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Recycling of Maize stalk for promoting plant growth and sustainable agriculture using Fungi

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Abstract: This study aimed to use maize stalks for preparation of compost using *Aspergillus brasiliensis* and *Penicillium brevicompactum*. The prepared compost was applied to promote maize growth. Fungal analysis during compost preparation resulted in isolation of sixteen fungal species belonging to 5 genera from maize stalks (MS), farmyard manure (FYM) and different treatments at both 28°C (15 species and 5 genera) and 45°C (6 and 2). *Aspergillus*, *Fusarium* and *Penicillium* were the most common genera. *A. flavus*, *A. niger*, *A. brasiliensis*, *A. fumigatus*, *A. quadrilineatus*, *A. sydowii*, *A. ustus* and *F. solani* represented the most predominant species. Both organic matter (OM) and organic carbon (OC) were declined during composting to 26.41, 15.3 (T₁), 30.02, 17.4 (T₂), 27.5, 15.9 (T₃) and 31.42, 18.2 (T₄), respectively. On the other hand, total N reached the maximum percentage after 60 days meanwhile, C/N ratio decreased during composting from 26.6 to reach 11.7, 10.8, 12.2 and 12.6 for T₁, T₂, T₃ and T₄ respectively. It is worthy to mention that, the best prepared compost was that treated with mixed culture of *A. brasiliensis* + *P. brevicompactum* (T₄). This compost was applied in soil with different doses to improve maize growth. From the results, there was a significant increase in all measured parameters in different treatments, especially in case of treatment with 100% compost + 100% mineral N. Furthermore, the treatment mentioned above produced the best compost enriched the soil with organic matter and the availability of nitrogen, phosphorus and potassium.

Keywords: Compost; *Penicillium brevicompactum*; *Aspergillus brasiliensis*; maize stalks; maize growth

Introduction

Corn or maize (*Zea mays* L.) is an important cereal crop of the world, ranked the first in seed yield production. In Egypt, maize is the third most important staple food crop both in terms of area and production after wheat and rice (Hafez and Abdelaal 2015). Agricultural wastes are one of the major environmental pollutants; their biotechnological conversion is not only a remedy for environmental problems but also the source of suitable microbial by-products like food, fuel and chemicals (Bhupendra et al. 2022). These wastes are characterized as coarse plant by-products and chemically low in protein and fat contents. Also, it is high in lignin, cellulose, and hemicellulose contents. In Egypt, more than 89.03 million tons of solid wastes were generated, about 12% from Sohag. The problem of agriculture wastes becomes obvious, due to their aggregation after the harvest of summer crops. Therefore, getting rid of the wastes has the highest priorities, usually by burning, which is not only considered an economic loss, but also has harmful effects on the

environment because of emissions to the air. The storing of these wastes in the field leads to reproduction and growth of both pests and pathogens that will attack new crops. The conversion of lignocellulosic biomass to useful by-products has long been recognized as a desirable enterprise, and there is an increasing concern on utilization of cellulosic wastes as a promising alternative source of valuable products (Bayer et al. 2007; Bohacz 2018). Agricultural wastes are the most popular substrates due to their low-cost and availability and a cheap source of cellulose to produce different useful products all over the world. Effective waste management is among big challenges in most countries. Implementation of such management systems is usually hampered by lack of several crucial ingredients, such as information, organization, financial resources, complexity, or system multidimensionality (Burnley 2007). Furthermore, there is a large demand for compost made from agricultural solid wastes. It was reported by Chennaoui et al. (2018)

that, about 1% only of the total wastes is converted to compost.

Composting is a biological conversion of solid organic wastes into usable end products. It is one of the best alternative ways in managing different wastes. Composting of organic wastes is a bio-oxidative process involving the mineralization and partial humification of the organic matter, resulting in a stabilized final product, free of phytotoxines and pathogens, with certain humic materials (Nguyen and Shindo 2010). Also, composting is a spontaneous biological decomposition process of organic materials in a predominantly aerobic environment. During the process, microorganisms break down organic materials to stable, usable simple organic substances called compost and this compost is beneficial for plants as a fertilizer. Composting is gaining interest as a suitable option for manures with economic and environmental profits. Since this process eliminates or reduces the risk of spreading of pathogens, parasites and weed seeds associated with direct land application of manure and leads to a final stabilized product which can be used to improve and maintain soil quality and fertility (Larney and Hao 2007). Fortunately, the existing and developed composting technology already offers economically and environmentally interesting solutions that could substantially improve the hitherto used system of waste management (Sefouhi et al. 2010). Compost prepared from organic wastes also reduces the use of chemical fertilizers and environmental problems such as eutrophication due to leaching and deposition of nutrients from water bodies (Panisson et al. 2021). The active component involved in the biodegradation and conversion process during composting is the resident microbial community including fungi, bacteria and actinomycetes, among which fungi play a very important role (Anastasi et al. 2005). The adequate C/N ratio for composting is in the range 25–35:1 (Bishop and Godfrey 1983). Application of compost as organic fertilizer can increase soil organic carbon content directly and then improve aggregation, hydraulic conductivity, total porosity, and penetration resistance (Anastasi et al. 2005).

Huge amounts of agriculture wastes are annually accumulated in Egypt, also, there are some disadvantages of chemical and mineral fertilizers. Thus, this study focused on simple strategy to manage maize wastes collected from

one feddan maize farm by converting to compost using *Aspergillus brasiliensis* and *Penicillium brevicompactum*. Application of the prepared compost to enhance maize growth was also investigated.

Materials and methods

1. Selected strains

The methods of this work implemented preparation of compost using two fungal isolates and then, studying the impact of the best prepared compost on maize growth and productivity. *Aspergillus brasiliensis* AUMC 10988 strain is highly pectinase producer, isolated in a previous study from rotted guava fruit and *Penicillium brevicompactum* AUMC 10987 isolated from rotted apple fruit is highly cellulase producer. The two strains were used for production of compost from maize stalks collected from one feddan maize farm from Sohag Governorate, Egypt.

2. Isolation of fungi from maize stalks (MS) and farmyard manure (FYM)

The dilution- plate method (Johnson and Curl 1972) was used for isolation of fungi from maize stalks materials, farmyard manure and different treatments of composting process. One ml of the appropriate dilution was transferred directly into sterile Petri dish, then, 15-20 ml melted Czapek's agar medium were poured. The plates were incubated at 28°C for mesophilic and 45°C for thermophilic/thermotolerant fungi for 7 days during which the developing colonies were identified and counted (expressed as colony forming units "CFUs" per g dry sample).

