)	L ascorbic acid (mg 100 g ⁻¹ FW)	Essential oil (%)	Oil yield ml pot ⁻¹
Control	29 ±0.3b	6.2 ±0.4c	38.5 ±4.3a	25.3 ±2.7c	0.67 ±0.09c	0.85 ±0.07c
NPK fertilizer	34 ±1.7a	8.5 ±0.9ab	37.5 ±3.1a	36.0 ±1.1a	1.0 ±0.12b	1.28 ±0.17b
Manure 20%	30 ±1.1b	$7.7 \pm 0.8b$	40.1 ±3.1a	27.6 ±2.6b	0.78 ±0.09bc	0.91 ±0.10c
Manure 40%	25 ±2.6c	5.7 ±0.4c	40.5 ±4.0a	23.7 ±1.6c	0.66 ±0.05c	0.73 ±0.09c
Vermi 20%	35 ±1.4a	9.8 ±0.9a	41.3 ±2.9a	36.3 ±2.3a	1.35 ±0.22a	1.7 ±0.18a
Vermi 40%	34 ±1.5a	8.8 ±1.1ab	40.6 ±3.7a	34.2 ±1.7a	1.03 ±0.10b	1.58 ±0.12a
Biophosphate	32 ±1.5b	7.6 ±0.7b	38.4 ±2.9a	28.7 ±1.9b	0.86 ±0.08bc	1.16 ±0.18b

Data with at least one common letter in each column are not significantly different at 5% level of Duncan multiple range test

Table 4. Leaf nutrient minerals concentrations affected by various organic fertilization treatments. Data are average of four replications ± SD

Treatments	N (%)	P (%)	K (%)	Mg (%)	Ca (%)	Fe (mg kg ⁻¹)	$Zn \atop (mg \ kg^{-l})$	$\begin{array}{c} Mn \\ (mg \; kg^{-l}) \end{array}$	Cu (mg kg ⁻¹)
Control	1.5 ±0.11c	0.36 ±0.05d	2.3 ±0.25b	0.32±0.04b	2.8±0.65b	123.3±15d	78.5±9b	161.5±18b	12.3±1.5b
NPK fertilizer	2.1 ±0.14a	0.5 ±0.02ab	3.1 ±0.14a	0.37 ±0.03a	3.4 ±0.43a	138.3 ±13c	83.2 ±8b	177.2 ±17b	13 ±1.8b
Manure 20%	1.4 ±0.12c	0.43 ±0.06c	2.6±0.15b	0.39 ±0.02a	2.9 ±0.51b	152.2 ±17b	75.2 ±10b	168 ±18b	10 ±1.8c
Manure 40%	1.4 ±0.10c	0.45 ±0.08bc	2.5 ±0.12b	0.33 ±0.04b	2.6 ± 0.60	148.5 ±13b	78.5 ±12b	164.5 ±17b	9.5 ±1.3c
Vermi 20%	1.8 ±0.11b	0.55 ±0.05a	3.3 ±0.30a	0.38 ±0.03a	3.3 ±0.37a	167.8 ±15a	95.5 ±14a	200 ±12a	15 ±0.8a
Vermi 40%	1.9 ±0.10ab	0.46 ±0.05b	3.2 ±0.21a	0.4 ±0.03a	3.2 ±0.30ab	171 ±10a	100.5 ±13a	188 ±20ab	14.8 ±1.0a
Biophosphate	1.6 ±0.11bc	0.47 ±0.06b	2.6 ±0.20b	0.32 ±0.06b	2.8 ±0.35b	127.8 ±11cd	82.8 ±12b	167.2 ±13b	12 ±1.4b

Data with at least one common letter in each column are not significantly different at 5% level of Duncan multiple range test

Table 5. Leaf nitrate content, total phenolic compounds and protein concentration of basil plants under various organic fertilization treatments. Data are average of four replications ±SD

Treatments	Nitrate (mg kg ⁻¹ FW)	Total phenols (mg galic acid g ⁻¹ FW)	Leaf protein con (% DW)
Control	1001 ±94b	1.46 ±0.21b	17.5 ±1.9bc
NPK fertilizer	1504 ±166a	1.38 ±0.11b	19.7 ±3.8b
Manure 20%	659 ±65c	1.73 ±0.09a	16.9 ±2.6bc
Manure 40%	617 ±115c	1.87 ±0.08a	15.4 ±2.0c
Vermi 20%	898 ±148b	1.31 ±0.11b	24.9 ±2.5a
Vermi 40%	1004 ±120b	1.28 ±0.10b	25.0 ±3.5a
Biophosphate	761 ±158bc	1.4 ±0.12b	18.2 ±3.4bc

Data with at least one common letter in each column are not significantly different at 5% level of Duncan multiple range test

