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Research Article Impact of Different Agro-Wastes on Quality of Vermicompost Made of Either Fish Sludge or Cow Dung

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Abstract

Background and Objective: Vermicompost technology application is attracting great attention these days for its multi-functions whether considered safely and friendly mechanism for agri-aguacultural wastes disposal, soil conditioner, decreasing soil degradation as a consequences of excessive agriculture activities. This study tested the theory that built on "for how far can manipulate the product quality according to the input (raw materials)". This study aimed to observe the impact of Different Agro-Wastes on Quality of Vermicompost. Materials and Methods: Different agricultural wastes (Rice Straw (RS), sugarcane (SC) and Banana Leaves (BL) were added during vermicomposting of Cow Dung (CD) and Fish Sludge (FS) by ratio (2 fish sludge or cow dung: individually). After entire degradation of wastes, samples were analyzed to determine vermicompost quality. Also, Colony Forming Units (CFU) and antimicrobial activity were evaluated. Results: Results indicated that, FS vermicompost types surpassed those from CD in dry weight and ash contents. Meanwhile CD vermicompost types surpassed those from FS in Organic matter and humidity content. Moreover, adding agro-wastes (RS, SC and BL) enhanced nutrient status in vermicompost whether produced from FS or CD. Generally, adding banana leaves wastes during vermicomposting enhanced K content in the product, while adding RS enhanced N content. Increasing nutrient levels in produced vermicompost than those in initial material may be related to activity of earthworm and microbial during degradation of organic wastes and increasing mineralization rate. In regard to amino acids and plant growth promoters content, adding agro-wastes had a positive effect. Besides, there is evidence that studied vermicompost types varied in its antagonistic to pathogenic microbes. Conclusion: There is a great potential to produce vermicompost with specific quality as growers needs or requirements of plant stage via using different agro wastes.

Key words: Vermicompost, cow dung, fish sludge, agro-wastes, rice straw, sugar cane, banana leaves, microbial activity, antimicrobial, nutrient contents, growth promoters and amino acids content

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

In Egypt a huge amount of agricultural wastes produced 30-35 million tons annually¹. Conversion these agro-wastes into compost by means of turned piles has resulted in a final product with poor physical and chemical characteristics. As the aquaculture solid-wastes cannot be applied directly in the crops land as an organic fertilizer hence it may cause several problems: offensive odor, oxygen depletion in the plant root zones resulting in plants die and if it dried it forms a solid crust on the soil surface that causes a seed germination failure as confirmed by Dauda *et al.*² and the potentiality of transporting pathogen as reported by Birch *et al.*³.

Vermicomposting technology utilizing earthworms, most frequently from the genus *Eisenia fetida* is reported by Kováčik *et al.*⁴ as multilateral natural bioreactors, which plays an essential role in decomposing of organic matter and agro-wastes, improving soil fertility, representing an efficient natural recycling and enhanced plants' growth^{5,6}.

During vermicomposting, the important plant nutrients such as (i.e., N, P, K, Ca and MG) present in the feed material are converted through microbial action into forms that are much more soluble and available to the plants than those in the original substrate⁷⁻⁹. Also, many authors reported that vermicomposts are very important in crop production as they contain biologically active substances such as plant growth regulators according to Alphonsine and Blok and Margit^{10,11}.

Besides, vermicomposting is a biological process, in which microorganisms play a key role in the evolution of organic material and in its transformation from waste to safe organic amendments or vermicompost. The effects that earthworms have on the microorganisms must be established because if earthworms stimulate or depress micro biota or modify the structure and activity of microbial communities, they will have different effects on the decomposition of organic matter, surface area and its guality. Knowledge of the microbial structure and functions in mature vermicompost is important to predict its potential impact on soil fertility according to other studies¹²⁻¹⁴. These activities may enhance OM turnover rate and productivity of microbial communities, thereby increasing the rate of decomposition Gómez-Brandón et al.¹⁵. Moreover, Edwards and Lofty¹⁶ showed that earthworms are avid to organic waste and using only a small portion (5-10%) on the organic waste for their body building, they excrete a large part of these consumed waste in a half digested form as vermicast. Since the intestine of earthworms harbor wide ranges of microorganisms, enzymes, hormones, etc., these half-digested substrates decompose rapidly and transform into a form of vermicompost within a short time. In addition,

this vermicast is mixed with mucus secretion of the earthworm's gut wall. Vermicomposting process is increasing polymerization of humic substances in parallel with a decrease of ammonium N and rising of nitric N^{17} .

