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Efficiency of Some Eco-friendly Treatments for Controlling Root-knot Nematodes on Banana Plants

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Abstract: The nematode populations in the soil and the roots of the banana plants were significantly reduced in the 2020–21 and 2021/22 growing seasons when different organic matters, such as compost, cow manure, and plant residue, were added to the organic farm of Williams banana at a rate of 1.2 kg/m^2 or drenched in a diluted 3:200/fed as recommended dose of vermicompost, *Azotobacter chroococcum*, and humic acid. Compost, as a banana-rich organic fertiliser at a rate of 1.2 Kg/m^2, caused the highest decrease in root knot nematode infestations, as well as the highest increase in vegetative growth pseudostem height and girth (cm), leaf area (cm), number of green leaves at bunch shooting, and period to bunch shooting (days); yield parameters; fruit quality "Total soluble solid (TSS), Vitamin C, protein",total sugar, chemical components of flavonoid and total phenol. There were no evident significant changes in plant growth characteristics across the other treatments.

Key words: Banana, Compost, humic acid, vermicompost, *Azotobacter chroococcum* and root-knot nematodes.

1. Introduction

Banana (*Musa spp*.) is a perennial, vegetative propagated crop, and the planting material used has a significant impact on plantation production **(Macharia *et al*., 2010)**. They grow in tropical settings with average temperatures of 27°C and moist, well-drained soil **(Amugune *et al*., 2007)**. Banana comes in fourth worldwide as the most valuable crop **(Muchui *et al*., 2013)** after rice, wheat and maize in terms of guaranteeing food availability in developing countries. Traditionally, sucker-derived planting materials have been utilized for growing banana plantations, but untreated suckers carry the danger of spreading pests like plant-parasitic nematodes. Banana crop, one of the most significant fruit harvests in Egypt, which is also the most significant due to its high nutritional value and abundance in vitamins A, B, and mineral salts. The vast majority of Egyptians consume bananas. The overall production of the 69,770 acres planted with productive bananas in Egypt is 1,292.8381 ton, with an average productivity per acre of 18.53 ton during the growing season 2021 **(Zedan and Helme, 2021)**.

Root-knot nematodes are a significant group of plant-parasitic nematodes that have an impact on crop productivity and significantly lower the quality of food. *Meloidogyne incognita* (Kofoid and White) Chitwood is one of the most prevalent and significant plant parasitic nematodes in tropical and subtropical areas of the world **(Sasser *et al*., 1983; El-Nagdi, 2001; Eissa *et al*., 2005;).** Its infection causes root galls in many crops and impede normal uptake of water and nutrients **(Mokbel *et al*., 2006 and Wachira *et al*., 2009)**. The diseases root rot and root knot nematode have a significant impact on banana productivity **(Khan and Hassan, 2010)**. In banana orchards, the root-knot nematode attracted attention **(Ragab *et al*., 2016 and Rahman *et al*., 2014)**.

The application of organic and bio-fertilizers has potential for bananas in the long run because they are sources of organic matter, vital nutrients, amino acids, natural hormones, antibiotics, and B vitamin complex. They also improve the physical and chemical characteristics of the soil **(Nijjar,1985; Kannaiyan, 2002; Kimenju *et al*., 2004)**. Organic amendments are essential for managing banana parasitic nematodes by releasing chemicals during their decomposition that are harmful to worms **(Wang and Hooks, 2009; Risede *et al*., 2010, Khan and Hassan 2010)**.

The use of biofertilizers as a Plant Growth Promoting Rhizobacteria (PGPR) resulted in increased N, P, and K% at the leaves of banana plants for controlling nematode-destroying fungi **(Bouajila and Sanaa, 2011; McSorley, 2011 and Hamdy and Mahmoud, 2014)**.

Due to its properties, such as nitrogen fixation, secretion of substances that promote plant growth, vitamins, antifungal metabolites, phosphate solubilization, soil aggregation, and tolerance to pesticides, the use of free nitrogen fixing bacteria, *i.e*., *Azotobacter* sp., as a bioinoculant is widely used for a wide variety of different plants to control nematode predators **(Inomdar *et al*. 2000; Shaheen et.al., 2009 and Mahfouz, 2011)**. **Abd El-Hamid, Marwa *et al*. (2010)** discussed the creation of siderophore, indol acetic acid (I.A.A), and hydrogen cyanide (HCN). Plants belonging to the genera Agave have reportedly shown biocidal activity against nematodes from the organic waste of goats and lambs **(Botura *et al*., 2011; Domingues *et al*., 2010; Silveira *et al*., 2012; Renčo, 2013)**.

The development of agricultural practices that integrate biofertilizers and natural products has been reported to be a method of increasing sustainable food production, the soil's ability to contain water and fertility, which promotes healthy plants and increases their resistance to nematodes **(Stirling *et al*., 2014; Thakur, 2014; Wezel *et al*., 2014 and Ragab et. al., 2016)**.