3. Preparation of compost

a. Inoculum preparation:

Aspergillus brasiliensis AUMC 10988 and *Penicillium brevicompactum* AUMC 10987 were cultured separately in sterilized wheat bran for 3 days and incubated at 28°C, then the spore suspension (10^8 spore/ml) was prepared using sterilized water to make the inocula.

b. Preparation of composted heaps:

Composting of maize stalks from one feddan maize was carried out by pile method. Maize stalks (MS) were grinded before compost process then mixed with farmyard manure (FYM) (1 : 2), then the heaps were enriched with water (50-60%) (Kluczek-Turpeinen 2007). The heaps were classified into four treatments (280 kg/heap):

Treatment 1 (T1): MS + FYM (control); treatment 2 (T2): MS + FYM + 3 litre spore suspension of *A. brasiliensis* (10^8 spore /ml); treatment 3 (T3): MS + FYM + 3 litre spore suspension of *P. brevicompactum* (10^8 spore /ml); and treatment 4 (T4): MS + FYM + *A. brasiliensis* + *P. brevicompactum* (the same as the above concentration). During composting, mixture was turned every week for air circulation and temperature homogeneity. Three replicates of each compost samples were taken at 0, 60, and 90 days to determine the physical and chemical properties. An appropriate amount of water was sprinkled onto the heap to keep the moisture content.

c. Physical and chemical analysis of the compost

The composts in each replicate per treatment were mixed thoroughly and 300 g samples were weighed and brought to the laboratory. The parameters measured in this study include indicators of compost quality such as the major plant nutrients as follows: total N (%), total P_2O_5 (%), total K_2O (%), O.M % and Physical (temperature, pH and electrical conductivity. The weight of the compost and fresh moisture content per replicate per treatment were also measured in this study for comparison. Other compost quality parameters like color, smell, and texture were also evaluated. Color and smell were described based on the physical appearance and odor of the compost. Texture was determined by feel method and were described accordingly soil-like, gritty, coarse, and the like (Navarro et al. 1993).

d. Pot experiment

According to the results obtained from the above experiment, the best compost treatment (T4: MS + FYM + *A. brasiliensis* + *P. brevicompactum*) was applied in five treatments. These treatments were allocated to the

Results

1. Chemical properties of maize stalks (MS) and farmyard manure (FYM)

The chemical characteristics of maize stalks (MS) and farmyard manure (FYM) were shown in Table (1). In MS, pH values were weak acidic (pH = 5.22), potassium content, organic matter (OM%) and organic carbon (OC%) were

recommended dose of nitrogen (120 Kg/fed), potassium (50 Kg/fed) and phosphorus (100 Kg/fed). The treatments were as follows; T1: Recommended dose of N (120 kg N/fed) (100% mineral N), T2: 100% compost + 100% mineral N, T3: 75% compost + 25% mineral N, T4: 50% compost + 50% mineral N, and T5: 25% compost + 75% mineral N.

Maize (single hybridized, No. 2055) was used as a test plant in order to study the impact of prepared composts on plant growth, nutrients uptake, and nutrient content in the soil after harvesting. Five kg dried soil were mixed carefully with the prepared compost + 0.25 g potassium sulphate + 0.5 g superphosphate in plastic pots. Then five maize grains were sown in each pot and irrigated to field capacity. Urea was added as a source of mineral N (46%N) after 15 days. Plant samples were taken after 60 days to determine fresh and dry weights. The samples were then dried and ground for further analysis.

e. Soil analysis:

Physical and chemical analysis of the best treated soil in the beginning of the experiment and after 60 days were assessed at Agriculture Research Center, Sohag, Egypt.

f. Plant growth parameters:

Chlorophyll content of maize leaves, leaf area, fresh and dry weights of plant shoot and roots and total nitrogen, phosphorus and potassium were measured (Reynolds et al. 1998).

4. Statistical analysis

The data were analyzed statistically using analysis of variance (ANOVA). Differences among the means were determined for significance at $P \leq 0.05$ or 0.01 using Duncan's multiple range test by SPSS V10 software.

relatively high (1.53, 80.52, 41.55%, respectively). But in general, MS had low phosphorus (0.14%) and nitrogen (0.48%) contents. On contrast, the C/N ratio was clearly high, representing 86.56%. Comparatively, FYM was characterized by higher pH (9.5%), phosphorus (0.40%) and nitrogen (0.76%); but lower OM% and OC% (38.98 and 22.6%, respectively).

Table 1. Initial chemical analysis of maize straw (MS) and farmyard manure (FYM).

Property	MS	FYM
pH	5.22	9.5
EC (dS/m)	4.52	8.62
OM%	80.52	38.98
OC%	41.55	22.6
C/N Ratio%	86.56	29.7
Macronutrients %: N	0.48	0.76
P	0.14	0.40
K	1.53	2.95

EC: Electrical conductivity; OM%: Organic matter percentage; OC%: Organic carbon percentage; C/N: Carbon/Nitrogen; N: nitrogen, P: phosphorus and K: potassium.

2. Fungal analysis

a. Fungi isolated from maize stalks (MS) and farmyard manure (FYM)

Seven identified fungal species belonging to 3 genera were isolated on Czapek's medium at 28°C from both maize stalks (MS) and farmyard manure (FYM). Maize stalks recorded higher fungal propagules (30×10^3 cfu/g) than farmyard manure (25×10^3). Four species belonging to 3 genera were isolated from maize stalks, while FYM contained 5 species belonging to two genera (Table 2). *Aspergillus flavus* and *A. niger* were the most common species, representing 29, 20, 3 CFUs/g (for *A. flavus*) from MS, MS + FYM and FYM, respectively and 15, 4 CFUs/g (for *A. niger*) from MS and MS + FYM, respectively.

b. Fungi associated with compost

Fungal genera and species were estimated during composting process on Czapek's agar medium at both 28 and 45°C.

c. Mesophilic Fungi

T3 and T4 (Fig. 1).

Aspergillus flavus, *A. niger* and *A. brasiliensis* were the most abundant fungal species in all treatments. There were significant differences between different treatments of compost in the qualitative and quantitative fungal diversity Table (2). The highest number of species (6 species) were reported from treatment with mixture of maize straw + farmyard manure + *Penicillium brevicompactum* (MFP) after three months, while control (MF) or compost treated with MFAP (maize straw + farmyard manure + *A. brasiliensis* + *P. brevicompactum*) showed the lowest number of fungal species (3).

3. Physical and chemical properties of compost

Temperature: Temperature of composted heaps was measured every week up to 90 days. The obtained results reported an increase in temperatures of all compost treatments reaching to 45°C or 50°C within the first month of composting process compared to control (40°C). Maximum temperature degree (50°C) was obtained in treatments nos.

Table 2. Total counts x 10³ (TC, calculated per 1 g) of fungi isolated from maize straw (MS), farmyard manure (FYM) and different treatments of compost on Czapek's agar (Cz) at 28°C.