Leaf micronutrient concentrations were significantly improved by vermicompost levels compared to unfertilized control plants (tab. 4). All fertilizer applications improved iron concentrations of leaf compared to control plants (tab. 4), however regarding Zn, only vermicompost treatments were significantly increased leaf Zn concentration compared to unfertilized control. Similarly, leaf iron concentration was highest in vericompost treatments followed by manure treatments and NPK fertilizer (tab. 4). The low-

est leaf Fe was observed in unfertilized control plants. All fertilizer treatments recorded higher concentration of leaf Mn, whereas statically Mn only in vermicompost 20% had significant higher concentration compared to unfertilized control. Determination of copper in basil leaves revealed that the significant highest concentration was in vermicompost treatments (tab. 4). There was no significant difference between control and biophosphate or NPK treatment. The significant lowest leaf Cu concentration was in

manure treatments (tab. 4). Although biophosphate has slightly increase all nutrients concentration compared to unfertilized control, but statically it had only significant higher concentration only for phosphorus not other nutrients.

Determination of leaf nitrate content (tab. 5) showed that the highest nitrate content of leaves was observed in NPK treatment followed in vermicompost 20 and 40% and unfertilized control plants. The significant lowest leaf nitrate content was in plants treated with 40% and 20% manure treatments, respectively.

Leaf phenols (tab. 5) were significantly higher in manure treatments, and the lowest phenols were recorded in those plants that were treated with vermicompost 40%, despite it had no significant difference to other treatments except to two manure levels. The highest leaf protein concentration (tab. 5) was recorded in vermicompost 20% and 40% treatments. The significant lowest leaf protein concentration was in those plants treated with 40% manure treatment.

DISCUSSION

In present study it was shown that fertilization of some organic fertilizers had quite various effects on sweet basil growth, productivity and nutritional values. The yield and many quality parameters were at best increased by 20% application of vermicompost. There was also significant highest essential oil in vermicompost treatments. The superiority effects of vermicompost on other organic and inorganic fertilizers have been well established in many studies [Arancon et al. 2005, Zaller 2007, Singh and Guleria 2013]. Vermicompost is one of the most effective organic fertilizers which have also high rates of application in conventional farming systems. It is one of the best fertilizers among all fertilizers for cropping systems [Arancon et al. 2005, Norman et al. 2005, Rai et al. 2013]. This organic fertilizer contains high amounts of N, P, K, Mg and many other essential (micro) nutrients in available form. Vermicompost is rich in macro and micro nutrients, vitamins, enzymes and plant growth regulators which in soil cause faster and better plant growth with higher marketable yield [Suthar 2009, Naiji and Souri 2014]. It has also high cation exchange capacity (more than 50 meq 100 g⁻¹) which is very important feature in soil fertility [Norman et al. 2005, Suthar 2009, Rai et al. 2013]. In organic systems, application of vermicompost through increasing the activity or soil microorganisms and supply of sufficient N, K, P, Mg and other nutrients can increase soil fertility and crop productivity [Singh and Ramesh 2002].

In garlic, vermicompost increased plant growth, yield and quality via faster bulb formation and longer period of growth and bulb filling [Suthar 2009]. Application of vermicompost to soil can improve soil infiltration, aeration, water holding properties, microbial activity and amounts of nutrient in soil solution [Atiyeh et al. 2000, 2002, Singh and Ramesh 2002, Arancon et al. 2005, Norman et al. 2005]. Production and release of plant promoting substances including growth regulators and semi-phytohormones and various enzymes play role in higher growth rate of many plants under vermicompost application [Vinotha et al. 2000, Zaller 2007]. Vermicompost create a best environment for activity of a range of beneficial microorganisms, and generally higher soil microorganism activity associated with vermicompost application. These organisms are a major source of various biostimulants including organic acids, amino acids, vitamins, effective enzymes and semihormonal substances that strongly can affect growth and improve plant productivity and nutrient uptake [Dahama 2007, Citak and Sonmez 2010]. The released compounds by these microorganisms mainly organic acids, amino acids and phenolics contributes in medium pH reduction and forming chelating bonds with unavailable forms of nutrients, making them more available to plant roots [Marschner 2011, Souri 2016].