Many studies emphasized on efficiency of vermicompost as organic fertilizers for several reasons, whereas Tajbakhsh *et al.*, Mustafa *et al.*, Moustafa *et al.*^{5,18,19}, indicated that vermicompost is considered as an excellent product since it has desirable esthetics, plant growth hormones, higher level of enzymes, greater microbial population and tend to hold more nutrients over a longer period without adversely impacting the environment. The plant growth regulators and other plant growth influencing materials, that include auxins, cytokinins, humic substances, etc. are produced by microorganisms in vermicompost as has been reported by Atiyeh *et al.*, Muscolo *et al.*^{20,21}.

In addition, Sartaj *et al.*²², showed that mixing cow dung with plant wastes such as sugar cane wastes helping to improve their acceptability by *E. fetida* and improved physico-chemical characteristics of produced vermicompost. In this context, this study came to illustrate the importance of mixing agro wastes with raw material (cow dung for fish sludge) of vermicompost to improve its quality. In view of aforementioned facts above, the present work was conducted to assess the potential of producing vermicompost with different levels of nutrients based on different nutrient requirements for plants.

MATERIALS AND METHODS

Study area: This work was carried out under the supervision of National Research Center, where vermicompost types were produced at the Central Laboratory for Aquaculture Research (CLAR) during (2019-2020) and all needed analysis were done at the National Research Centre (NRC), except microbiological analysis was done at the Faculty of Science, Tanta University.

Specimen collection: Mixing three species of earthworm (*Eisenia fetida, Lumbricus rubellus* and *Perionyx excavatus*) were raised on cow dung and fish sludge until utilized in the experiment Preparation of different feeding materials:

 Cow Dung (CD) processing: Fresh cow dung was obtained from a cow farmer adjacent to the Central Lab for Aquaculture Research (CLAR) and applied directly to the treatments, assuming the moisture is about 50% of the wet weight

- Fish sludge collection and preparation: Fish Sludge (FS) was collected from the concrete ponds of Nile tilapia *Oreochromis niloticus* brood stock and fry, at the Nile tilapia hatchery belonging to CLAR, during fry harvesting from the broodstock ponds as well as from fry rearing ponds. The produced FS, with a moisture content of 96.5% and dry solid content of 3.5%, was collected in barrels and then spread out in a thin layer on a cement floor for drying over fourteen days, so it can be stored safely until being used
- Banana Leaves (BL) processing: Banana leaves were collected from CLAR nearby farm, then sun-dried for 3 days and were crushed into small pieces
- **Rice Straw (RS):** Rice straw was bought from CLAR nearby farm after harvesting a rice crop. Rice straw was chopped into small pieces, less than 2 cm length
- Sugar cane (SC): Sugar cane wastes were collected from sugar cane juice's shop and sun-dried for 3 days then minced into small pieces by machine

Earthworm inoculation and vermicompost production: Both

of fresh cow dung and dried fish sludge were mixed individually with agro-wastes (RS, SC or BL) at a ratio of 2:3, respectively and moistened to 60-70% in Styrofoam boxes with dimensions of $60 \times 40 \times 30$ cm. After 24 hrs, three species of earthworm (Eisenia fetida, Perionyx excavatus and Lumbricus rubellus) were added to the media at a rate of 50 g worm per 1000 g media. For eight weeks the boxes were checked weekly and re-moistened and mixed until the vermicompost matured. All boxes were kept indoors and the temperature was maintained between 18-25°C during the vermicompost maturation. At harvest time, vermicompost was checked manually on white plastic surface and the adult as well as pre-adult earthworms were collected then the vermicompost was returned to the boxes again for one more month. Later, the vermicompost was re-checked again and all hatched earthworms were collected. The harvested vermicompost was packed in plastic bags and delivered to laboratories to be analyzed.

Experimental treatments: Eight setup (treatments) containing different feeding materials (Cow Dung (CD) alone, Fish Sludge alone (FS) and CD or FS supplemented with BL, RS and SC respectively) with three replicates of each were prepared as following:

CD, FS, CD+BL, CD+RS, CD+SC, FS+BL, FS+RS and FS+SC

Samples of all these treatments delivered to different laboratories in order to be analyzed.

