

RESEARCH ARTICLE

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Suppressive effect of compost /pomegranate peel tea combination against *Fusarium oxysporum* f. sp. *lupini*, and *Rhizoctonia solani* as an alternative synthetic fungicide

ABSTRACT:

Plant diseases continue to cause severe damage to most agricultural crops resulting in heavy losses of both yield and quality. In addition, the agricultural wastes cause real problems to get rid of them. Therefore, intensive concern was directed toward compost utilization, as they are available for no cost no side effect. So, in this study different suggested formulations of aerated compost tea in combination with pomegranate peel powder were evaluated for the possibility of using them as an environmentally safe alternative to fungicides. *In vitro* antifungal activity as well as, the *in vivo* probability of these combinations to suppress damping-off and wilt diseases caused by *R. solani* and *F. oxysporum* was done. Pathogens were isolated from diseased lupine plants with root rot and wilt symptoms. *In vitro* experiment results illustrated that high inhibition linear growth was obtained by compost/sour pomegranate tea prepared using alkaline water by up to 60% and 70% for *F. oxysporum* and *R. solani* respectively. Meanwhile, under field condition all tested combinations showed significant reduction in disease severity over un-composted control, indicated by low wilting and high survival % of lupine plants, no significant differences were detected between treatments. All compost treatments, in the field experiment, performed significantly better than the un-composted control in terms of growth parameters, NPK accumulation, yield and its components with no significant differences. Findings of this study suggest that mixture of pomegranate (*Punica granatum* L.) peels extract with compost tea might be a promising combination for controlling phytopathogenic fungi and can be used as effective natural fungicide against two of the most dangerous soil-borne fungi looking for reducing the dependence on the chemical fungicides.

KEY WORDS:

Lupine, *F. oxysporum* *R. solani*, compost tea, pomegranate peel.

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INTRODUCTION:

White lupine (*Lupinus albus* L.), has been cultivated in Egypt for human and animal nutrition, also for medical and industrial purposes, as it can be considered a friendly crop to the environment according to efficiency in nitrogen fixation and its improvement to the traditional cereal rotation and protein supply in low input farming systems. Its seeds contain considerable nutrition due to its high protein (35 - 45%) and oil content (10 - 15%) and are good dietary source of minerals such as phosphorus and potassium (Abd El-Hai *et al.*, 2016). The soil borne fungal diseases such as vascular wilts, root rot and damping-off represent the major causes of decreased yields of agricultural crops and economic losses all over the world, and their control represents a major issue in

the management of cropping systems (Makovitzki *et al.*, 2007). Damping-off, root-rot and wilt diseases still represent the most difficult problems to be managed in the fields and the most important factors limiting seed yield of legume crops (Pal and Garedener, 2006; Farahat *et al.*, 2016). *Fusarium* wilts are widespread diseases affect all agricultural crops as well as legumes; the pathogen colonizes the vascular tissue after root infection causing wilting and finally death of the plant (Rubiales *et al.*, 2015). Damping off disease is one of the most important diseases caused by *R. solani* which may infect a seedling prior to emergence or after emergence (Kucharek, 2000). Damping-off disease of seedlings is reported to affect up to 80% of the seedlings and thereby induce heavy economic losses and once established in the nursery soil, damping-off pathogens are able to survive in the soil for many years, even in the absence of host plants, either as saprophytes or as resting structures that are capable of surviving the adverse conditions (Lamichhane *et al.*, 2017). Overall, the economic losses due to damping-off are represented by a direct cost, due to damages of seed or seedlings, and an indirect cost, which consists of an additional cost of replanting and the consequent lower yields due to the later planting dates (Horst, 2013). In Egypt, Damping-off and root rot diseases are among the most dangerous diseases attacking lupine. Some synthetic substances are compelling in controlling plant infections, yet they are costly and not environmentally friendly, therefore, alternative control methods are needed (Abd El-Hai *et al.*, 2016). Several studies announced the antifungal activity of plant extracts against soil borne pathogens such as *R. solani* and *F. oxysporum* (Abd El-Ghany *et al.*, 2015; Khan *et al.*, 2016; Selim *et al.*, 2016). This clearly suggests that damping-off problem is multifaceted and requires more research efforts to generate further knowledge for a durable and sustainable management of damping-off. Most of these soil-borne pathogens are common in agricultural soils and can be spread *via* non-anthropogenic and anthropic activities, including water run-off through irrigation, air or rain (Zappia *et al.*, 2014).

Chemical Fungicides are effective in controlling such diseases but cause many risks such as water contamination, persistence of residues both in the environment and in the food, accumulation in animal and human fat tissues. In addition, regular and random use of fungicides leads to development of fungicidal resistance variants (Zian *et al.*, 2013). Several studies announced the antifungal activity of plant extracts against soil borne pathogens such as *R. solani* and *F. oxysporum* (Abd El-Ghany *et al.*, 2015; Khan

et al., 2016). Pomegranate (*Punica granatum* L.) is reported to have strong antibacterial, antifungal, antiprotozoal and antioxidant properties (Shafiq *et al.*, 2013; Hamouda *et al.*, 2015). About half of the total pomegranate fruit weight corresponds to the peel, which is an important source of bioactive compounds such as tannins, flavonoids, polyphenols and anthocyanins (Tholkappiyan *et al.*, 2011; Kumar and Vijayalakshmi, 2013). Besides, the pomegranate fruits peel is rich with the most nutritious minerals mainly P, K, Na, Fe, Zn, Mn, and Cu (Rowayshed *et al.*, 2013).