Fungal taxa	MS	FYM	Ms + FYM 0	Treatments							
				MF		MFA		MFP		MFAP	
				2	3	2	3	2	3	2	3
<i>Aspergillus</i>	44	18	24	15	12	6	27	12	20	16	6
<i>A. awamorii</i>					6						6
<i>A. brasiliensis</i>						2	12			5	
<i>A. flavus</i>	29	3	20	9			1	5	3	1	
<i>A. nidulans</i>		4				1			6		
<i>A. niger</i>	15		4	6	5	3	14	7	10	6	
<i>A. ochraceous</i>										4	
<i>A. sydowii</i>		8			1						
<i>A. terreus</i>									1		
<i>A. ustus</i>		3									
<i>Curvularia lunata</i>									1		
<i>Fusarium</i>							6	2			6
<i>F. solani</i>							6	2			4
<i>F. verticillioides</i>											2
<i>Mucor racemosus</i>	1	3									
<i>Penicillium</i>	2			1		1		1	3		
<i>P. aurantiogriseum</i>				1		1					
<i>P. brevicompactum</i>	2							1	3		
Sterile mycelia (White)		4									
Total counts	27	35	44	16	12	18	33	15	24	16	12
No of genera (5)	3	2	4	2	1	2	2	3	3	1	2
No of species (15)	4	5	7	3	3	4	4	4	6	4	3

MF: Maize straw + farmyard manure (control). MFA: maize straw + farmyard manure + *Aspergillus brasiliensis*. MFP: maize straw + farmyard manure+ *Penicillium brevicompactum*, MFAP: maize straw + farmyard manure+ *Aspergillus brasiliensis* + *Penicillium brevicompactum*; 0 = at zero time, 2 = After two months and 3 = After three months of composting.

d. Thermophilic and thermotolerant fungi

At the end of composting, the total fungal count isolated at 45°C from compost treated with *A. brasiliensis* and *P. brevicompactum* were 6x10³ and 4x10³cfu/g, respectively. While no fungal species recorded after three months in both control and

compost treated with mixed culture of *A. brasiliensis* and *P. brevicompactum*. Thermotolerant fungi were represented mainly by 5 species belong to genus *Aspergillus* (Table 3).

Table 3. Total counts x 10³ (TC, calculated per 1 g) of fungi isolated from maize straw, farmyard manure and different treatments of compost on Czapek's agar (Cz) medium at 45°C.

Fungal taxa	Maize Straw	FYM	MS + FYM (0)	Treatments							
				MF		MFA		MFP		MFAP	
				2	3	2	3	2	3	2	3
<i>Aspergillus</i>	1	14	6	4		1	6	1	4	3	
<i>A. fumigatus</i>	1		2	4			1	1			
<i>A. niger</i>							5		3		
<i>A. quadrilineatus</i>		4				1			1		
<i>A. sydowii</i>			2							1	
<i>A. ustus</i>		10	2							2	
<i>Mucor racemosus</i>		3				2					
Total counts	1	17	6	4	0	3	6	1	4	3	0
No. of genera (2)	1	2	1	1	0	2	1	1	1	1	0
No. of species (6)	1	3	3	1	0	2	2	1	2	2	0

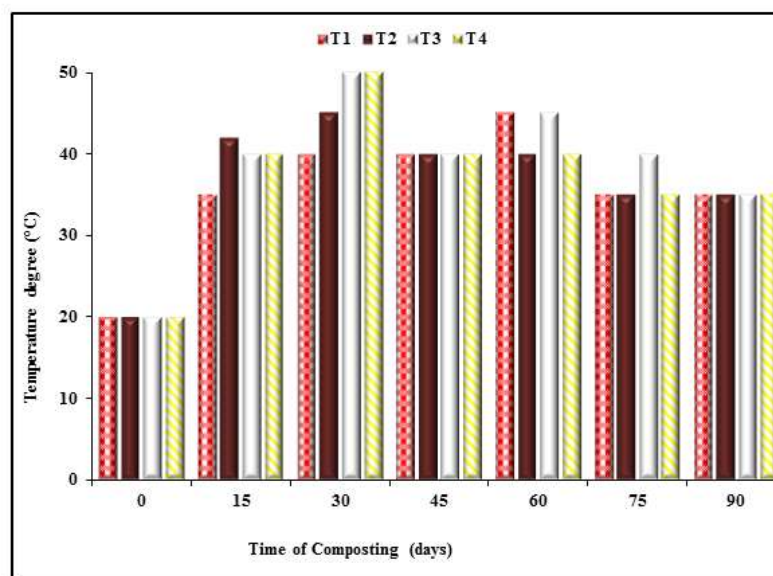


Fig. 1 Temperature changes of composted heaps during 90 days.

pH values: In all treatments, pH values increased after 15 days of composting process, reporting the highest value in T4 (8.55), then decreased gradually to 7.53 after 75 days (Table 4).

Table 4. Changes in pH values and electrical conductivity during composting process.

Parameters	Time (in days)	Treatments			
		T1	T2	T3	T4
pH	0	7.83	7.83	7.83	7.83
	15	8.3	8.42	8.2	7.89
	30	8.4	8.45	8.5	8.45
	45	7.66	7.33	7.28	6.89
	60	7.69	7.52	7.72	7.44
	75	7.7	7.22	7.65	7.63
	90	7.06	7.40	7.05	7.55
EC (mS)	0	5.6	5.6	5.6	5.6
	15	5.66	5.81	5.74	5.82
	30	5.71	5.86	5.94	6.02
	45	5.77	5.88	6	6.33
	60	5.86	5.94	6.05	6.48
	75	5.63	5.83	5.68	5.54
	90	5.63	5.27	5.54	5.52

T1: MS + FYM (control), T2: MS + FYM + *A. brasiliensis* AUMC 10988, T3: MS + FYM + *P. brevicompactum* AUMC 10987 and T4: MS + FYM + *A. brasiliensis* + *P. brevicompactum*.

Electrical conductivity (EC): Electrical conductivity slightly increased in some treatments reaching its maximum value (6.48 mS) in the second month in compost treated with the mixture of *A. brasiliensis* and *P. brevicompactum* (T4), then, the EC decreased to 5.52 at the end of experiment. However, in other treatments, it was fluctuated between 5.6 to 6.05 mS (Table 4).

Organic matter and organic carbon: The most valuable obtained results were significant decrease (to more than 50%) in organic matter (OM) and organic carbon (OC) during the time of composting up to 26.41, 15.3 (in T1), 30.02, 17.4 (in T2), 27.5, 15.9 (in T3) and 31.42, 18.2 (in T4), respectively (Table 5).

Total nitrogen percentage (N%): It was observed that, total N reached its maximum value after 60

days in T4 (2.1%), then decreased within the third month to 1.45% (Table 5).

Carbon / Nitrogen ratio (C/N): Results presented in Table (5) revealed high significant decrease in C/N ratio during composting in all treatments, reporting the least value (10.8) after 90 days in T2.