Analysis of chemical and microbial parameters:

- Dry weight (g)
- Organic matter (%)
- Humidity (%)
- Ash
- C/N ratio
- Nitrogen % (Kjeldahl method)
- Available Phosphorus (Modified Olsen's method)
- Available Potassium (Ammonium acetate method)
- Amino acids content (mg/100g dry weight)
- Growth promoters content (g/100g sample)
- Bacterial and Fungal population (Serial dilution and plate count method)

Physiochemical analysis for vermicompost: Vermicompost samples were dried in a ventilated oven at 70°C to constant weight. Samples were grinded in stainless steel mill with 0.5 mm sieve and kept in plastic containers for chemical analysis. The samples (1 g of each sample) were dry-ashed in a muffle furnace at 450°C for 6 hrs.

Macronutrients were extracted using the dry ashing digestion method according to Chapman and Pratt²³. Nitrogen was determined by using the Kjeldahl method, the ash was dissolved in HCI (2N) and phosphorus was photometrical determined in the digested solution using vanado-molybdate color reaction according to the method described by Jackson²⁴. Potassium was measured in the digested suspension using the Flame photometer, (Eppendorf, DR Lang). Organic matter content was determined according to Walkely and Black²⁵.

Free amino acids and growth promoter's analysis: To determine total free amino acids, the modified of ninhydrin colorimetric method that described by Rosein, Selim *et al.*^{26,27}, was used for this purpose. Besides, growth promoters in samples of vermicompost were estimated according to method described by Dobrev *et al.*²⁸.

Microbiological analysis of vermicompost samples

Sampling and sample preparation: Five grams from vermicompost samples were placed in sterile Stomacher bags and treated by a Stomacher 400 Circulator for 60 sec at middle speed after adding 45 mL sterile 0.85% NaCl. The Stomacher blending step was repeated three times and the microbial suspension was obtained.

Estimation the counts of total viable bacteria count: Tenfold serial dilution of the microbial suspensions obtained with the

protocol described above made with sterile 0.85% NaCl were plated onto plate count agar medium for the estimation of total viable counts, counts of Colony Forming Units (CFU) were estimated after 3 days of incubation at 28°C and were calculated per gram vermicompost. The total resistant bacteria were estimated by planting the same dilution onto plate count agar medium sublimated with (20 mg L⁻¹) for (penicillin, ampicillin, erythromycin and tetracycline, respectively).

Antimicrobial activity assay: This method was done by agar well diffusion test according to Schillinger and Luck²⁹. To determine the antimicrobial activity of the vermicompost samples against the selected identified pathogenic bacteria e.g., *Citrobacter freundii, Enterobacter cloacae, Pseudomonas aeruginosa* and *Klebsiella pneumonia*. The prepoured nutrient agar plates were over layed with 100 µL of overnight culture of tested pathogens (in nutrient broth), then spread well with L-shaped glass rod. After 15 min, wells of 5 mm diameter were made with a sterile cork borer. Samples of vermicomposts extract were placed into wells. Plates were then incubated at 30°C for 12 hrs. The inhibition zones were measured to assay the antimicrobial activity of vermicompost samples.

Data analysis: Vermicompost samples were analyzed using the standard procedures in the laboratory at National research Centre and Faculty of Science, Zoology department, Tanta University. All data are the means of triplicates. Statistical analysis of data, analysis of variance (ANOVA) and mean separation were carried out using Duncan's multiple range test and significance were determined at the (p≤0.01) level(Duncan)³⁰. Data analysis was performed using ASSISTAT version 7.7 beta (2015).

RESULTS AND DISCUSSION

Physio-chemical parameters: Several physiochemical data were represented in Table 1. Dry weight for all FS

vermicompost types was significantly higher than CD vermicompost types. The significantly highest dry weight was recorded at the treatment of (FS+BL), while the significantly lowest dry weight was at the treatment of (CD+BL). For organic matter (O.M), it was noticed that CD vermicompost types had higher O.M than FS vermicompost types and vermicompost of CD alone without plant wastes additives recorded the highest O.M comparing with other treatments. While the significantly lowest value of O.M was determined at the treatment of FS only. For humidity, CD vermicompost types contained higher humidity comparing with FS vermicompost types. Adding BL wastes to CD yielded the markedly highest humidity. For ash content, the trend was different whereas the FS vermicompost types recorded higher ash content comparing with DC vermicompost types. Adding different agro-wastes (RS, SC or BL) to CD increased ash content on contrary to the decreased ash content in FS treatments. With respect to the C: N ration, the significantly highest ratio was calculated in CD alone treatment, this may be attributed to its higher O.M content. Meanwhile, the lowest ratio was calculated in CD+BL as in FS+SC.