The fruit processing industries generate considerable amount of solid wastes in the form of peels and seeds, and these wastes if not disposed correctly are seen to cause serious environmental problems such as water pollution, unpleasant odors, and greenhouse gas emissions (Roy and Lingampeta, 2014). Composting is a perfect recycling technology for organic wastes through which microorganisms convert them into humus like substance called compost which is very rich of plant nutrients with beneficial effects on physical, chemical, and biological properties of the soils (Pane *et al.*, 2013). Numerous studies showed that incidence of several soil-borne plant pathogens have been reduced by using composts made of different raw materials (Zaccardelli *et al.*, 2013). Additionally, compost-based treatments can stimulate an enhanced plant physiological status with improvements in quantity and quality of crop productions (Liguori *et al.*, 2015). El-said *et al.* (2018) found that the mixed treatments of agricultural residues (aerated compost tea and olive pomace) with promoting microorganisms were significantly superior against *R. solani* and *F. oxysporum*. Compost tea has been evaluated as fungicides alternative to control a variety of plant pathogens according to Islam *et al.* (2019) and may be one of promised applications of compost which is made by infusion of compost in water for a period, then compost tea is obtained by filtration (Islam *et al.*, 2019). Significant reduction of the linear growth of the tested fungi (*Fusarium solani*, *Macrophomina phaseolina* and *Rhizoctonia solani*) was observed by Attia (2019) when treated with compost tea. Nowadays, biological control, compost and many natural waste extracts has become an important ecological and safer alternative to chemicals for treating and managing many soil-borne diseases. *In vitro* and *in vivo* study were performed to assess the potential role of combined enriched compost and compost tea with pomegranate peel powder in management of *R. solani* and *F. oxysporum*, the main cause fungal pathogens of lupine damping-off and wilt diseases.

MATERIAL AND METHODS:

Pomegranate peels:

Three distinct kinds of commercial pomegranate including two Palestinian (sweet and sour) and one Egyptian (sweet) were utilized in this investigation. Palestinian pomegranate fruits were gathered from commercial markets in Gaza Strip while the Egyptian type was gathered from commercial markets in Cairo. Fresh fruits of pomegranate (*Punica granatum L.*) were physically peeled. Peels were cut into smaller pieces and washed with distilled water. The peel pieces were air dried enough and then oven dried at $50 \pm 2^\circ\text{C}$ for 3 - 4 days, and ground using a mechanical grinder.

Compost:

Mature compost was provided from Ismailia Agriculture Research Station, Ismailia Governorate, Egypt. Compost was prepared from rice straw, farmyard manure, bentonite, rock phosphate, feldspar, urea, and elemental sulfur and vinasse solution according to Abdel-Wahab *et al.* (2008).

Organisms used:

a- *Rhizoctonia solani* Kühn and *Fusarium oxysporum* f. sp. *lupini* were provided by Legume and Forage Diseases Research Department, Plant Pathology Research Institute, ARC, Giza, Egypt, which were isolated from diseased lupine plants with root rot and wilt symptoms. The fungi were regularly sub-cultured and maintained on Potato Dextrose Agar (PDA) medium at $5 \pm 1^\circ\text{C}$ until used.

b- *Rhizobium (Bradyrhizobium lupini)*, ARC 408) inoculum was provided by Biofertilizer Production Unit. Soils, Water and Environment Res. Inst., Agric. Res. Center, Giza, Egypt.

Lupine seeds:

White lupin (*Lupinus albus L.*) seeds cultivar (Giza 2) was obtained from Legume Crop Res. Dep., Field Crop Res. Inst., Agric. Res. Center, Giza, Egypt.

Preparation of aqueous pomegranate peel extract:

Extraction was done by soaking 10 g of each pomegranate powder in 100 mL water for 24h and filtered through filter paper Whatman No.1. Filtrate was dried in hot air oven at $45 - 50^\circ\text{C}$ and stored in dark glass bottle at 4°C for further studies (Selvamohan *et al.*, 2012).

Preparation of aerated compost tea:

Compost tea was prepared according to Ingham (2003). The following ingredients (g) were added to one-liter tap water:

Mature compost, 143 (compost to water ratio was 1:7); $(\text{NH}_4)_2\text{SO}_4$, 4.5; KH_2PO_4 , 0.5; $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, 0.25; NaCl, 0.1; sucrose, 10

and $\text{MnSO}_4 \cdot \text{H}_2\text{O}$, 0.2. Air was bubbled through the mixture using aquarium pump for five days. Then the liquid mixture was filtered through a 100-mesh screen to remove solid particles. Alkaline compost tea was prepared by adding the previous ingredients to alkaline water (0.1M KOH/L). Potassium hydroxide has been used to obtain humic extract from compost, and it produces high humic acid yield and contains high amount of K, so it is the preferred choice as an extracting agent (Charest *et al.*, 2005).

Preparation of aerated pomegranate peels/compost tea:

Pomegranate peel/compost tea was prepared as described previously by adding the pomegranate peel powder (each of the three tested types) to compost at ratio 5: 100. Alkaline pomegranate peel/compost tea was also prepared.