The aim objective of conducting this investigation is to evaluate the application of different safe organic matters and biofertilizer as natural products alternative to chemical pesticides to control root knot nematodes *in vivo* and to increase the quality and quantity of banana fruits under organic farming systems to achieve environmental sustainability for soil, agriculture and food safety.

2. Materials and Methods

Trials

1. Plant materials used during the investigation

a. Banana cultivar

Banana seedlings of Williams cultivar were purchased from the nursery of the Horticulture Institute Research Station in Qanater El-Khairiya, Qalyubia Governorate, Egypt.

b. Humic acid

Humic acid was obtained from the Central Lab. of Organic Agriculture (CLOA); ARC; Egypt, which used as soil drench separately at the rate of 3 lit/200 lit water/fed.

c. Compost

The extensive ripening compost was applied in this farm during the preparation of soil and before transplanting on the 2020/21 and 2021/22 growing seasons, respectively. Compost was obtained from the same farm, consisting of "extensive cow manure + plant residue and was applied at the rate of 1.2 Kg /m^2. Composting operation taken (2.5-3 months) to enhance soil with organic matter and enrich the fertility. This amount provides the following nutrients according to the soil analysis (Table,1) at Water and Environmental Res. Inst., ARC, Giza, Egypt. Data in Table (1) show the high concentrations of macro elements which improve the quality and quantity of the target crop.

Table (1). Main parameters characteristics of mature compost

Different Organic matter	Bulk weight (kg/m^3)	Dry Matter (g/L)	%Humidity	pH (10:1)	EC (10:1) dS/m	Soluble N (mg/kg)		C/N	Total N (%)	Organic Matter (%)	Organc Carbon (%)	Total P (%)	Total K (%)
						NH 4+	NO 3-						
Compost	1020	100	40	6.50	2.39	43	27	1:12	1.55	83.35	16.55	1.25	0.80
Cow manure	850	45.36	55.74	7.50	1.25	69	20	1.18	0.85	81.20	14.45	0.65	0.70
Plant residue	750	85.20	13.50	6.50	3.50	53	18	1.13	1.40	82. 60	17.60	0.80	0.60

d. Vermicompost

It was obtained from the Central Lab. of Organic Agriculture (CLOA); ARC; Egypt and used as the rate of 3 L/200 L water/fed. Vermicomposting was prepared according to the method of **(Znaïdi, 2002 and Morales-Corts *et al*., 2018)**.

This amount provides the nutrients (Tab., 2) according to the analysis carried out at Soil, Water and Environmental Res. Inst., ARC, Giza, Egypt. Data in Table (2) show the high concentrations of macro and micro elements which improve the quality and quantity of the target crop.

Table (2). Main parameters characteristics of vermicomposting.

pH (10:1)	EC (10:1) dS/m	C (%)	N (%)	C/N (%)	P (%)	K (%)	Fe (ppm)	Mn (ppm)	Zn (ppm)	Cu (ppm)
7.50	1.19	15.10	1.50	1:12	1.55	1.25	480	230	145	25.00

e. Soil analysis

A representative sample of a mixture of clay and sandy soils (1:1 w/w) was collected from Badr Center, Nubaria, Beheira Gov., Egypt. The collected soil for pot experiment was air dried, crushed and prepared to physical and chemical properties determinations according to the methods described by Piper (1966). The physical and chemical analysis of soil used in testing the activities of efficient different organic matter against knot rot nematode in vivo are given in Tab. (3).

Table (3). Some Physio-Chemical Properties of Giza Governorate Soil

Soil Compositions					
Physical Properties		Chemical Compositions		Exchangeable bases (mol (+) kg)	
Clay %	07.76	pH (Kcl)	07.90	Zn (ppm)	1.80
Silt %	19.35	Total Carbon (g/kg)	00.24	Mn (ppm)	3.10
Sand %	72.00	Total Nitrogen (g/kg)	17.82	K (ppm)	160.25
EC (dS. m -1)	01.60	P (ppm)	65.88	Cu (ppm)	1.05
ECE (cmolc-Kg-1)	13.80	CaCO3 %	05.00	CEC	5.85
Textural class	Sandy loamy	---------		Fe (ppm)	14.50

2. Isolation and detection of root-knot nematode (*Meloidogyne spp.*)

In order to detect infection with the root-knot nematode (*Meloidogyne spp*.) infection using conventional nematode testing, samples of 250 cm3 dirt was created from the rhizosphere 200g soil and 5g roots samples taken from the top 30 cm of each plot from the commercial banana fields at Badr Centre, Nubaria, Beheira Gov., Egypt, were immediately transferred to the Environmental Studies and Research Institute University of Sadat City.

Then, nematodes were collected from 250 cm^3 soil using Cobb's sifting and decanting method **(Fallis, 1943)**, followed by a modified Baermann's funnel technique **(Schindler, 1961)**, and counted. Eggs of *Meloidogyne* spp., were extracted from banana roots infected with the nematode using sodium hypochlorite solution **(Hussey and Barker, 1973)**. Second-stage juveniles (J2) were collected daily from eggs and stored at 15°C. The juveniles used in the experiments were less than 5 days old.