In respect of nutrients status, generally it can be observed that adding agro-wastes to raw material of vermicompost enhanced nutrient content whereas these plant wastes improved level of N and K. For nitrogen, most considerable values were recorded with vermicompost types produced from CD supplemented with agro-wastes and the highest value (1.89%) was noticed in (CD+RS) followed with (CD+SC) and (CD+BL) with no markedly differences among these types. Meanwhile, the lowest N content was observed in treatment (FS). However vermicompost produced from (FS+SC) didn't differed markedly than CD vermicompost types in nitrogen content. For phosphorus, generally, adding the plant wastes resulted in a decrease in the P content of the produced vermicompost. Except for the adding BL to FS, this resulted in the highest P content that may be attributed to the higher P content in the FS than that of CD.

Table 1: Impact of different agricultural wastes on physiochemical parameters of vermicompost

	Initial weight (g)	Dry weight (g)	O.M (%)	Humidity (%)	Ash	C/N ratio	N (%)	P (%)	K (%)
CD Control	10	6.70 ^b	46.60ª	33.0 ^B	20.40 ^d	20.40ª	1.33 ^{ab}	0.35 ^{ab}	1.08 ^d
CD+RS	10	6.72 ^b	37.87 ^b	32.8 ^B	29.33 ^{cb}	11.51 ^b	1.89ª	0.13 ^{bcd}	1.50 ^{cd}
CD+SC	10	6.78 ^b	33.68 ^{cb}	32.2 ^B	34.12 ^{bcd}	11.52 ^b	1.68 ^{ab}	0.08 ^{cd}	1.40 ^{cd}
CD+BL	10	6.38°	33.62 ^{cb}	36.2 ^A	30.18 ^{cd}	11.50 ^b	1.68 ^{ab}	0.01 ^d	1.98ªb
FS control	10	7.27ª	21.06 ^c	27.3 ^c	54.94ª	11.58 ^b	1.09 ^b	0.40ª	1.88 ^{ab}
FS+RS	10	7.48ª	23.20 ^c	25.2 [⊂]	51.63ªb	11.63 ^b	1.16 ^b	0.29 ^{abc}	2.13 ^{ab}
FS+SC	10	7.45ª	26.48 ^{cb}	25.5 ^c	48.02 ^{abc}	11.50 ^b	1.34 ^{ab}	0.32 ^{ab}	2.13 ^{ab}
FS+BL	10	7.50ª	23.13 ^c	25.0 ^c	51.87ªb	11.80 ^b	1.14 ^b	0.50ª	2.38ª

Values are represented as average of three replicates, different letters within the same column express significant differences at L.S.D. p>0.05, CD: Cow dung, SC: Sugar cane, RS: Rice straw, BL: Banana leaves, FS: Fish sludge

The N trend differed in potassium case, whereas K values were higher in all fish sludge vermicompost types (ranged from 2.13-2.38%) comparing with those values came from CD vermicompost types (ranged from 1.4-1.98%)) and the highest value of K achieved with vermicompost produced from (FS+BL) followed by (FS+RS) and (FS+SC) without significant differences (2.38 and 2.13%, respectively). In general, supporting FS or CD with plant wastes resulted in an increase in K content of the produced vermicompost.

The present study's results are in consistence with the finding of Chaulagain *et al.*⁹, who reported that mixing banana wastes with cow dung enhancing the nutrients status of the produced vermicompost.

Similar nitrogen content 1.108% nitrogen, in vermicompost using *E. eugeniae* had been reported by khucharoenphaisan and Kanokkorn³¹. However, the present study yielded higher potassium content in the produced vermicompost than that (1.318%) reported by khucharoenphaisan and Kanokkorn³¹.

Moreover, Bhat *et al.*^{32,33} mentioned that vermicompost contains a lot of macro and micro nutrients, such as nitrogen, potassium and phosphorus and that vermicompost is a new promising bio-fertilizer for sustainable agriculture as well as aquaculture as reported by Bhat *et al.*³⁴.

The obtained results of the current study indicated that providing CD or FS with agro-wastes during the vermicomposting process may enhance the nutrient status of the produced vermicompost, which is confirmed by several researchers, who elucidated that nutrients (i.e. N, P, K, Ca and Mg) are released and converted through microbial action, during the vermicomposting process, into more available soluble forms to the plants than those in the raw materials of the vermicompost and that the earthworm retain only a small percent of these nutrient (5-10%) within their bodies for growth⁷⁻⁹.