Chemical NPK fertilize also resulted in higher values of many growth and quality parameters compared to control. In such soil with rather low level of fertility (tab. 1), the plant response to NPK fertilization is quite expected. This is the best promotion of farmers for more application of chemical fertilizers, which during last decades in conventional farming has resulted in serious challenges including reduced soil quality and fertility, contamination of surface and deep waters, reduced biodiversity and disruption in

ecosystem activities [Socolow 1999, Souri 2016]. Application of different levels of chemical fertilizers generally results in increase of plant growth, yield and nutrient concentrations [Singh and Ramesh 2002, Anwar et al. 2005, Singh and Guleria 2013]. However, in present study NPK fertilization was the next after vermicompost regarding plant growth and quality improvement, indicating superior effects than NPK application. Meanwhile other organic fertilization treatments recorded lower growth and quality values compared to chemical NPK fertilization. In general the yield reduction of organic products is about 10% which can be compensated by their higher quality (through higher nutrients and secondary metabolites) as well as lower energy and inputs [Dahama 2007, Bahadur et al. 2009, Michael et al. 2010, Rai et al. 2013].

Basil leaf phytochemicals were significantly influenced by various fertilization treatments (tabs 3 and 5). Vermicompost application resulted in highest ascorbic acid, essential oil and protein content of leaves, while phenolics was higher in manure treatment and nitrate was highest in NPK fertilization. It has been shown that organic products of vegetables and fruits have nearly 20% more of phytochemicals with anticancer properties [Dahama 2007, Rai et al. 2013]. They have also better accepted flavor compared to conventional products [Anwar et al. 2005, Bahadur et al. 2009, Blaise et al. 2005, Michael et al. 2010]. The highest vitamin C in cabbage, tomato and spinach was observed in application of vermicompost, or integration of manure and chemical fertilizers [Rai et al. 2013, Zaller 2007, Citak and Sonmez 2010].

In spinach the highest plant height and stem diameter were obtained from chemical fertilizer application, and the highest vitamin C was observed in organic manure application treatment [Citak and Sonmz 2010]. Application of organic fertilizers has been reported to increase essential oils of basil [Anwar et al. 2005, Singh and Guleria 2013]. Nevertheless, different plants may have various response to chemical and organic fertilizers; as the highest yield and nutrient uptake in radish was in 100% chemical fertilizer (NPK), in amaranths from 2.5 t ha⁻¹ poultry manure + chemical fertilizer, and in spinach in 5 t ha⁻¹

organic wastes + chemical fertilizer [Islam et al. 2011]. In present study a similar results was confirmed.

In this study, manure particularly in 40% rather than 20% volume resulted in no improvement or restriction in plant growth and quality parameters. Lower plant growth due to organic manure has been shown in other studies [Eghball and Power 1999, Badran and Safwat 2004, Anwar et al. 2005, Arancon et al. 2005]. This probably is due to various factors associated with organic manure including its microbial and pathogen load, chemical properties including EC and pH, and more importantly depletion of such composted materials from N, as its physicochemical characteristics favorite N volatilization. The "pot" effect should not also be ignored in this regard; as a given plant may have different responses to manure application in pot and field experiments. It has been reported that available N in first year for cow manure is about 40-50%, which significantly reduces in following years [Eghball and Power 1999, Atiyeh et al. 2000, Bahadur et al. 2009]. The manure which was used was composted and at time of analysis it contained 1.1 % nitrogen; however the relative content of N with regard to the absorbtion affinity of decomposed material for ammonium or nitrate must be considered. In addition, the soil under this treatment probably has been depleted of N by efficient competent of microbes and manure organic fixing properties [Souri 2016, 2017]. This is more supported as plants in manure treatment particularly with higher manure volume of pot showed chlorosis symptoms similar to N deficiency symptoms. The effects of manure under field soil conditions may be quite different and in lettuce the highest leaf number, plant height, marketable yield and leaf dry weight were achieved by poultry and then cow manure [Masarirambi et al. 2012].

In this study some growth and quality traits were improved by biophosphate application compared to unfertilized control. There has been positive response of many crops and under various conditions to biophosphate fertilizers particularly in vegetable crops [Badran and Safwat 2004, Bahadur et al. 2009]. These fertilizers contain a range of beneficial microorganisms that produce organic acids and amino

acids that reduce medium pH or forming chelate bonds with less available nutrients for better plant root uptake. However, various bioenzymes have also released from these microorganisms such as phosphatases which play important role in p availability for plant roots [Vinotha et al. 2000]. Biological fertilizers are economic and can improve soil sustainability, long term soil fertility and environmental protection. On the other hand, products from organic fertilizing systems are safer and healthier to consumers and environment [Dahama 2007].

CONCLUSION

In this study basil plants in a rather dry climate had quite different response in terms of growth, quality and nutrients concentration to various organic fertilization compared to NPK application. Application of eight months composted cow manure resulted in poor plant performance while vermicompost resulted in significant better plant growth conditions. The highest value of many growth and quality traits was recorded from vermicompost treatments. Biophosphate was also improved some traits compared to control, but still much lower value compared to NPK fertilizer. The results indicate that vermicompost was superior to other forms of organic fertilizers, and processing of manures in form of vermicompost can probably mitigate its negative effects on plant growth and productivity.

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