Total Phosphorus and Potassium (%): Compost treated with either *A. brasiliensis* or *P. brevicompactum* or both lead to significantly increase in phosphorus and potassium concentrations during the second month of composting then decreased, at the third month. The highest values of total P and K % (0.88 and 2.21 respectively) were observed in T3 (Table 5).

4. Soil analysis

Results showed that, the soil used in pot experiment was characterized by clay loam and low organic matter content (Table 6).

Table 5. Changes of some chemical properties of compost during 90 days.

Parameters (%)	Time (in days)	Treatments			
		T1	T2	T3	T4
OM	0	62.35	62.35	62.35	62.35
	60	40.3	34.75	30.6	30.03
	90	31.42	30.02	27.5	26.41
OC	0	36.2	36.2	36.2	36.2
	60	23.04	20.2	17.7	17.5
	90	18.2	17.4	15.9	15.3
Total N	0	1.36	1.36	1.36	1.36
	60	2.1	2	1.5	1.6
	90	1.45	1.6	1.3	1.3
C/N ratio	0	26.6	26.6	26.6	26.6
	60	11.5	10.1	11.8	10.9
	90	12.5	10.8	12.2	11.7
Total P	0	0.443	0.443	0.443	0.443
	60	0.84	0.85	0.88	0.82
	90	0.81	0.84	0.85	0.71
Total K	0	1.78	1.78	1.78	1.78
	60	1.94	2.2	2.21	1.95
	90	1.84	2.01	1.91	1.62

Table 6. Initial physical and chemical properties of soil used in pot experiment.

Soil Properties	Values	Soil Properties	Values
Soil fractionation (%): Sand	23.9	CaCO ₃ %	2.8
Silt	38.5	Soluble anions (meq/L): CO ₃ ²⁻	-
Clay	37.6	HCO ₃ ⁻	0.9
Soil texture	Clay loam	Cl ⁻	11.4
Saturation point (%)	70	SO ₄ ²⁻	3.2
EC mmhos/cm (1 : 2.5)	1.95	Available nitrogen (ppm)	83
pH (1 : 2.5)	7.85	Available phosphorus (ppm)	18
Organic matter %	0.81	Available potassium (ppm)	168
Soluble cations (meq/L): Ca ⁺⁺	5.8	Iron (ppm)	19.3
Mg ⁺⁺	2.5	Manganese (ppm)	7.28
Na ⁺	6.1	Copper (ppm)	0.32
K ⁺	1.1	Zinc (ppm)	0.44

5. Impact of the prepared compost on maize plant growth and nutrient uptake

The effect of the compost, prepared from maize wastes and farmyard manure with the best

properties (T4), on maize growth parameters and nutrients uptake was studied. Chlorophyll contents of maize leaf was measured *in vivo* after 60 days. Also surface leaf area, stem height, fresh and dry weight of shoot and root systems, as well

as nutrients uptake were determined after harvesting. Generally, most of the measured parameters (except dry weight of roots) were significantly increased in different treatments (Table 7 and Figures. 2-5).

The measurements of total N, P and K uptake in maize plant were recorded after 60 days from application of different compost treatments (Table 8). There is a notable increase in total N content of shoots, ranging between 0.17 to 0.52 g/pot, but reaching its maximum value in T2 (100% compost + 100% mineral N). Also, phosphorus uptake showed significant increase when using a combination of compost with mineral N compared to those of mineral N alone. Precisely, K uptake

increased significantly when plant cultivated in soil treated with 100% compost + 100% mineral N (T2). Other treatments showed non significant effect on K uptake (Table 8).

6. Impact of the prepared compost on organic matter and N, P, and K contents of soil

The above results revealed that, treatment of soil with 100% compost +100% mineral N (T2) gave the best output on maize growth, therefore, this soil was analyzed after harvesting of maize plant to assess its organic matter and N/P/K contents. From the results, the organic matter content, as well as nitrogen, phosphorus and potassium contents of soil were totally increased (Table 9).

Table 7. Chlorophyll content of leaf, surface leaf area, stem height, fresh and dry weight of roots and shoots of maize plant cultivated for 60 days in soil treated with compost prepared using mixed culture from *A. brasiliensis* and *P. brevicompactum*.

Plant parameters Treatments	Mean ± Std. Error						
	Chlorophyll content of leaf (spad unit)	surface leaf area (cm ²)	Stem height (cm /plant)	Fresh weight (g/pot)		Dry weight (g/pot)	
				Shoot	Root	Shoot	Root
T1: (100% mineral N)	19.33 ±0.09 ^a	200.1±3.5 ^b	24.27±0.15 ^b	113.43 ± 3.17 ^a	4.78±0.36 ^a	21.43±0.09 ^b	1.50±0.05 ^{ab}
T2: 100% compost+ 100% mineral N	22.57±0.13 ^d	223.84±1.61 ^c	27.39±0.20 ^d	144.47 ± 0.57 ^c	7.29±0.19 ^c	26.27±0.64 ^c	1.70±0.03 ^b
T3: 75% compost + 25% mineral N	21.07±0.09 ^c	188.23±4.75 ^a	26.11±0.56 ^c	131.11 ± 4.22 ^b	5.12±0.16 ^{ab}	20.81±0.46 ^b	1.34±0.01 ^a
T4: 50% compost + 50% mineral N	19.30±0.30 ^a	192.48±1.04 ^a _b	25.93±0.35 ^c	115.8 ± 2.54 ^a	5.93±0.34 ^b	20.35±0.48 ^b	1.51±0.05 ^{ab}
T5: 25% compost + 75% mineral N	20.27±0.26 ^b	188.36±4.50 ^a _b	22.61±0.43 ^a	113.41 ± 1.05 ^a	4.83±0.23 ^a	18.65±0.15 ^a	1.39±0.30 ^{ab}
F value	48.712 ^{**}	18.905 ^{**}	24.925 ^{**}	26.543 ^{**}	15.485 ^{**}	46.736 ^{**}	2.409 ^{ns}

One way–ANOVA was performed for each amendment treatment; Means in the same amendment treatment group with different letters are significant (* P < 0.05), high significant (** P < 0.01) or NS= non- significant difference according to the Duncan test.