Identification of Meloidogyne spp.

According to **Taylor and Sasser (1978)**, adult female root-knot nematodes were isolated from galled banana plant roots and identified as *M. incognita* by inspection of their circular perineal pattern (PCP) and morphological traits by stereo microscope and used for further studies.

3 - Isolation and Identification of Azotobacter bacteria from soil around roots

Isolation was carried out using the method described by **Mahmoud (1997) and Ahmed (2005)**. Samples of soil (1g) was collected from a rhizosphere of banana and soybean plants were weighed and each sample was added to a flask containing 25ml sterile water, shacked periodically for approximately 3 hours, left to deposit. This soil suspension was streaked onto plates contained modified Ashby’s medium **(Abd-El-Malek and Ishac, 1968)**. The inoculated plates were incubated for 5 days at 25°C. Azotobacter bacterium (white milky, mucoid, colonies growth, morphology is large ovoid cells, negative gram stain) was identified according to **Becking (1974)**. Azotobacter bacterium was subcultured on slants of Ashby’s agar medium and stored at refrigerator. In addition used as Plant Growth Promoting Rhizobacteria (as indirect effect).

4. Field experiment

The Williams banana plants (*Musa cavendishii* L.), produced in a private orchard in Badr Centre, Nubaria, Beheira Gov., Egypt, were the subject of this investigation during the 2020/21 and 2021/22 growing seasons at 3.5 × 3.5 metres, plants were produced. Sand loamy soil with drip irrigation system was present **(El-Nagdi, Wafaa, 2001)**. The field was selected because its soil nematode population density was larger than the economic threshold level (> one juvenile/g soil **(Shanthi and Rajendran, 2006)**. In these experiments, all composted types were added at the rate of 1.2 Kg on dry basis/1 m^2) to the naturally infested soil in the field and irrigated daily. In all trails the soil of Williams banana cv. was drenched in diluted (3:200/fed) as recommended dose of vermicompost, *Azotobacter chroococcum* and humic acid. Plots without using any treatment were used to serve as control treatment. Three plots were used as replicates for each treatment. Each one composed of 3 ridges prepared as mentioned before **(Ahmed, 2013)**.

Plots were irrigated periodically by drip system from the well source and received all other normal organic agricultural practices. The recorded temperature under field conditions was around 25 ± 5o C, while the relative humidity was 60 ±10 R.H. Each treatment was added twice, on 7-21, April and 05 May.

The following variables were looked at:

a. Samples

Three samples of 5g roots and 200g soil were collected from each treated banana as well as untreated control after one week of each treatment for different examinations. Then, the samples were immediately transferred to Environmental Studies and Research Institute University of Sadat City, for the following studies. Females and eggs in roots per 5g were counted according to **Franklin and Goodey (1957).**

Reductions (%) of *M. incognita* population in soil and roots were determined according to the formula of Handerson and Tilton **(Puntener, 1981)**: Total population= numbers of juveniles in soil (Js2) + developing stages+females+egg mass

b. The development of plants and flowering

Data on the flowering and vegetative development included

According to **Murry (1960)**, the pseudostem's height (cm) and girth (cm) were estimated, and the leaf area using the third full-sized leaf (from the top) was calculated in (m^2). Number of green leaves at bunch shooting, period to bunch shooting (in days), and leaf area (leaf area = length × width × 0.8) were all noted.

c. There is some yield measurement

According to **Abou-Aziz *et al*. (1970) and Abd El-Naby (1988)**, the bunches were picked at the point of bunch maturity using the angulation criteria of fingers. The bunch weight (kg), hand weight (kg), and finger weight (g) were calculated at harvest.

d. Chemical components determination

1- Total proteins

Total proteins were determined by the method of **Bradford (I976)**.

2- Determination of Vitamin C

Vitamin C or ascorbic acid method **(Offor *et al*., 2015)** is based on measurement of the extent to which a 2,6-dichlorophenol-indophenol dye solution is decolorized by the presence of ascorbic acid.

3- Determination of total sugars

Total sugars were extracted from plants by 80% ethanol and were estimated by the phenol-sulphuric acid reaction of **Dubois *et al*. (1956) and Dubois *et al*. (2013)**. Total carbohydrate is expressed as :µg glucose / gm fresh weight. Since sugar beetroot consists of sucrose as major sugar content, sucrose calculated by difference **(Quin *et al* ,1980)**.

Sucrose % = Total sugar % - reducing sugars %.

4- Total soluble solid (TSS)

A total soluble solid (TSS in °Brix) of the banana juice was measured by the method described by **(A.O.A.C., 2005)**.

5-Quantification of total phenolics

The amount of total phenolics in extracts was determined by Folin – Ciocateu method as modified by **Singleton *et al*. (I999)**.

6- Determination of total flavonoids content

The flavonoid content is expressed as milligrams of rutin equivalents per gram of sample (mg RE/g) according to the method of **(Rice-Evans *et al*., 1996)**.