Inhibition activity of aerated compost tea and pomegranate peel/compost tea:

The antifungal effect was studied *in vitro* by poisoned food technique (Agarwal *et al.*, 2001). Ten mL of each of the prepared teas was added to 100ml sterilized PDA medium and shacked carefully. The medium was then poured into 9 cm diameter Petri dishes (three replicates), and untreated plates were used as negative control. Five mm diameter disc of the fungus (one-week-old culture) was placed at the center of each of the poured plates. Plates were incubated at $25 \pm 2^\circ\text{C}$ until the fungal growth in the control dishes was completed. Plates containing the fungicide Rizolex-T were used as positive control. Percentage of inhibition was calculated according to Whitehead (1957) due to the treatments against control using the following formula

$$\text{Inhibition} = (\text{C} - \text{T} / \text{C}) \times 100$$

Where **C** is hyphal extension (mm) in control plates

T is hyphal extension (mm) in treated plates

Field experimental design and measured parameters:

Under natural conditions, the experiment treatments were carried out at Agric. Res. Centre, Giza, Egypt, during two successive winter seasons to evaluate the bio-suppression of lupine damping-off and wilt diseases by seed treatment and soil application of enriched compost and compost tea with pomegranate peel powder, pomegranate type will be chosen depending on the results of the previous laboratory experiment. The treatments used were as follow: **a-**compost powder which is mixed with soil at the rate of 4000 Kg/fed. **b-** compost with the selected pomegranate peel powder was added at the rate of 4000 and 200 Kg/fed, respectively. **c-** aqueous extract of the selected pomegranate peel powder **d-** alkaline

compost tea. e- selected pomegranate/ compost tea . The last three treatments were applied as soil drench at rate of 75L/fed three times (one at sowing and two at 20 days intervals). f- finally fungicide treatment was carried out to the seeds by applying commercial Rizolex-T at the recommended dose (positive control). Treatments were arranged in a complete randomized block design (three replicates). The field plot was 5.4 m² with four rows, in each plot two hundred lupine seeds were sown under normal field conditions and naturally infested soil with the casual organisms of damping-off and root rot of lupine, and untreated plot was left as negative control. *Rhizobium* inoculum was added at sowing to the experimental soil at rate of 600 g inoculum/fed. All plots received the recommended dose of superphosphate at the rate of 100 kg/fed and potassium sulphate at the rate of 50 kg/fed. Nitrogen fertilizer in the form ammonium sulphate was used at the rate of 15 kg/fed as activation dose, and the agricultural practices were done as recommended. Disease incidence percentage of pre, post-damping-off, wilt and survived plants was calculated 15, 45, 90 and 120 days of sowing. After 75 days from sowing, plants were randomly uprooted from each plot to evaluate the following morphological growth parameters (plant height, shoot dry weight/plant, root dry weight/plant), physiological parameters (chlorophyll content of leaves, nitrogen, phosphorus and potassium content) and nodulation status (number of nodules/plant, dry weight of nodules per plant and nitrogenase activity). At harvest (150 days from sowing), the following morphological and physiological measurements were evaluated: plant height, root length, number of pods/plants, number of seeds/plant, seed yield/plant, hundred-seed weight, and crude protein content of seeds (%). The biological weight of each plot was reported to determine seed yield (Kg/fed.) and straw yield (Kg/fed.).

Pre – Emergence% = (Total No. of ungerminated seeds / Total No. of planted seeds) × 100

Post – Emergence% = (Total No. of dead seedlings / Total No. of planted seeds) × 100

Wilt planted % = (Number of wilted planted / Total No. of planted seeds) × 100

Survived seedlings % = (Number of survived seedlings / Total No. of planted seeds) × 100

Analytical methods:

All determinations of compost, compost tea and pomegranate peel/compost tea analysis were done at Soils, Water and Environmental Res. Inst. Agric. Res. Center, Giza, Egypt.

Digestion of plant samples were performed according to Jackson (1973). Shoot and root nitrogen content was determined in

digested solution using Macro-Kjeldahl method, phosphorus determined using stannous chloride reagent by spectrophotometer at 640 nm and potassium using flame photometer according to Page *et al.* (1982).

Chlorophyll content in fresh leaf samples were extracted by 80% methanol after grinding. The sample was filtered through filter paper. The absorption of constant volume of filtered was measured at optical density 650 and 665 (Arnon, 1949). Total chlorophyll (mg/g fresh leaves) = 25.5 OD 650 + 4.0 OD 665

The nitrogenase activity of mature roots nodules was estimated using the reduction assay method according to Hardy *et al.* (1973).

Crude protein of seeds was calculated as % nitrogen multiplied by 6.25 (Ranganna, 1977).

Statistical analysis:

Differences in treatments were tested using one-way analysis of variance (ANOVA 1) according to SPSS software (SPSS, 2006).

RESULTS AND DISCUSSION:

Analytical characterization of compost and compost teas:

Assessment of physical (water holding capacity), chemical (electrical conductivity, pH, heavy metals and potentially toxic elements) and biological (absence of pathogens and presence of promoting organisms) is an important requirement for compost selection (Brinton, 2000). Many researchers accepted different compost substitutions based on many factors as germination rate and indices with some limitations (Bustamante *et al.*, 2008; Ostos *et al.*, 2008). It is clear from data present in table 1 that preparation of tea using alkaline water is more benefit since higher microbial counts over those prepared using water were obtained. This was likewise observed for all chemical measurements determined. Additionally, higher considerable levels of the measured chemical parameters were identified for the blend of compost with pomegranate peel tea prepared using alkaline water compared to compost or compost tea. Interestingly, the lower germination index % was recorded for compost powder which indicates higher phytotoxic compounds and further confirmed that compost tea suitable for application (Kuo *et al.*, 2004). Compost is very rich with humic substances that outcome by microbial disintegration of organic wastes under controlled conditions (Ahmad *et al.*, 2007). The low E₄/E₆ ratio than 5 indicate to some extent high degree of condensation of aromatic humic content according to Pare *et al.* (1997), interestingly, the present data showed high level of humification in the

prepared teas. It was reported that humic substances have molecular characteristics that may bring about higher biological activity due to enzymatic activation of nutrient uptake or modification of bacterial cell permeability to

nutrients (Tejada *et al.*, 2006). This may clarify the higher count of bacteria, fungi and actinomycetes in compost teas prepared using alkaline water than those of compost teas prepared using water.