Numerous authors showed that, earthworm feeding affects the final product physiochemical characteristics. Differences can be also found grain size as well as water retention capacity^{35,36}. Vermicompost texture can be characterized as sandy loam³⁷.

According for Chauhan and Singh³⁸ findings, earthworms consumed various types of agricultural waste, such as vegetable waste, cattle dung, soybean meals, agricultural residue, as well as cow dung, sewage sludge and other industrial refuse. Thereby, biochemical quality of raw materials affect the quantity and quality of the produced vermicomposting³⁹⁻⁴¹.

Khucharoenphaisan and Kanokkorn³¹ showed that the quality of vermicompost is classified mainly based on the

macro nutrient content, such as nitrogen (N), phosphorous (P) and potassium (K) and adding soybean meal to the raw material during vermicomposting resulted in increasing nitrogen level to 1.1%, while the standard organic fertilizer has range from of (1.00-0.05%) of nitrogen according to Singh *et al.* and Sinha *et al.*^{42,43}. They also indicated that with testing six different material (cow dung, elephant dung, coconut shell's hair, watermelon peel, coffee ground and soybean meal) organic matters of vermicompost after 60 days of vermicomposting process the produced vermicompost was homogenized and the raw material could not be distinguished.

The increment in the nitrogen contents that is coincident with plant wastes adding can be explained by: More ventilation conditions (particularly with RS and SC) that facilitate more nitrification rate than that in CD and FS treatments, where the vermicompost tend to be compacted. Similarly, Dominguez⁴⁴ attributed the higher nitrogen content in vermicompost to higher nitrification rate. Also, in well ventilation conditions, earthworm can consume more wastes and increase the N content of the produced vermicompost, through addition of their excretory products, mucus. etc. as reported by Tripathi and Bhardwaj⁴⁵, more than in low ventilated environment.

For potassium high content in the vermicompost can be referred to high rate of mineralization within the earthworm gut as a result of the microbial activity. This explanation is confirmed by Suthar⁴⁶. Also, the addition of banana leaves to either FS or CD resulted in the highest K content compared to the other treatment, which can be attributed to higher K content in banana leaves compared to RS and SC wastes. For phosphorus content in the CD treatments of the present study are comparable to the general range of P in the vermicompost (0.1-0.3%) as reported by Kale *et al.*⁴⁷. However, the treatments of FS had higher P content, which required further studies to clarify the factors that controlling the P mineralization during the vermicomposting.

In general, different nutrients (i.e., nitrogen potassium and phosphorus) pattern and mineralization activities depend on total content of nutrients (i.e., nitrogen) in the initial waste and on the earthworm species and microbial activity during the waste decomposition^{46,48}.

Effect of different plant wastes and raw material on amino acids and growth promoter's content in vermicompost: Data in Table 2 show that the highest amino acids content was observed in (CD+BL) meanwhile the lowest amino acids content was noticed in the treatment of CD. Also, it can noticed that all vermicompost types that produced from

Asian J. Plant Sci., 20 (3): 370-379, 2021

TRT	Amino acids (mg1 00 g ⁻¹)	ABA (g 100 g ⁻¹)	GA (g 100 g ⁻¹)	IAA (g 100 g ⁻¹)
CD (Control)	0.27 ^e	0.33°	1.08 ^d	0.04 ^e
CD+RS	0.67 ^c	0.43 ^b	1.49 ^c	0.12 ^d
CD+SC	0.98 ^b	1.72ª	3.23ª	0.15 ^c
CD+BL	1.17ª	0.12 ^f	0.50 ^e	0.03 ^e
FS control	0.44 ^d	0.01 ^g	0.16 ^f	0.03 ^e
FS+RS	0.39 ^d	0.41 ^b	1.68 ^b	0.18 ^b
FS+SC	0.43 ^d	0.19 ^e	1.39 ^c	0.12 ^d
FS+BL	0.30 ^e	0.26 ^d	1.46 ^c	0.28ª

Table 2: Impact of agro-wastes on amino acids and growth promoters content of produced vermicompost

Values are represented as average of three replicates, different letters within the same column express significant differences at L.S.D. p>0.05, CD: Cow dung, SC: Sugar cane, RS: Rice straw, BL: Banana leaves, FS: Fish sludge