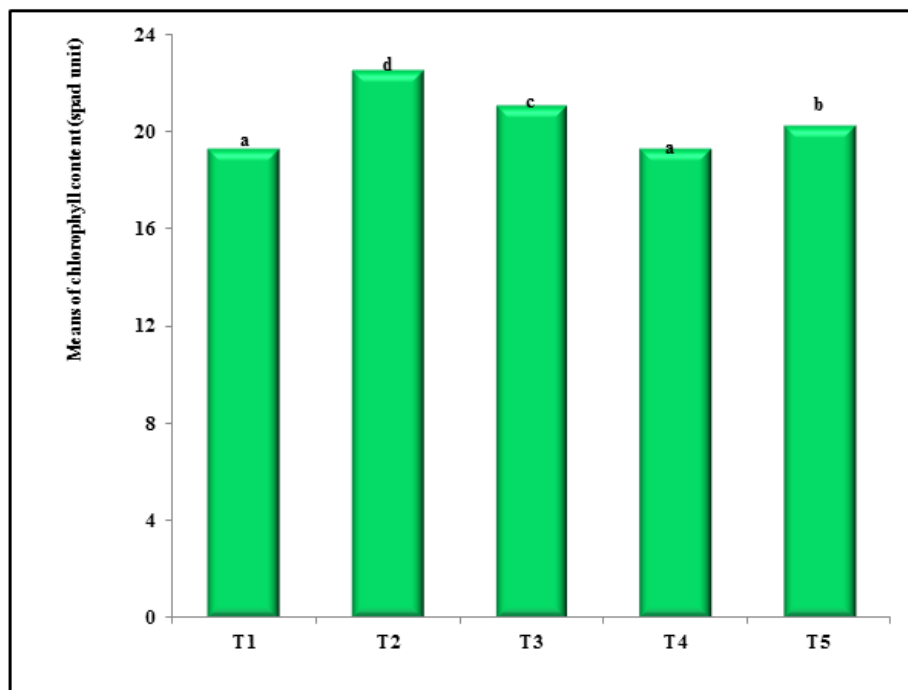


Fig. 2 Means of chlorophyll content (spad unit) of maize leaves after 60 days of planting.

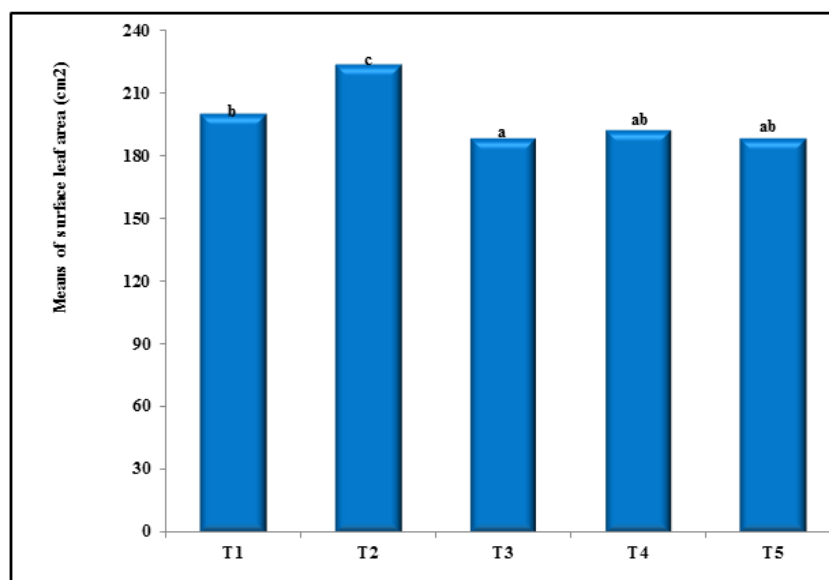


Fig. 3 Means of surface leaf area (cm²) of 60- days old maize plant.

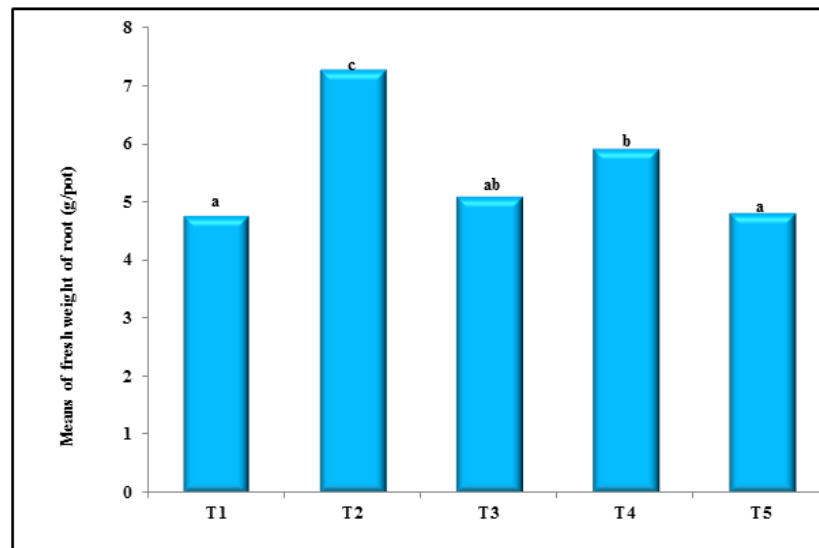


Fig. 4 Means of fresh weight of root (g/ pot) of maize plant after harvesting (after 60 days).

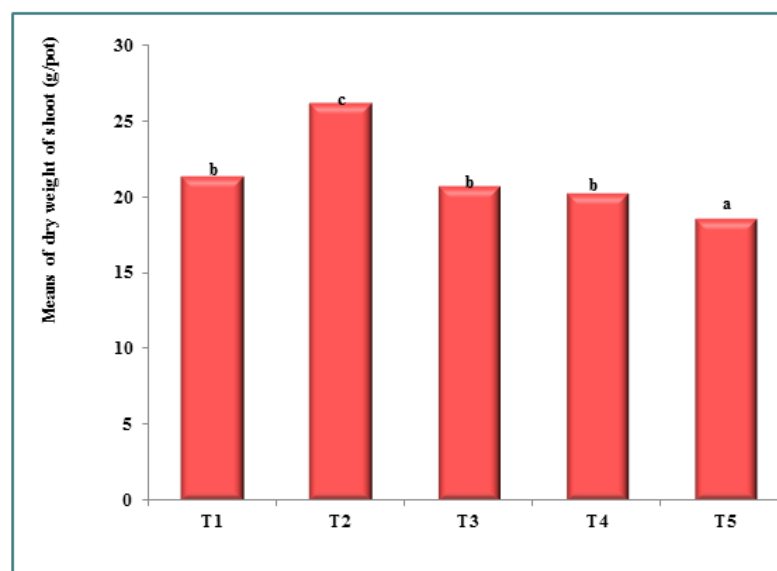


Fig. 5 Means of dry weight of shoot (g/ pot) of maize plant after harvesting (after 60 days)

Table 8. Nitrogen, phosphorus, and potassium uptake of maize plants cultivated for 60 days in soil treated with affected compost prepared using mixed culture from *A. brasiliensis* and *P. brevicompactum*.