Statistical analysis

All the obtained data were subjected to statistical analysis and compared according to the least significant difference (LSD) as mentioned by **Snedecor and Cochran (1990)**.

3. Results

Morphological Identification

Root-knot nematodes (*Meloidogyne* spp.) have been identified using morphological and morphometric characters on second-stage (J2) juveniles, males and perineal patterns of mature females **(Hunt and Handoo, 2009)**. These body traits include perineal style shape, style length, body width, body length, head shape for females and males, juveniles in the second stage **(Hunt and Handoo, 2009)** and differential diagnosis **(Perry *et al*., 2009)**.

Effect of different organic matters on vegetative growth and flowering of Williams banana under field conditions during 2020/21 and 2021/22 growing seasons

Disease control

The use of organic matter to banana plants as soil amendments (Tables, 1 and 2) resulted in a better soil quality and greater plant disease suppressiveness. Data in tables (4a and b) show that the efficacy of organic matters, *i.e*., plant residue, cow manure and compost "plant residue + cow manure", *Azotobacter chroococcum*, vermicompost or humic acid separately reduced the numbers of nematodes in the soil and the roots of banana plants. Data in Tables (4a and b) illustrate that the quantity of galls, developmental stages, females, and egg-laying females in banana roots was significantly reduced with application of all treatments involving different organic compounds and biofertilizer during the two growing seasons 2020/21 and 2021/22. The number of young nematodes in the soil, the overall nematode population, and the rate of nematode build-up were all found to be similar to those of the control check.

Additionally, as compared to the control as well as other treatments, the number of nematodes in the soil (J2s) in the compost treatments suppressed the other investigated compounds. While nematode development stages were dramatically reduced in treatments with plant residue when compared to other treatments. As a result, the use of tested materials had a significant impact on the nematode population. For example, compost treatment outperformed all other studied materials in terms of nematode population reduction.

Table (4 a). Effect of different organic matter and biofertilizer on banana root knot nematode under field condition during 2020/21 growing season

Treatments	Time of application	No. of galls/root	No. of Juveniles in soil	Nematodes in root			Total population
				Developmental stages	females	Egg-masses	
Azotobacter chroococcum	7 April	27.50	3968.75	27.00	45.00	23.75	4064.50
	21 April	16.50	2381.25	16.20	27.00	14.25	2438.70
	5 May	5.50	793.75	5.40	9.00	4.75	812.90
Plant residue	7 April	32.50	4156.25	25.50	41.50	25.75	4249.00
	21 April	19.50	2493.75	15.30	24.90	15.45	2549.40
	5 May	6.50	831.25	5.10	8.30	5.15	849.80
Cow manure	7 April	30.00	2479.50	26.50	39.50	33.00	2578.50
	21 April	18.00	1487.70	15.90	23.70	19.8	1547.10
	5 May	6.00	495.90	5.30	7.90	6.60	515.70
Compost	7 April	26.75	2274.00	21.71	41.50	22.50	2359.71
	21 April	16.05	1364.40	13.03	24.90	13.50	1415.83
	5 May	5.35	454.80	4.34	8.30	4.50	471.94
Humic acid	7 April	51.50	3285.25	66.00	48.75	39.75	3439.75
	21 April	30.90	1971.15	39.60	29.25	23.85	2063.85
	5 May	10.30	657.05	13.20	9.75	7.95	687.95
Vermicompost	7 April	28.50	3969.75	28.00	46.00	24.75	4068.50
	21 April	17.10	2381.85	16.80	27.60	14.85	2441.10
	5 May	5.70	793.95	5.60	9.20	4.95	813.70
Control	Untreated	114.0	5740.00	41.00	102.50	79.00	5962.5
LSD at 5%		1.22	4.22	1.20	0.78	1.18	4.33

Table (4 b). Effect of different organic matter and biofertilizer on banana root knot nematode under field condition during 2021/22growing season

Treatments	Time of application	No. of galls/root	No. of Juveniles in soil	Nematodes in root			Total population
				Developmental stages	females	Egg-masses	
Azotobacter chroococcum	7 April	28.50	3969.75	28.00	46.00	24.75	4068.50
	21 April	17.10	2381.85	16.80	27.60	14.85	2441.10
	5 May	5.70	793.95	5.60	9.20	4.95	813.70
Plant residue	7 April	33.50	4157.25	26.50	42.50	26.75	4253.00
	21 April	20.10	2494.35	15.90	25.50	16.05	2551.80
	5 May	6.70	831.45	5.30	8.50	5.35	850.60
Cow manure	7 April	31.00	2480.50	27.50	40.50	34.00	2582.50
	21 April	18.60	1488.30	16.50	24.30	20.40	1549.50
	5 May	6.20	496.10	5.50	8.10	6.80	516.50
Compost	7 April	27.75	2275.00	22.71	42.50	23.50	2363.71
	21 April	16.65	1365.00	13.63	25.50	14.10	1418.23
	5 May	5.55	455.00	4.54	8.50	4.70	472.74
Humic acid	7 April	52.50	3286.25	67.00	49.75	40.75	3443.75
	21 April	31.50	1971.75	40.20	29.85	24.45	2066.25
	5 May	10.50	657.25	13.40	9.95	8.15	688.75
Vermicompost	7 April	29.50	3970.75	29.00	47.00	25.75	4072.50
	21 April	17.70	2382.45	17.40	28.20	15.45	2443.50
	5 May	5.90	794.15	5.80	9.40	5.15	814.50
Control	Untreated	115.00	5742.00	43.00	103.00	80.00	5968.00
LSD at 5%		1.24	4.28	1.30	0.88	1.20	4.34