Samples	CFU counts g ⁻¹ vermicompost	Samples	CFU counts g ⁻¹ vermicompost
Manure (CD alone)	9.62±0.28 ^{bc}	FS alone	3.48±0.18 ^e
CD+RS rice straw	9.96±0.10ª	FS+RS	3.7±0.03 ^d
CD+Sc sugar cane	9.78±0.15 ^b	FS+SC	3.34±0.02 ^e
CD+BL banana leaf	9.50±0.17°	FS+BL	2.66±0.12 ^f
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Values are represented as average of three replicates, different letters within the same column express significant differences at L.S.D. p>0.05, CD: Cow dung, SC: Sugar cane, RS: Rice straw, BL: Banana leaves, FS: Fish sludge

mixing CD with plants wastes (RS, SC or BL) were higher in content of amino acids comparing with other treatments that produced by mixing FS with the same plant wastes (RS, SC or BL). The effect of adding agro-wastes (i.e., RS, SC and B) during vermicomposting process resulted in an increase in amino acids content in the CD treatments and a decrease in amino acids content in the FS treatments. Interestingly, the BL addition achieved the significantly highest amino acids content when added to CD, however, achieved the significantly lowest amino acids content when added to FS. In respect to the growth promoters, data in Table 2 revealed that (CD+SC) treatment surpassed all combinations in its content of Abscisic acid and gibberellins compounds, meanwhile this trend differed in auxin content (IAA) whereas (FS+BL) surpassed all tested combinations. As a general trend, the agro-wastes addition resulted in an increase in the plant growth promoter's content. Using SC with CD resulted in the significantly highest content of ABA and GA, while using RS with FS resulted in the significantly highest ABA and GA. However, the significantly highest content of IAA was measured in the treatment of FS and BL.

These results may be related with type of wastes that used for earthworm feeding and activities of both of earthworm and microbial during decomposing the organic wastes. Whereas, if the crops wastes rich in proteins and amino acids compounds thereby more amino acids will be found in vermicomposting as a result of protein and amino acids compounds degradation. Meanwhile, the situation will differ for growth promoter because it is related with earthworm activity during the agro-wastes decomposing and their excretion them from earthworm guts. Obtained results concern with growth promoter also indicated that adding crop wastes can be critical factor for plant growth promoters producing in vermicompost through indirect effect whereas these crop wastes affected the activity of earthworm and earthworm in turn produce these growth promoters in its gut as confirmed by Sinha *et al.*, Abul-Soud *et al.*, Xing *et al.*⁴⁹⁻⁵¹. Several studies indicated that these plant growth promoters are excreted from earthworm's guts and depended on its activities and population^{52-54,11}.

Also, Muscolo *et al.*²¹ showed that existing growth promoters (i.e., auxin-like) in vermicompost may be related with producing humic material during vermicomposting process and microbial action. Besides, organic wastes that are also offered to earthworms may have a great impact on earthworm's activities and reproduction rate and thereby secrete growth promoters from its guts and intestine^{16,17}.

Microbiological measurements: From Table 3 it could be noticed that there were markedly differences among studied vermibed combinations in CFU values. Generally, all CD combinations surpassed other FS combinations. In addition, adding agro wastes to vermibed enhanced microbial community comparing with CD or FS alone. Moreover, utilizing RS wastes in vermibed led to significantly increase in microbial CFU whether with CD or FS, comparing with individually treatment of CD and FS respectively. Meanwhile, the lowest value of Microbial community (CFU) was achieved with combination of (FS+BL).

On other side, combination (CD+RS) recorded the highest value of CFU comparing with all the studied combinations for CD and FS. Also, (CD+SC) combination as vermibed came in

Asian J. Plant Sci., 20 (3): 370-379, 2021

	<i>Citrobacter freundii</i> (mm)	<i>Enterobacter cloacae</i> (mm)	<i>Pseudomonas aeruginosa</i> (mm)	<i>Klebsiella pneumoniae</i> (mm)
CD	13	29	19	11
CD+RS	11	20	21	9
CD+SC	14	25	14	6
CD+BL	10	25		
FS	20	19	12	18
FS+RS	22	24	18	15
FS+SC	10	15		10
FS+BL	12	20	10	11

Table 4: Antimicrobial activity of vermicompost samples against identified pathogenic bacteria *Enterobacter cloacae, Pseudomonas aeruginosa*, Citrobacter *freundii* and *Klebsiella pneumonia* represented in inhibition zone diameter (mm)