Treatments	Mean ± Std. Error		
	N uptake (gm/pot)	P uptake (gm/pot)	K uptake (gm/pot)
T1: (100% mineral N)	0.38±0.009 ^c	0.21±0.01 ^a	0.06±0.001 ^a
T2: 100% compost+ 100% mineral N	0.52±0.020 ^d	0.43±0.02 ^c	0.10±0.004 ^b
T3: 75% compost + 25% mineral N	0.25±0.020 ^b	0.26±0.01 ^b	0.07±0.004 ^a
T4: 50% compost + 50% mineral N	0.17±0.005 ^a	0.30±0.01 ^b	0.07±0.003 ^a
T5: 25% compost + 75% mineral N	0.22±0.003 ^b	0.27±0.01 ^b	0.06±0.002 ^a
F value	92.695 ^{**}	38.969 ^{**}	23.51 ^{**}

One way ANOVA was performed for each amendment treatment; Means in the same amendment treatment group with different letters are significant (*, P < 0.05), high significant (**, P < 0.01) or NS= non -significant differences according to the Duncan test.

Table 9. Organic matter, N, P, and K contents of soil before and after treatment with 100% compost + 100% N (T2).

Soil contents	Values	
	Before	After
Organic matter %	0.81	1.2
Available nitrogen (ppm)	83	175
Available phosphorus (ppm)	18	19.91
Available potassium (ppm)	168	225

Discussion

Application of *Aspergillus brasiliensis* AUMC 10988 and *Penicillium brevicompactum* AUMC 10987 for compost production

This work could improve solid waste management converting it into valuable compost. Composting is the importance idea for remediation of organic waste in Egypt and other agricultural developing countries. Composting is an economical and safe technique for transformation of solid organic matter to more stable and mature humic substance by the activity of microorganisms under controlled conditions. In the current study, maize wastes could be converted into valuable compost using spore suspensions from *A. brasiliensis* and/or *P. brevicompactum*. Crop residues are considered the main resources available for composting (Ancuta *et al.* 2011). Composting was extensively used in bioremediation and biodegradation of green wastes (Unmar and Mohee 2008). Maize stalks is rich in carbon and has low density and moisture content, making it suitable for use as a bulking agent during composting (Kumar *et al.* 2010). The present work revealed that, the C/N ratio of maize stalks (MS) was high (86.56:1) for this reason, its break down is very slow (Rashwan 2016). Farmyard manure (FYM) is a valuable resource for organic fertilizers because of its high organic matter and nutrient content (Nada 2015). The results showed that the C/N ratio of FYM was 20.79 : 1 which is nearly similar (18.06 : 1) to the findings of Mahmoud (2010).

Mycobiota of maize stalks (MS) and farmyard manure (FYM)

Our results about number of isolated genera (3,2) and species (4,5) from MS and FYM, respectively are disagreement with the data representing by Rashwan (2016) who could isolate 11 species from sorghum wastes, 8 species from FYM. Also, Mahmoud (2010) isolated 8 and 11 fungal species from FYM and date palm residues, respectively. Furthermore, Kumar *et al.* (2008) could isolate 10 species of thermophilic fungi from wheat straw, farmyard manure and soil.

Mesophilic mycobiota of compost

Fungi play an important role in composting by breaking down tough debris and enabling other microorganisms to continue the decomposition process. So, the fungal diversity

during composting process was studied. The results showed that the total counts of fungi in all compost treatments after three months ranged between 12×10^3 - 33×10^3 cfu/g and the highest total count (33×10^3 cfu/g) was recorded in compost inoculated with *A. brasiliensis* (MFA). While the highest number of species (6 species) was isolated from compost inoculated with *P. brevicompactum* (MFP). In agreement with the current results, it was reported that, the total count of fungi in compost ranged between 10^5 - 10^6 cfu/g (Mahmoud 2010). Determination of the resident microbial diversity of the compost and their benefit role is essential for: (1) detecting pathogenic microorganisms, optimizing compost quality standards; (2) identifying the active microbiota in compost that are beneficial to soil and plant health and (3) production of quality and efficient compost suitable for agricultural and commercial applications (Ashraf *et al.* 2007, López-González *et al.* 2015).

Aspergillus niger was observed to be the most dominant species in all compost treatments. This may be due to the antagonistic activity of *A. niger* that suppress the activity of other fungi (Suárez-Estrella *et al.* 2007). Fungal species isolated at the end of composting process (*Aspergillus brasiliensis* and *Curvularia lunata*) are in agreement with the results of Olabode *et al.* (2016) who isolated the same species at the final stage of composting (10 weeks) of wood dust with cow dung. It was shown that, fungal biomass in compost exceeds bacterial biomass, this is probably related to low temperatures and moisture content during composting (Mehta *et al.* 2014). Also, fungi have superiority in degradation of lignocellulosic polymers and other complex carbon substrates and tolerance to extreme conditions, therefore, they are mainly responsible for compost maturation (De Gannes *et al.* 2013).

Most of the fungal species recorded in the current study are known to be common saprobes on soil and dead plant tissue. Fungal total count decreased gradually during composting process, reaching the minimal count at the end of composting in MF and MFAP treatments. In agreement, Gebeyehu and Kibret (2013) recorded high fungal count at the beginning of composting process, which decreased at the end of composting period (after 90 days), may be due to increase in temperature during thermophilic stage of composting process. Also, it was suggested that, the decline in the number of fungi is due to the

presence of conditions unfavorable to their proliferation (Chennaoui *et al.* 2018).

Thermophilic and thermotolerant mycobiota

Mesophilic and/or thermophilic fungi play the main role in the decomposition of organic materials, such as cellulose, hemicellulose and lignin from raw materials forming valuable end products (Desoki 2004; Chennaoui *et al.* 2018). There is a notable variation in fungal diversity during composting by different treatments. This variation may be depending on change of physico-chemical features in different stages of composting and competition between microbial flora. In the present study, fungi isolated at 45°C were mainly represented by five species of the genus *Aspergillus* (*A. fumigatus*, *A. niger*, *A. quadrilineatus*, *A. sydowii* and *A. ustus*). These results agree with those obtained by Olabode *et al.* (2016) who isolated *A. flavus*, *A. fumigatus*, *A. niger* and *A. ochraceus* at the final stage of composting wood dust with cow dung. Ashraf *et al.* (2007) stated that the cellulolytic fungi, such as *Aspergillus*, *Trichoderma*, *Penicillium* and *Trichurus* accelerate composting for efficient recycling of dry crop wastes with high C:N ratio and reduce the composting period. In the final stages of composting, fungi, in cooperation with bacteria, have very important role in acceleration of the decomposition process and in production of mature compost by eliminating microbial pathogens. These microorganisms continue to degrade polymer compounds in the waste material resulting stable and high quality compost at the end of the cycle.