Table 1. Physico-chemical and biological characterization of compost, compost tea and pomegranate peel/ compost tea

Measured parameter	Compost type				
	Compost	Compost tea		Sour pomegranate / Compost tea	
		-	Water	Alkaline water	Water
pH	6.79	7.43	8.57	6.41	7.75
EC (ds/m)	5.53	6.63	8.22	6.31	8.62
Organic carbon (%)	23.60	4.4	5.3	6.33	7.1
Organic matter (%)	40.59	8.3	9.8	8.8	8.2
Total N (%)	1.42	0.9	1.30	0.7	0.9
NH ₄ (ppm)	441.0	469	587	496	661
NO ₃ (ppm)	371.0	30	42	30	41
Available P (ppm)	311.0	40	54	60	78
Available K (ppm)	613.5	257	400	320	460
E ₄ /E ₆	2.54	1.547	2.038	1.924	2.253
Germination index for cress (%)	79.5	90	93	91	94
Dehydrogenase activity (mg *TPF/100g or ml / 24hr)	89.8	93.548	100.324	95.838	118.614
Total count of bacteria (CFU/g or mL)	3.2 × 10 ⁷	1.8 × 10 ⁷	7 × 10 ⁷	1.7 × 10 ⁷	12 × 10 ⁷
Total count of fungi (CFU/g or mL)	2.1 × 10 ⁶	1.5 × 10 ⁶	4.3 × 10 ⁶	1.3 × 10 ⁶	3.1 × 10 ⁶
Total count of actinomycetes (CFU/g or mL)	1.5 × 10 ⁶	1.2 × 10 ⁶	4 × 10 ⁶	0.8 × 10 ⁶	5 × 10 ⁶

*Tri-Phenyl-Formazan.

In vitro screening for the inhibitory effects of aerated compost tea and aerated different pomegranate peel / compost tea on linear growth of *F. oxysporum* and *R. solani* was done to be used as predictors of disease suppression for the further *in vivo* (field) experiment. Clearly, the compost tea and the suggested formulation of pomegranate compost teas prepared using water and or alkaline water possess positive effect on the

studied fungi and inhibited the linear growth with significant different degrees (Fig. 1). In general, prepared compost tea using alkaline water (KOH) strongly inhibiting mycelium development of the two tested fungi compared with compost prepared with water. The most effective combination was recorded for the mixture of compost with the sour pomegranate prepared by alkaline water (Figs 2 & 3).

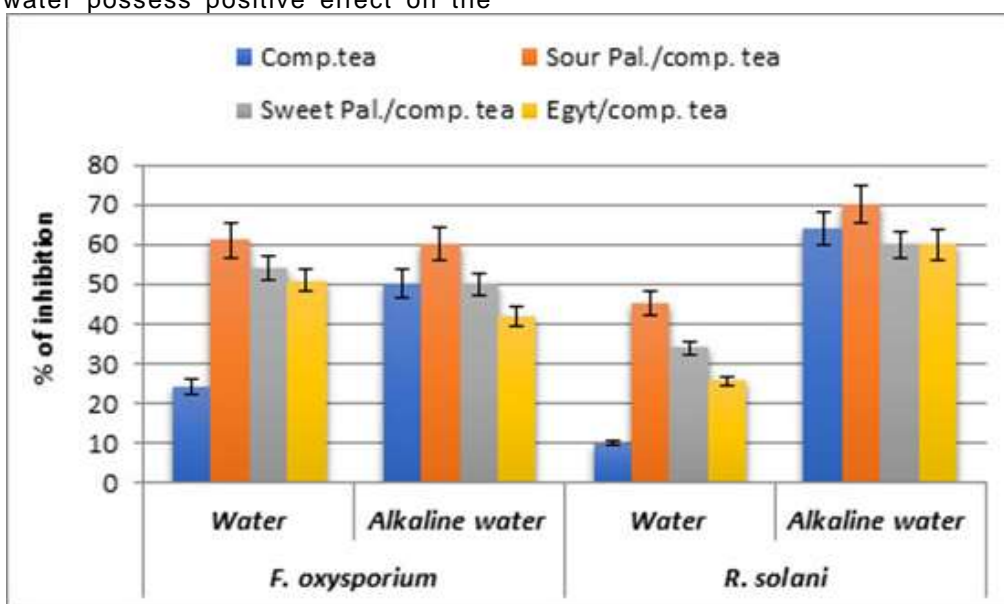


Fig. 1. Effect of different compost teas on linear growth of *F. oxysporium* and *R. solani*

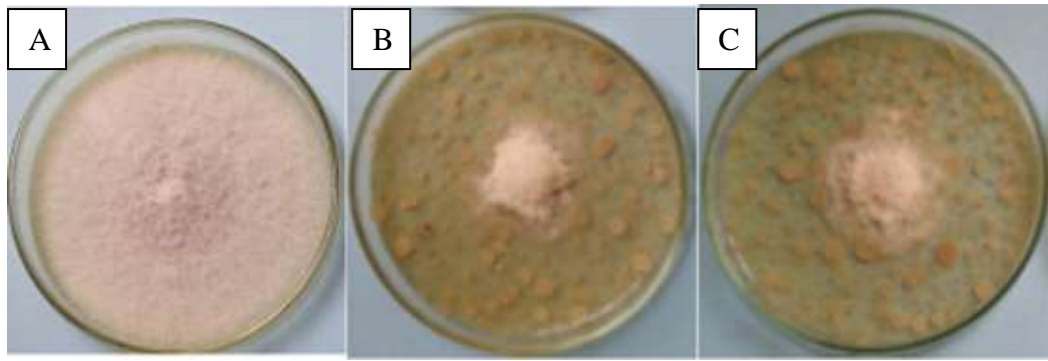


Fig. 2. Effect of sour pomegranate/compost tea on *F. oxysporium*: A control, B with alkaline water, C with water.