Vegetative growth and Flowering

Data in Table (5-a & b) reveal that all the added organic matters, i.e., plant residue, cow manure and compost "plant residue + cow manure" to the soil of banana plants at the rate of 1.2 Kg/m^2 and transplants treatment with diluted of 3:200 as a recommended dose of *Azotobacter chroococcum*, vermicompost or humic acid separately significantly increased vegetative growth and the best of earlier flowering i.e. pseudostem height, and girth (cm), leaf area (cm), No. of green leaves at bunch shooting and period to bunch shooting (days) in Williams banana plants in comparison with control treatment during 2020/21 and 2021/22 growing seasons. Compost was the most effective one followed by *A. chroococcum* then vermicompost in both growing seasons, respectively compared with control treatment. On the other hand, humic acid was the least effective one compared with other treatments rather than control treatment.

A yield measurement

The information obtained in Table (6) demonstrates that throughout the 2020/21 and 2021/22 growth seasons, the effectiveness of different types of organic matter and biofertilizer has an important influence on the number of fingers and hands, as well as the bunch and hand weights.

Resulted data in Table (6) reveal that using any of organic matters or biofertilizer to banana plants led to significant increase in the estimated yield parameters in two growing seasons 2020/21 and 2021/22.

Table (5 a). Effect of adding different organic matters and biofertilizer on vegetative growth and flowering of Williams banana plants under field conditions during 2020/21 growing season

Different treatments	Pseudostem height (cm)	Pseudostem girth (cm)	Leaf area (m2)	No. of green leaves at bunch shooting	Period to bunch shooting (days)
Azotobacter chroococcum	265.4	80.40	2.36	14.10	411
Plant residue	260.7	77.20	2.28	13.00	425
Cow manure	264.6	78.30	2.30	13.50	420
Compost	266.9	81.50	2.38	14.20	402
Humic acid	258.2	74.60	2.25	12.60	428
Vermicompost	262.3	79.70	2.34	13.80	418
Control	247.6	69.10	1.70	10.70	431
LSD at 5%	2.33	1.14	0.12	0.66	2.10

Table (5 b). Effect of adding different organic matters and biofertilizer on vegetative growth and flowering of Williams banana plants under field conditions during 2021/22 growing season

Different treatments	Pseudostem height (cm)	Pseudostem girth (cm)	Leaf area (m2)	No. of green leaves at bunch shooting	Period to bunch shooting (days)
Azotobacter chroococcum	266.4	80.50	2.37	14.20	412
Plant residue	261.7	77.30	2.29	13.10	426
Cow manure	265.6	78.40	2.32	13.60	421
Compost	267.9	81.60	2.39	14.30	404
Humic acid	259.2	74.70	2.26	12.70	429
Vermicompost	263.3	79.80	2.35	13.90	420
Control	248.6	69.20	1.71	10.72	432
LSD at 5%	2.44	1.16	0.10	0.70	2.20

Table (6). Effect of adding different organic matters and biofertilizer on yield and fruit quality of Williams banana plants under field conditions during 2020/21 and 2021/22 growing seasons

Treatments	2020/21 growing season				2021/22 growing season			
	No. of fingers / hand	No. of hands / bunch	Hand weight (kg)	Bunch weight ⁄ plant (kg)	No. of fingers / hand	No. of hands / bunch	Hand weight (kg)	Bunch weight ⁄ plant (kg)
Azotobacter chroococcum	16	9	2.87	25.83	17	9	2.88	25.92
Plant residue	13	7	2.68	18.76	14	7	2.69	18.83
Cow manure	14	8	2.78	22.24	15	8	2.88	23.04
Compost	17	10	2.93	29.30	18	10	2.94	29.40
Humic acid	13	6	2.63	15.78	14	7	2.65	54.54
Vermicompost	15	8	2.83	22.64	16	8	2.84	22.72
Control	11	5	2.18	10.09	12	6	2.19	13.14
LSD at 5%	0.45	0.25	0.10	1.14	0.46	0.27	0.11	1.16

Compost which consists of cow manure and plant residue showed the best results and caused significant increase in number of fingers/hand, number of hands/bunch and the averages of hand weight (g) and bunch weight/plant (Kg), respectively, being (17, 10, 2.93 g and 29.30 Kg) in 2020/21 growing season and (18, 10, 2.94 g and 29.40 Kg) in 2020/2021 growing season, respectively compared with untreated plants. On the contrary, humic acid was the least effective one during the two growing seasons compared with other treatments.