Changes in physical and chemical properties of compost

Temperature is one of the physical characteristics indicating the level of decomposition during the composting process. It may be considered as one of the key factors in the selection of species in microbial communities during composting. Similarly, Baffour-Asare (2009) and Gebeyehu and Kibret (2013) found that the compost heaps reached their highest temperatures within the first two weeks of composting and maintained temperatures above 45°C for almost four weeks. The composting materials subjected to three typical degradation phases: mesophilic, thermophilic as well as cooling and maturation (Saludes *et al.* 2008). Increasing in temperature at the beginning may be due to high available carbon content which provides favorable

conditions for high rate of growth and biological activity of microorganisms (Novinscak *et al.* 2007). In the present work during composting, the temperature values decreased rapidly to reach the minimum values at the end of experiment, in accordance, Ogunwande *et al.* (2008) recorded that, the temperature tends to decrease after the thermophilic phase due to depletion of the substrate, leading to reduction in microbial activity. The fluctuation in temperature during composting process explains that the compost subjected to different stages of fermentation.

pH of the compost is necessary for most biochemical reactions catalyzed by enzymes which allows the bioavailability of nutrients and the solubility of mineral elements for microorganisms. In the current study, pH values decreased after the first 45 days of composting process in all treatments, reaching its minimum value in T3 (7.28). In studying compost inoculating with *Bacillus cereus*, Ribeiro *et al.* (2017) found that pH value the decomposition of sugarcane bagasse and coast-cross grass decreased, giving 6.3. The decrease in pH values at the beginning of composting process was probably due to production of organic acids during the bio-oxidation and active degradation stage of composting process (Kiehl 2002). Afterwards, these organic acids react with the released bases from the organic matter or subjected to metabolic degradation and/ or lost by volatilization or ammonification process, therefore, the pH values thereafter increased and become alkaline. Such pH variation during composting was recorded previously (Gebeyehu and Kibret 2013; El-Hagar *et al.* 2015; Adamcová *et al.* 2016).

The electrical conductivity (EC) evaluates the salinity level in the compost and characterizes the inhibitory and harmful impacts on the plant growth. The highest value of Electrical conductivity (EC) was observed in T4 (up to 6.48 ms), followed by T3 (up to 6.05), this may be related to high concentration of ammonia released during the ammonification of proteins, amino acids and peptides, in addition to releasing soluble inorganic and nutrient ions during the mineralization of organic materials (Badawi 2003; Desoki 2004; El-Hagar *et al.* 2015). Also, in consistence with the current results, Chennaoui *et al.* (2018) recorded that, the EC showed an increase from the initial value of 4.9 ms/cm to a maximum of 7.5 ms/cm, then decrease gradually at the end of

the composting process. It was noticed an increase in EC values to 19.74 mS/cm in compost studied by Kianirad *et al.* (2010). It is worthy mentioning that, releasing of ammonia and the precipitation of mineral salts could cause the decrease of the EC at the final stage of composting (Wong *et al.* 1995). Compost with a low EC can be used directly while the high EC compost must be thoroughly mixed with soil or other low EC materials before it can be used for crops (Chennaoui *et al.* 2018).

The decrease in organic matter (OM) and organic carbon (OC) after 3 months may be attributed to the loss of carbon as CO₂ by microbial oxidation during the composting process (Charisiou *et al.* 2012). Also, the OM is decomposed and transformed to stable humic compounds usually substantial during the bio-oxidative stage, when the temperature and the microbial activity are high, because of the organic matter provides the major energy to microbial metabolism (Amir *et al.* 2004). This could be because most of the easy biodegradable substances have been depleted by microorganisms during the first stage of the composting process. Then, when compost becomes stable and mature; the activities of microorganisms decrease as the result of lack of energy (Gebeyehu and Kibret 2013). Baffour-Asare (2009) reported a similar pattern of reduction in organic carbon in dewatered sewage sludge and sawdust compost. At the end of composting, CO₂ was the main source of carbon loss, the remaining carbon losses were caused by the emission of CH₄ and other volatile organic compounds. In the study of Chennaoui *et al.* (2018), the organic matter decreased during the composting process, from about 92% to about 48% after 60 days of composting.

In the current work, the total N% increased during composting period in all treatments from 1.36 to 2.1% after 60 days, then decreased to 1.45 % in T4 after 90 days. Similar results were recorded by El-Hagar *et al.* (2015), who found that the nitrogen content increased during composting period of dry leaves or bagasse. An increase in the nitrogen content during composting process is indication to the biomass lost, or the nitrogen may be lost through volatilization (Ribeiro *et al.* 2017). Furthermore, Gebeyehu and Kibret (2013) recorded reduction in nitrogen content during composting, which may be due to the utilization of inorganic nitrogen by bacteria in the composting process and the conversion of nitrogen into bacterial proteins or

could be attributed to mineralization of organic nitrogen by microbial activity during decomposition. Differences in nitrogen content among all treatments may be caused by immobilization of nitrogenous fractions, volatilization as NH₃ during oxidation at thermophilic phases (Jurado *et al.* 2015). The decrease in NH₄⁺ is characteristic for a good maturation of compost, also nitrogen in the form of NO₃⁻ content stimulate growth of microorganisms during the composting process (Chennaoui *et al.* 2018).

The initial carbon to nitrogen ratio (C/N) is one of the most important factors influencing compost quality. The results obtained showed that, C/N ratio sharply decreased at the end of the process. In this respect, Van Heerden *et al.* (2002) found that composting of citrus wastes was completed after 3 months giving 7 C/N ratios. Moreover, the results of Kianirad *et al.* (2010) showed that, the C/N ratio did not stabilize after 75 days because of continuous decomposition of the organic matter. In general, initial C/N ratios of 25–30 are considered ideal for composting (Kumar *et al.* 2010). However, recently some researchers have successfully carried out composting at lower initial C/N ratios, which can increase the amount of manure treated, and increase the loss of nitrogen as ammonia gas. Gebeyehu and Kibret (2013) found that the C/N ratio was reduced from 19.7 to 10.7 and the temperature increased, and this is of crucial importance for efficient mineralization, which in turn results in reduction in C/N ratio. The total nitrogen contents of treatments increased during composting, while the initial C/N ratio mainly influenced by the maturity of the final compost. In the current study, C/N ratio in the final stage of composting (12.9) are markedly higher than that resulted by Adamcová *et al.* (2016) (12) and Chennaoui *et al.* (2018) (11). This decrease could be explained by those consuming microorganisms more carbon than nitrogen. The study of Czekala *et al.* (2016) on composting of maize stalks content with sewage sludge revealed that, C/N ratio ranged between 9.2-26.4 in different treatments. Some workers have suggested that changes in the C/N ratio reflect organic matter decomposition and stabilization during composting; therefore, the C/N ratio cannot be used as an absolute indicator of compost maturity (Benito *et al.* 2006). It was suggested that a C/N ratio equal to or less than 20 is a standard for mature compost Hirai *et al.* (1983).