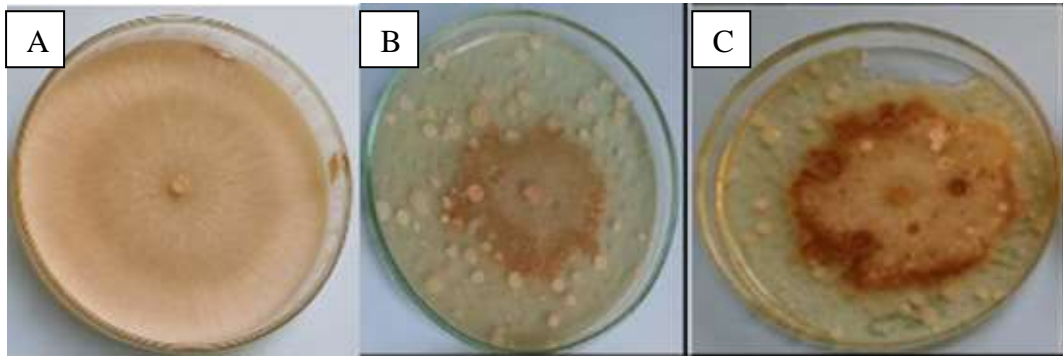


Fig. 3. Effect of sour pomegranate/compost tea on *R. solani*: A control, B with alkaline water, C with water.

High suppressive effect of this combination indicated by inhibition fungus mycelium development reached 60 % against *F. oxysporum* and 70 % against *R. solani*. In contrast, very weak reduction in linear growth of the studied fungi was recorded for compost tea with water as a single treatment against *F. oxysporum* (24.3%) and *R. solani* (10%). As shown from the obtained results that mixture of aerated compost with sour pomegranate prepared using alkaline water resulted in inhibition growth higher than 50% for *F. oxysporum* f. sp. *lupini* and *R. solani* which was considered the reference value for determining a high suppressor effect (Bernal-Vicente *et al.*, 2008). While many of the prepared compost with water did not reach a suppressive effect of 50%. El-saiid *et al.* (2018) recorded that the mixed treatment of agricultural compost tea and olive pomace was significantly superior over the individual treatments and showed 63.52% and 44.07% percent inhibition against *R. solani* and *F. oxysporum* respectively. Results are also in good accordance with recent studies demonstrated significant reduction the linear growth of different soil borne pathogenic fungi (*Fusarium solani*, *Macrophomina phaseolina* and *Rhizoctonia solani*, *Fusarium oxysporum*) (Attia, 2019; Islam *et al.*, 2019).

Potassium hydroxide has been used to obtain humic extract from composts, and it is the preferred choice as an extracting agent because it produces high humic acid yield and contains high amount of K (Charest *et al.*, 2005). Concerning the effect of additive botanical powder to compost tea, these additives might promote or affect the growth

of bacteria, furthermore the enhancement of the blended formulation may be attributed to the nutritive value of pomegranate peel which improved the quality of compost tea as concluded by Rowayshed *et al.* (2013). Pomegranate fruits peel powder is rich with the most nutritious minerals and considered a good source of macro and micro elements. In addition, pomegranate peel is a rich source of many bioactive substances as hydrolysable tannins, polyphenols specifically gallic acid and flavonoids (Faria and Calhau, 2010; Glazer *et al.*, 2012). Moreover, physicochemical properties of the compost teas, namely plant nutrients and organic molecules such as humic or phenolic compounds may protect the plant against disease through improved nutritional status, direct toxicity toward the pathogen and/or induced systemic resistance (Dionne *et al.*, 2012).

Compost tea is a fermented extract of composted substances to obtain a watery extract with or without active aeration (Ingham, 2003). Aerated compost teas are actively fermented and supplying with microbial food, catalysts and air bubbles pumped in the solution providing plenty of much-needed oxygen (Litterick and Wood, 2009). During brewing process of compost tea, increasing dissolution of nutrients is predicted due to the agitation caused by aeration, and then according to Ingham (2003), aerated composts are more efficient against diseases and phytopathogens because they tend to have higher microbial populations and diversity. According to the illustrated results alkaline Palestinian sour

pomegranate /compost formulation is the most effective one against the two studied soil borne pathogens, therefore it was used for management these fungi in a field experiment.