CD: Cow dung, SC: Sugar cane, RS: Rice straw, BL: Banana leaves, FS: Fish sludge

the second rank in values of CFU whether on the level of CD or FS treatment separately. These results may be referred to that RS is considered a rich carbohydrates biomass that contains 38% cellulose, 25% hemicellulose, besides nutrients contents that are relevant to several application such as soil conditioner. Besides, adding rice straw to vermicompost bedding adjust humidity near to suitable range (80-90%) for rapid growth of *Eisenia fetida* and thereby for microbes during the process of vermicomposting according for findings by Domínguez and Edwards⁵⁵.

Moreover, current results showed that nitrogen content was high in vermicompost that produced from (CD+RS) comparing with other studied combinations whether for CD and FS combinations. In view of the facts of the highest N content which was mentioned by several studies^{56,57}, highly N content in vermicompost that produced from (CD+RS) combination) indicated that this combination was more preferred by earthworm to be feed on than the other treatments thereby will lead to more activities of earthworms in degradation of raw material that will be reflected positively on microbes activity.

Effective vermicompost extraction as antimicrobial: Among the eight vermicompost mixtures almost the studied samples had antagonistic activity against the four identified pathogenic bacteria (Citrobacter freundii, Enterobacter cloacae, Pseudomonas aeruginosa and Klebsiella pneumonia) except sample (CD+BL) against (Pseudomonas aeruginosa, Klebsiella and sample (FS+Sc) against pneumonia) (Pseudomonas aeruginosa). In contrary, CD, CD+SC, CD+BL and FS+RS had higher antimicrobial activity to Enterobacter cloacae and the inhibition zone diameter was (29, 25, 25 and 24 mm, respectively). While FS+RS, CD+RS and FS were higher antagonist with Citrobacter freundii, Pseudomonas aeruginosa and Klebsiella pneumoniae with 22, 21 and 18 mm diameter, respectively (Table 4).

These results may be due to a wide range of enzyme activities in vermicompost samples, which match with finding

of Devi *et al.*⁵⁸, who reported that there are fluctuations during the composting period whereas the maximum enzyme activities were observed during 21-35 days in vermicomposting and on 42-49 days in conventional composting. Moreover, the antimicrobial activity may due to production of extracellular enzymes of the isolates of vermicomposts which inhibit the working mechanism of selected pathogenic bacteria. Also, these results are coming in parallel with findings of Tiquia and Pietronave *et al.*^{59,60}, who showed that the antimicrobial activity of vermicompost may be attributed to activity of isolates microbial community in vermicompost that working against pathogenic bacteria. Generally, antimicrobial activity was characterized on the basis of the production of extracellular enzymes and antimicrobial substance which are important characters in vermicompost.

CONCLUSION

Utilizing agro-wastes (i.e., rice straw, sugar can and banana leaves) in vermicompost as promising organic fertilizers, is fetching several benefits for rural people, consumer health and the environment. Also, it is very important to determine the chemical composition and the best mixture ratio in order to achieve a good quality and stabilized final product. With vermicompost technology that acts as recycling nutrients from the agro-aquaculture wastes to soil several points will be achieved: raising awareness with concept of environment protection or friendly-environmental activities, relieve burden of agricultural production cost, developing rural life and reducing pollution sources through decreasing amount of usage of mineral fertilizers and pesticides. Finally, these efforts can contribute in developing vermicompost technology (as a source for nutrient, organic compost, amino acids, growth promoters and antimicrobial) to be sustainable industry that can produce vermicompost with specific quality as growers needed or requirements of plant stage. In addition, the results suggest that the vermicompost is a very promising untraditional organic

fertilizer with unique characteristics that can be used in fish pond fertilization to boost the phytoplankton production and resist pathogenic organisms. As, it will enhance the economic efficiency of the fish farms and animal farms as well as agricultural activities through varying their products.

SIGNIFICANCE STATEMENT

This study emphasized that there is a great potentiality to manipulate the vermicompost quality through adding different agro-wastes and that can be beneficial for growers and farmers through reducing doses of used mineral fertilizers thereby reduce agricultural cost and protect environment. This study will encourage the researchers to exert more effort to set an efficient protocol for vermicompost production with adequate quantity and high quality. Thus supporting exerted efforts to replace mineral fertilizer with vermicompost".

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