Therefore, the current prepared compost could be considered as good quality compost that can be applied for agricultural purposes.

In the present work the loss in compost weight caused by reduction in OC% led to the increase in total phosphorus (P) and potassium (K) percentages (0.88 and 2.21 respectively), and consequently increase in the percentage of nutrient concentrations. But witting of composted heaps resulted in K% decrease. Reversely, Jamaludin *et al.* (2017) recorded an increase in total phosphorus and potassium in the prepared compost. Also, Lin (2008) showed that the increase in total phosphorus during composting was possibly caused by concentration effect arising from the higher rate of carbon loss that occurs when organic matter is decomposed. Furthermore, Siddiquee *et al.* (2017) found that the highest percentages of phosphorus (P) and potassium (K) (2.13 and 6.68 respectively) were found in compost which inoculated with *Trichoderma* strains.

Generally, the compost prepared in this study can be used for agricultural purposes, because it had a good C/N ratio and high rich in total N, P and K compared with control. These valuable results were also reported by other researchers (Azim *et al.* 2017).

Impact of treatment of compost on plant growth:

Application of the best prepared compost (T4) significantly enhanced different growth parameters of the plant such as: chlorophyll content of leaf, surface leaf area, stem height and fresh and dry weight of shoot and root systems. In agreement with these results, Palanivell *et al.* (2013) found that application of sawdust compost significantly increased maize plant diameter, height, and dry matter production. Composts offer a rich source of residual nutrients, enzymes, xenobiotic degrading microorganisms, and have been employed to treat soils contaminated with a variety of contaminants (Anastasi *et al.* 2008). Thus, the composts may increase timely retention and nutrients release in the soil for plant uptake. This play an important role as a fertilizer by preventing nutrient immobilization and continued supply of nutrients to the plant for better growth and development (Zai *et al.* 2008).

Composts are low in density function as a bulking agent and improve soil structure by loosening it and increase the porosity for aeration

(Celik *et al.* 2010). Also, it allows roots to get oxygen and increase in growth and penetration in soil. Hence, it enables roots to absorb water and nutrient from soil. The significant increase in plant height and growth of maize plants as the result of application the recommended dose of N as a combined form (100% compost +100% mineral N) was supported by the results obtained previously (Hashem *et al.* 2016), and this could be because the compost has a high content of nutrients and biologically active enzymes, as well as hormone-like substances. These substances enhance the root growth and increase the ability of root systems to explore a large volume of soil and consequently increase the amount of nutrients uptaken by the plant (Weber *et al.* 2014). Furthermore, enhancing plant growth in the soil containing a mixture of compost and mineral N reflected with increasing chlorophyll content and photosynthetic capacity of the plant, consequently leading to increased biomass production and (Condon *et al.* 2002; Adamtey *et al.* 2010).

Effect of selected composts on nutrient uptake:

Maize plant grown in soil amended with the prepared compost had greater N, P and K uptake achieving its maximum concentration in T2. Also, Palanivell *et al.* (2013) found that application of sawdust compost significantly increased N, P and K uptake by maize plant. The results of N uptake by maize plant reflected the vital role of organic materials to increase the availability of nutrients in the soil. Addition of organic materials such as corn stalks, soybean straw and plant residues compost in combination with N fertilizer increased the N uptake by wheat plants than using inorganic N fertilizer only (Weber *et al.* 2014).

During the decomposition of the organic constituents of the compost, a lot of soluble organic acids and humic substances are released, which enhance the growth of roots and facilitate the turnover of unavailable P to available form increasing its uptake by the growing plants. Badawi (2003) pointed that the decomposition of organic materials in soil had a positive effect in solubilizing of phosphate by producing organic acids which decrease pH and increase the dissolution of bound forms of phosphate. In addition, some hydroxyl acids may chelate calcium and iron resulting in effective solubilization and utilization of phosphates.

Effect of compost on organic matter and NPK contents in the soil:

Addition of the prepared compost to the soil increased the organic matter and availability of nitrogen, phosphorus, and potassium after harvesting maize plants as observed in the results of soil analysis. Perez-Piqueres *et al.* (2006) reported that soil amendment with compost is an agricultural practice commonly used to improve soil quality and manage organic wastes. Using the agricultural wastes as soil amendments on farmland as alternative fertilizer instead of burning them improved soil physical properties and recycles carbon into the soil. Recycling composted organic residues in agriculture can reduce the need for mineral fertilizers and improve the physicochemical and biological properties of cultivated soils (Aranda *et al.* 2015). Addition of compost to soil can also result in plant disease suppression through the mechanisms of antagonistic action of compost microbiota against plant pathogens (Jovičić-Petrović *et al.* 2016). Gebeyehu and Kibret (2013) showed that high concentration of soil organic matter in the compost treatments contributed to improving the soil's capacity to hold water, that encourages the solubility and availability of nutrient as well as the retention of K by organic colloids (Desoki 2004).

The high amount of total N in soil after plant harvesting could be resulted by many processes taking place during the growth period of plants. Firstly, the decomposition of organic fertilizers has a substantially effect on increasing the amount of N (Mohamed and Hussein 2005). Secondly, the high amount of total N in soil treated with organic fertilizer could be due to the enhancement of the activity of soil microorganisms that fix the atmospheric N (Elbordiny *et al.* 2003). Our results are similar to those obtained by Badran *et al.* (2000) who found that the addition of organic

materials significantly increased the soil moisture retention and availability of phosphorus and potassium. They explained the increase of available P by the production of CO₂ and thus the formation of H₂CO₃ during organic matter decomposition, which lead to phosphate solubility.

Conclusion

Maize plants grown in soil amended with the prepared compost and mineral N showed a significant increase in chlorophyll content, surface leaf area, stem height, fresh and dry weights, as well as NPK uptake. It is assumed that enhancing plant growth may be due to the improving in physical, chemical, and biological properties of the soil after addition of compost. This stimulative effect may be related to good equilibrium of nutrients and water around the root medium or to the beneficial effect of fungi on vital enzymes and hormones that induced the plant growth. Application of such strategy will lead to the reduction of chemical input in the biosphere and establishing the equilibrium in soil characteristic. The current investigation made some valuable contributions: (1) provides a comprehensive knowledge of relative composition of microorganism in composts based on different substrates; (2) provides an overview about recycling of maize stalks using *Aspergillus brasiliensis* and *Penicillium brevicompactum* for production of a valuable and high quality compost; (3) shed light on using prepared compost for promoting plant growth and nutrient uptake; and (4) use of organic fertilizer (maize stalks compost) with mineral fertilizer enhanced plant growth and nutrient uptake, as well as increase organic matter and N/P/K content of soil. Consequently, composting of waste agricultural materials by fungi is recommended for potential use as a low cost organic fertilizer with value-added products.

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