Field evaluation of compost and compost tea formulations against damping-off and wilt disease:

- Disease assessment:

The most important factors limiting the yield production of lupine are soil-borne fungal diseases resulting in severe economic losses. Among the most destructive pathogens are damping-off *Fusarium oxysporum* f. sp. *lupine* and *Rhizoctonia* root-rot (*Rhizoctonia solani* Kühn). To obtain high yields production and protect the environment, replacement of chemical fertilizers has always received increasing attention. Therefore, evaluation of compost amendments for potential control of damping-off and wilt diseases of lupine plants in a field trial naturally infested was performed. The disease-suppressive effect was assessed in terms of the following parameters:

Damping-off and wilt diseases:

Disease incidence (% pre-, post-damping-off, wilt and survived plants) were

calculated 15, 45, 90, and 120 days of sowing, respectively. All compost amendments and chemical treatment provided significant improvement in seedling emergence (pre- and post-) damping off lupine plants over the un-composted control (Table 2). The highest reduction in wilt disease incidence was obtained with compost/pomegranate tea, aqueous extract of pomegranate tea and Rhizolex-T fungicide with no significant differences which is clearly illustrated by the lower percent of wilted plants and higher percent of survived plants compared with high percent of wilted plants (11.67%) and low percent of survived plants (62%) for untreated control. In addition, results illustrated that the tested composting formulations as well as the chemical fungicide significantly increased the lupine seeds germination over untreated control. Remarkable reduction in percentage of pre- and post-emergence damping-off ranging from 6.33 to 1.33% for pre- and from 2 to 0.33% for post- emergence over untreated control. Finally, all treatments showed high significant level of disease suppression in comparison to control plots indicated by high percent of survived lupine plants.

Table 2. Influence of different compost formulations on incidence of damping-off and root-rot diseases

Treatment	Measurement			
	Damping-off (%)		Dead plants (%)	Survived plants (%)
	Pre-emergence	Post-emergence		
Untreated soil (Control)	12.00 ± 2.00 ^a	14.33 ± 0.58 ^a	11.67 ± 2.52 ^a	62.00 ± 2.00 ^d
Compost	6.00 ± 1.00 ^b	2.00 ± 1.00 ^b	7.67 ± 1.15 ^b	84.33 ± 1.53 ^c
Compost with sour pomegranate peels	6.33 ± 1.53 ^b	1.67 ± 1.53 ^b	6.00 ± 1.00 ^b	86.00 ± 1.00 ^{bc}
Compost tea(alkaline water)	2.33 ± 1.15 ^c	2.33 ± 1.53 ^b	7.00 ± 1.00 ^b	90.33 ± 3.21 ^{ab}
Compost/sour pomegranate tea (alkaline water)	4.00 ± 2.65 ^b	0.67 ± 0.58 ^b	4.33 ± 1.53 ^b	91.00 ± 1.00 ^a
Aqueous extract of sour pomegranate peels	4.00 ± 1.00 ^b	2.00 ± 1.00 ^b	4.00 ± 3.00 ^b	90.00 ± 1.73 ^{ab}
Rizolex-T	5.33 ± 2.08 ^b	0.33 ± 0.58 ^b	4.33 ± 3.21 ^b	90.00 ± 4.58 ^{ab}
F value	10.84 ^{***}	67.25 ^{***}	4.93 ^{**}	52.88 ^{***}

Means followed by the same letter are not significantly different at * P< 0.05 ** P< 0.01 *** P< 0.001.

These results are proved by many investigators who reported that incidence of several soil-borne plant pathogens have been reduced by using composts made of different raw materials. For instance, the obtained results by Mokhtar and El-Mougy (2014) indicated that under field conditions, compost reduced root rot incidence disease of bean caused by *F. solani* and *R. solani*. In other work, El-Mohamedy *et al.* (2015) recorded that drenching soil with different bio composts prepared from sugarcane bagasse, rice straw and soybean straw and inoculated with isolates of *Trichoderma* before sowing potato tuber significantly reduce *Fusarium* dry-rot and *Rhizoctonia* black scurf diseases during two cultivation seasons. It was also suggested

that this reduction could be attributed to the direct effect of such organic materials or bioactive compounds to suppress (or to kill) the pathogen population indirectly enhance both the microbial population and activity in rhizosphere of the affected plants (El-Gizawy, 2005; Scheuerell *et al.*, 2005).

Although there is a significant effect for the prepared compost formulations on the severity of lupine plants damping-off and wilt diseases compared to untreated control under field conditions, but no compost type was a significant factor affecting microbial and chemical characteristics of compost teas which is not consistent to the present results.

2-Growth parameters:

Measurement of growth features of plants may represent a good indication about the different effect of various treatments tested. Effect of different formulated compost teas on plant height, shoot and root dry weight and leaf chlorophyll content of lupine plants after 75 days of planting are illustrated in table 3. Clearly all growth measures of the untreated control recorded low values with significant difference over measures of treated plants; such reduction in the measured values confirms the adverse effect of soil borne pathogens on plant health and survival. Results also illustrated higher values of all measured parameters for treated plants with no significant difference between types of composting used. Importantly, plants treated with the chemical fungicide exhibited nearly the same values of plant height, shoot and root dry weight and chlorophyll content. In accordance, Scheuerell and Mahaffee (2002) proved that compost teas, with their rich biochemical composition have a considerable role in controlling much fungal disease. Furthermore, El-Shinnawi *et al.* (2011) illustrated

that application of composts or their teas as soil manuring resulting in constant suppression of damping-off disease of peanut plants caused by *Rhizoctonia solani* and increased health and survival, as compared with the un-composted control. El-Masry *et al.* (2002) proposed that composts and compost tea may control soil borne pathogens by production of different bioactive inhibitory compounds, hydrolytic enzymes or through nutrients competition. Further explanation by Kone *et al.* (2010) illustrated that the nutritional elements of different compost tea especially the organic molecules with specific chemical characters as humic or phenolic compounds may play a role to improved growth status of the plants which protect them against diseases or exert direct toxicity toward pathogen. El Shimi *et al.* (2015) recorded significant high increase in vegetative growth parameters (plant length, number of leaves, leaf area, fresh and dry weight and leaf and chlorophyll content) of sweet pepper plants when soil amended with formulated compost.