Total soluble solid (TSS), Flavonoids and total phenols as chemical components determination

Data in Table (7) indicate that all tested organic matter treatments at the rate of 1.2 Kg/ m^2 to the soil of banana plants affected positively the activities of total soluble solid (TSS), flavonoids and total phenols in leaves of banana plants in comparison with control treatment during the two successive seasons (2020/21 and 2021/22).

Table (7). Effect of adding different organic matters and biofertilizer on the activity of TSS, flavonoids and total phenols of Williams banana plants under field conditions during 2020/21 and 2021/22 growing seasons

Different treatments	2020/21 growing season			2021/22 growing season		
	TSS	Flavonoids	Total phenols (mg/100g FW)	TSS	Flavonoids	Total phenols (mg/100g FW)
Azotobacter chroococcum	21.4	45.60	95.1	21.7	46.20	95.3
Plant residue	19.7	38.90	93.5	19.9	39.80	94.0
Cow manure	20.1	42.70	94.2	20.3	43.70	94.5
Compost	21.5	47.50	95.5	21.9	48.30	95.7
Humic acid	19.2	35.40	91.9	19.4	35.90	92.1
Vermicompost	20.6	44.80	94.7	20.8	45.50	94.9
Control	17.9	29.00	83.8	18.1	30.10	84.2
LSD at 5%	1.21	2.11	2.22	1.22	2.12	2.23

In this respect, the highest effective treatment on total soluble solid (TSS), flavonoids and total phenols was compost treatment, where it recorded 21.5, 47.50 and 95.5 % during 2020/21 growing season and 21.9, 48.30 and 95.7 % during 2021/2022, respectively, followed by *Azotobacter chroococcum*. On the other hand, humic acid showed the lowest effect in comparison with the other treatments.

Effect of applying different organic matters and biofertilizer to the soil of banana plants on the fruit quality under field conditions

Data in Table (8) illustrate the role of different organic matters or biofertilizer on the nutrition of banana plants. Results revealed that there were changes occurred in fruit quality Total sugar (g/100g FW), Vitamin C (mg/100g FW) and protein (g/100g FW) due to these treatments in comparison with control treatment during the two seasons (2020/21 and 2021/22). Presented data in Table (8) indicate that extensive compost treatment was the highest effective one concerning Total sugar being 17.6 and 17.7, Vitamin C, being 5.3 and 5.4 mg/100g FW, protein being 3.11 and 3.12 g/100g FW during both seasons, respectively followed by *Azotobacter chroococcum* then vermicomposting. On the opposite trend, humic acid showed the least effective treatment compared to control treatment. Finally, all organic matters improved the fruit quality and the results in the 2021/22 growing season were higher than in 2020/21 growing season.

Table (8). Effect of adding different organic matters and biofertilizer on fruit quality of Williams banana plants under field conditions during 2020/21 and 2021/22 growing seasons

Different treatments	2020/21 growing season			2021/22 growing season		
	Total sugar (g/100g FW	Vitamin C (mg/100g FW)	Protein (g/100g FW)	Total sugar (g/100g FW	Vitamin C (mg/100g FW)	Protein (g/100g FW)
Azotobacter chroococcum	17.1	5.1	3.01	17.3	5.2	3.09
Plant residue	15.9	4.3	2.73	16.1	4.5	2.75
Cow manure	16.3	4.6	2.85	16.5	4.8	2.87
Compost	17.6	5.3	3.11	17.7	5.4	3.12
Humic acid	15.3	3.8	2.59	15.5	4.0	2.63
Vermicompost	16.8	4.8	2.97	16.9	5.0	2.99
Control	11.6	3.1	0.85	11.8	3.3	0.88
LSD at 5%	1.12	0.20	0.12	1.13	0.22	0.13

4. Discussion

Recently, human realized that using many chemical pesticides might have injury on the environment and human health because they highly toxic substances in agricultural, which led to great disturbance in the biological balance. This disturbance led to the appearance of new pests, caused reduction in number of natural enemies and increased the accumulated toxic chemicals in human food chain. New group of food products appeared in the markets under different names, i.e. organic biodynamic and ecological food. All these names showed that no synthetic chemicals were added during production or processing.

The present work was designed to reduce using toxic chemicals in agriculture process to produce food of high quality in sufficient quantity, enhance biodiversity system, maintain and increase long-term fertility of soils. In addition to, find out the most suitable non-chemical methods to protect banana plants against nematodes diseases.

Root-knot nematodes (*Meloidogyne spp*.) have been identified using morphological and morphometric characters on second-stage (J2) juveniles, males and perineal patterns of mature females **(Hunt and Handoo, 2009)**. These body traits include perineal style shape, style length, body width, body length, head shape for females and males, juveniles in the second stage **(Hunt and Handoo, 2009)** and differential diagnosis **(Perry *et al*., 2009)**.