Table 3. Growth parameters of lupine plants treated with compost formulations in field experiment (75 days)

Treatment	Measurement			
	Plant height (cm)	Shoot dry wt. (g/plant)	Root dry wt. (g/plant)	Chlorophyll content (mg/g)
Untreated soil (Control)	53.00 ± 2.00 ^c	6.50 ± 0.75 ^c	1.53 ± 0.22 ^c	2.50 ± 0.10 ^b
Compost	63.00 ± 2.65 ^{ab}	7.62 ± 0.18 ^b	2.18 ± 0.03 ^b	2.83 ± 0.06 ^a
Compost with sour pomegranate peels	65.33 ± 2.52 ^{ab}	8.39 ± 0.34 ^b	2.28 ± 0.14 ^{ab}	2.93 ± 0.32 ^a
Compost tea(alkaline water)	67.33 ± 2.52 ^{ab}	9.62 ± 0.62 ^a	2.37 ± 0.17 ^{ab}	2.90 ± 0.10 ^a
Compost/ sour pomegranate tea (alkaline water)	69.00 ± 2.00 ^a	10.25 ± 0.72 ^a	2.51 ± 0.11 ^a	3.00 ± 0.10 ^a
Aqueous extract of sour pomegranate peels	67.00 ± 2.00 ^{ab}	8.54 ± 0.20 ^b	2.32 ± 0.09 ^{ab}	2.77 ± 0.06 ^a
Rizolex-T	66.00 ± 3.00 ^{ab}	9.50 ± 0.50 ^a	2.37 ± 0.12 ^{ab}	2.80 ± 0.10 ^a
F value	14.79 ^{***}	18.47 ^{***}	16.34 ^{***}	3.67 [*]

Means followed by the same letter are not significantly different at * P < 0.05 ** P < 0.01 *** P < 0.001.

Nodulation status:

Table 4 illustrates the effect of the investigated compost and different compost tea formulations on nodulation status of lupine plants after 75 days of planting. Results clearly showed that under natural infested soil conditions, fungal infection remarkably decreased nodulation capacity of the roots for more than one half of each nodule number, dry weight and nitrogenase activity values. It was suggested that reduction of nodule formation is due to the pathogenic fungus invasion, which shortened the energy transported to the roots under disease intensity and impair the root system proliferation (Hassanein *et al.*, 2006; El-Sayed, 2007). Importantly, unmistakable impact of the aerated compost/sour pomegranate tea on nodulation status over untreated control or even chemical fungicide treatment is observed. Percentage increase of nodules number, nodules dry weight and nitrogenase activity were 123.86, 377.26 and ISSN: 1687-7497

664.04% over un-composted control. On the other hand, the lowest significant improvement in lupine nodulation status was observed for plots treated with solid compost alone which exceed only of about 28.57, 114.6, and 245.86% for nodules number, dry weight and nitrogenase activity over the untreated control. According to El-Shinnawi *et al.* (2011) this could be attributed to a suppressive biocontrol action of the efficiency of compost and their teas for stimulation of plant growth as a result of nutritional support and presence of growth-promoting substances. The present findings are in accordance with Malik and Sindhu (2011) who demonstrated that organic materials produced a suitable habitat for legume growth and enhancing the performance of the biological nitrogen fixation process.

Table 4. Nodulation status of lupine plants treated with compost and compost teas formulations in field experiment (75 days)

Treatment	Measurement		
	No. of nodules/plant	Dry wt. of nodules (mg/plant)	Nitrogenase activity (µmole/pl./hr.)
Untreated soil (Control)	7.00 ± 1.00 ^d	102.67 ± 13.50 ^e	2.42 ± 0.32 ^f
Compost	9.00 ± 1.00 ^{cd}	220.33 ± 16.50 ^d	8.37 ± 0.17 ^e
Compost with sour pomegranate peels	10.33 ± 1.53 ^{bc}	249.00 ± 8.00 ^{cd}	8.82 ± 0.15 ^{de}
Compost tea(alkaline water)	11.67 ± 1.53 ^b	257.67 ± 13.50 ^c	10.78 ± 0.50 ^c
Compost/ sour pomegranate tea (alkaline water)	15.67 ± 1.53 ^a	490.00 ± 29.00 ^a	18.49 ± 0.21 ^a
Aqueous extract of sour pomegranate peels	12.00 ± 1.00 ^b	389.00 ± 24.00 ^b	9.22 ± 0.23 ^d
Rizolex-T	12.67 ± 1.53 ^b	413.00 ± 11.00 ^b	14.88 ± 0.28 ^b
F value	13.11 ^{***}	166.33 ^{***}	952.74 ^{***}

Means followed by the same letter are not significantly different at * P < 0.05 ** P < 0.01 *** P < 0.001.

Effect of copost preparations on NPK contents in lupine plant tissues.