The morphological details of these species are important for identifying and defining evolutionary relationships **(Karssen and Moens, 2006)**. In addition, these morphological details are often used to determine physiological function **(Shepherd and Clark, 1983)**.

Improvement of banana plants growth by the application of these biological products appear to undergo physiological changes that render them unsuitable for nematode penetration and development, thus inducing a certain degree of resistance in plants against nematode infestation soil **(Mahfouz, 2011)**. The beneficial effects of organic amendments for improving the physical, chemical and biological properties of soil are well recognized **(Mayad *et al*., 2013)**.

It has been suggested that the depressive effect of some amendments on fungi and nematodes may be due to highly specific fungistatic and nematicidal substances released during their decomposition or due to nitrogen starvation resulting from greater capacity of saprophytes to multiply or greater sensitiveness of parasitic fungi to liberation of CO_2 during decomposition. Use of organic amendments also leads to the increase of natural enemies' level in soil **(Chavarria-Carvajal & Rodriguez-Kabana, 1998)**.

The volatile fatty acids like formic, acetic, propionic and butyric acids, ammonia, formaldehyde, hydrogen sulphide, phenols and amino acids that are released during the decomposition of amendment, have been reported to be toxic to nematodes **(Alam, 1993)**.

The suppression of root knot nematodes by organic amendments used in this study is probably based on a complex mode of action involving multiple mechanisms. Changes in soil enzyme activities cause a shift in specific groups of microorganisms after the application of organic amendments.

The use of organic matter to tomato plants as soil amendments (Tables, 1 and 2) resulted in a better soil quality and greater plant disease suppressiveness **(Ahmed,2013)**; however, in this study it depended on the type of organic fertilizer. Applications of added organic matters, *i.e*., plant residue, cow manure and compost "plant residue + cow manure" to the soil of banana plants at the rate of 1.2 Kg/m^2 and transplants treatment with diluted of 3:200 as a recommended dose of *Azotobacter chroococcum*, vermicompost or humic acid treatments into the soil suppressed the population of *Meloidogyne incogonita* on banana and increased plant growth. All the tested products reduced significantly the number of J2 and root damages.

The lowest nematode population density was observed in treated plant with compost (**Mayad *et al*., 2013**). The results support earlier reports of decreased infestations of root knot nematode following compost products application in different crops (**Ferji *et al*., 2004a, and 2004b)** and direct effect of chemical compounds of compost on J2 larvae of *Meloidogyne spp*. (**Mayad *et al*., 2013)**.

The added organic matters, i.e., plant residue, cow manure and compost "plant residue + cow manure" to the soil of banana plants at the rate of 1.2 Kg/m^2 and transplants treatment with diluted of 3:200 as a recommended dose of *Azotobacter chroococcum*, vermicompost or humic acid separately significantly increased vegetative growth and the best of earlier flowering *i.e.* pseudostem height, and girth (cm), leaf area (cm), No. of green leaves at bunch shooting and period to bunch shooting (days) in Williams banana plants during 2020/21 and 2021/22 growing seasons. The effective occur was expressed both by reducing the number of galls on the root infected plants and the final nematode number in soil. In the present study, the treatments were applied to control root-knot nematode, *M. incognita* on banana plants; they exhibited antagonistic action and most of them affected on total nematode numbers **(Mayad *et al*., 2013)**. The majority of tested materials have been known to possess nematicidal properties that may be release in soil. Likewise, organic matter and biofertilizers produce a great variety of secondary metabolites **(Gerwick *et al*., 2001)**, such as nitrogen-containing compounds, polyketides, lipopeptides, cyclic peptides and many others.

Animal dung is rich in nitrogen, phosphorus, and potassium, which the nematode-destroying fungi use saprophytically as they grow, according to **Swe *et al*. (2011)**. As an illustration, consider the species of *Arthrobtrys* spp., which are known to flourish in soil both as saprophytes and predators. These findings are consistent with previous research by **Jaffe (2004), Farrell *et al*. (2006), Xue *et al*. (2011)**, and, which reported that *A. oligospora* gains its carbon and energy from organic matter in its capacity as a saprophyte and from ensnaring nematodes as a parasite, allowing it to adapt to a wide variety of habitats **(Wachira *et al*. (2009b)**.

Compost fertilization is the limiting factor for banana plant's growth and productivity. Bananas owing to its large size and rapid growth rate requires relatively high amounts of compost to get high yield with good fruit quality **(Nijjar, 1985)**. Pollution is one of the most negatively problems that affecting on human health, especially when the edible part of the plant is contaminated with any pollutants. Using mineral-N fertilizers causes accumulation of harmful residual substances in the pulp of the fruits. In this connection, similar report given by **Ahmed, 2013 and Ahmed, *et al*., 2017** who came to the same conclusion. Some investigators dealt with the effect of organic manure on vegetative growth, yield and chemical constituents.

They stated that application of organic manure increased dry weight/plant; N, P and K contents and the yield measures **(Athani *et al*., 2009, Bhutani *et al*., 2012, Gaikwad *et al*., 2010 and Suhasini *et al*., 2018)** on banana plants in comparison with control treatment. The organic fertilization has a positive action in increasing activity of microflora, water holding capacity, soil structure aggregation, soil organic matter, soil humus content and the availability of most nutrients. Such stimulation on the uptake of nutrients leads to improving the biosynthesis of organic foods and cell division **(Miller *et al*., 1990)**.

Bio-fertilization has an important role on biological, as well as, activating the availability and incidence of soil borne diseases, facilitating the fixation of atmospheric N, and then improving soil fertility **(Kannaiyan, 2002; El-Salhy, 2004 and El-Salhy *et al*., 2019)**.

The best results regarding to growth, fruit quality and yield due to use compost were alternated to positive action of compost on enhancing soil fertility, the availability of nutrient, organic matter, root development, activity of organisms and N-fixation **(Formowit *et al*., 2007)**.

Continuous application of organic and bio-fertilizers is promising in the long run of banana, as sources of organic matter, essential nutrients, amino acids, natural hormones, antibiotics and vitamins. Also, improving both physical and chemical characters of soil.

Accordingly, it can be concluded that the fertilization using bio and organic fertilizers affect positively on improving the plant vigor expressed as an increase in leaf surface expansion and its nutrient status. These findings emphasize the vital importance of these fertilization sources in order to overcome the losses of nutrients by leaching, volatilization and mobility of movement elements. These sources also, improve the soil fertility due to the highest values of the residual nutrients, the enhanced solubility of nutrients and the increased activity of microorganisms. In addition, the importance of such fertilization treatments is considered for the organic farming production.

The results were consistent with what had been achieved by **Bhalerao *et al*. (2009), Roshdy (2010), Badgujar *et al*. (2010), Barakat *et al*. (2011), Kuttimani *et al*. (2013), Abdel-Rahman and Mansour (2015), Baiea and El-Gioushy (2015) and Abdel-Hafiz *et al*. (2016)**. They concluded that application in either organic or bio-form along mineral-N, P and K sources was effective on improving growth vigor and nutrient status of banana plants in favor of improving the fruiting.

In this respect, the highest effective treatment on total soluble solid (TSS), flavonoids and total phenols was compost treatment, followed by *Azotobacter chroococcum*. On the other hand, humic acid showed the lowest effect in comparison with the other treatments. These results are in harmony with those reported by **Ahmed (2013)** who came to the same conclusion when tomato plants were dealt with the effect of organic manure on vegetative growth, yield and chemical constituents (Tables, 1 and 2) it can improve soil physical and chemical properties and nutrient dynamics **(Ahmed, *et al*., 2017, Ahmed, *et al*., 2018 and Ahmed *et al*., 2022)**.

Data illustrate the role of different organic matters or biofertilizer on the nutrition of banana plants. Results revealed that there were changes occurred in fruit quality Total sugar (g/100g FW), Vitamin C (mg/100g FW) and protein (g/100g FW) due to these treatments in comparison with control treatment during the two seasons (2020/21 and 2021/22). Presented data indicate that extensive compost treatment was the highest effective one followed by *Azotobacter chroococcum* then vermicomposting.

On the opposite trend, humic acid showed the least effective treatment compared to control treatment. Finally, all organic matters improved the fruit quality and the results in the 2021/22 growing season were higher than in 2020/21 growing season. The obtained results are in harmony with those obtained by **(Gaikwad *et al*., 2010; Ahmed *et al*., 2017; Ahmed *et al*., 2022)**, who explained That instead of the increment in physical and chemical quality attributes may be due to the increase of macro nutrient content of plant tissues which affect photosynthetic assimilation rate and in turn increased accumulation of total soluble solids in fruit and ascorbic acid, total acidity and carbohydrate, which was intermediate products during photosynthetic assimilation process. It is also contributed to increase the aeration capacity of soil, which improving and increasing the quality, quantity, dry matter, vitamin C and sugars of vegetable crops as improved by **Abdel-Rahman and Mansour (2015); Ahmed, *et al*. (2017)**.

5. Conclusion

The purpose of this research is to find alternatives to chemical materials, such as adding different organic materials, such as organic fertilizer, cow dung, and plant residues, to the organic farm of banana plants. Williams banana at 1.2 kg/m^2 or soaked at 3:200/fed as the recommended dose of vermicompost, *Azotobacter chroococcum*, and humic acid in response to the significant decrease in the number of nematodes in the soil and roots of banana plants during the growing seasons 2020/21 and 2021/22, as well as to increase soil fertility and preserve biological diversity, thereby increasing banana fruit productivity and quality. Compost, a banana-rich organic matter at 1.2 kg/m^2, resulted in the greatest reduction in root-knot nematode infestation, as well as the greatest increase in vegetative growth, pseudostem height and girth (cm), and leaf area. The quantity of green leaves when nodules are released, as well as the time period till veins are released (days); Yield Parameters Fruit quality indicators include total soluble solids (TSS), vitamin C, protein, total sugar, flavonoid chemical components, and total phenolics.

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