Nitrogen, phosphorus and potassium accumulation in root and shoot of lupine plant tissues as a result of the application of different compost treatments under study followed a similar trend (Table 5). application of solid compost alone or with pomegranate peels powder and their extracts significantly elevating the shoot and root NPK accumulation of lupine plants compared to untreated control, similar trend was obtained by El-Sayed (2007). Clearly, compost with sour pomegranate tea recorded the highest level of NPK existence. This was clarified by the ability of organic substances (humic and other various organic acids) and biological agents to improve the availability and uptake of macro-elements, beside the helpful role in the biological nitrogen fixation process (Tilak *et al.*, 2005; El-Tahlawy, 2006). Fuente *et al.* (2006) showed that organic fertilizers and soil enhancers are used for their organic matter contribution and nutrients. Ayoola and Makinde (2009) illustrated that application of organic fertilizer has positive effects in preserving the organic matters to soil; the

organic matters liberate the nutrient elements through mineralization process that can be used to assist in plant growth. The nutrients from organic fertilizers help rapid root development (Baldi *et al.*, 2010). Results illustrated by El-Shinnawi *et al.* (2011) revealed that artificial infection of the sandy soil with *Rhizoctonia solani* caused a significant depression in N, P and K contents of the peanut tissues, while use of composts or their tea resulting in significant increase of such nutrients accumulation. It was explained by El-Shinnawi *et al.* (2011) that the decrease in N content in peanut tissues indicated that the nitrogen fixation process was somewhat weakened as effected by the fungal infection. Furthermore, under fungal infection reducing of the uptake of both phosphorus and potassium occur as a result of root system damage. It was revealed by Haggag *et al.* (2014) that application of compost tea as soil drench with or without mineral NPK gave the best results for increasing nitrogen and potassium contents in leaf seedlings of olive compared to control treatments without using mineral NPK or compost tea (Table 5).

Table 5. NPK accumulation in shoot and root of lupine plants treated with compost and compost teas formulations in field experiment (75 days)

Treatment	Measurement					
	Shoot (mg/plant)			Root (mg/plant)		
	N content	P content	K content	N content	P content	K content
Untreated soil (Control)	131.93 ± 9.38 ^f	15.31 ± 0.83 ^f	75.36 ± 4.92 ^d	8.86 ± 0.16 ^e	0.65 ± 0.05 ^e	9.64 ± 0.69 ^d
Compost	175.11 ± 8.00 ^e	19.03 ± 0.30 ^e	112.45 ± 1.58 ^c	15.13 ± 0.78 ^d	1.67 ± 0.05 ^d	15.71 ± 0.38 ^c
Compost with sour pomegranate peels	206.36 ± 7.68 ^d	21.79 ± 0.05 ^d	115.16 ± 2.83 ^c	15.71 ± 0.50 ^{cd}	1.82 ± 0.07 ^{cd}	16.16 ± 0.30 ^{bc}
Compost tea (alkaline water)	292.89 ± 16.37 ^b	31.35 ± 0.65 ^b	133.17 ± 4.23 ^b	16.31 ± 0.71 ^c	1.94 ± 0.07 ^c	18.12 ± 0.73 ^a
Compost/sour pomegranate tea (alkaline water)	358.30 ± 23.28 ^a	32.75 ± 1.27 ^a	142.05 ± 8.34 ^a	18.57 ± 0.74 ^a	3.26 ± 0.10 ^a	18.56 ± 0.35 ^a
Aqueous extract of sour pomegranate peels	244.06 ± 16.25 ^c	28.74 ± 0.34 ^c	115.27 ± 3.14 ^c	16.35 ± 0.30 ^c	2.20 ± 0.13 ^b	15.98 ± 0.60 ^{bc}
Rizolex-T	245.11 ± 13.33 ^c	28.47 ± 0.72 ^c	137.30 ± 3.84 ^{ab}	17.51 ± 0.44 ^b	2.27 ± 0.18 ^b	16.81 ± 0.61 ^b
F value	80.76 ^{***}	269.36 ^{***}	72.46 ^{**}	93.65 ^{***}	177.56 ^{***}	87.77 ^{***}

Means followed by the same letter are not significantly different at * P < 0.05 ** P < 0.01 *** P < 0.001.

Effect of compost preparations on lupine yield and its components:

Regarding lupine plant yield and its major components, they showed a trend comparable to that above mentioned for the growth features and nodulation. Joshi *et al.* (2009) suggested that this is an outcome of the beneficial action accumulation of the different forms of compost additions. Management the damping-off and root rot diseases to improve yield component aim to maximize the total seed yield of grain legume crops. Result shown in table 6 illustrate that addition of any composted formulation had a distinct positive effect on the harvested lupine yield and its components over the uncomposted treatment. The highly effective compost treatments which exhibited nearly the same positive effects with a distinguishable impact over the other treatments were compost sour pomegranate tea and the chemical fungicide Rexolex-T with no significant differences over each other. The effective treatment of compost /sour pomegranate tea recorded increase percentages reached 86.66, 15.14, 22.21, and 39.75% higher than untreated control treatment for number of pods/plant, 100 seed weight, seed protein content and seed yield in respective order. The seed yield of legumes is

the result of different plant growth processes, which are in the end expressed in the yield components of pods /plant, seeds /pod, mean seed weight and seed yield. The highest seed yields are generally obtained when all observable characters are maximized (Ayaz, 2001). Several researchers have been recorded that bio compost application as soil amendment could suppress diseases caused by *R. solani* and *Fusarium spp.* on many economic crops (Godwin-Egein and Arinze, 2000; El-Mohamdy, 2004). Abdel-wahab *et al.* (2006) showed that the application of rhizobacteria and compost treatment result in significant increases in peanut yield and its components under field experiment. The positive effect of compost tea on plant growth and yield could be attributed to the presence of beneficial rhizobacteria, or their metabolites which enhanced plant growth by their ability to stimulate nodulation ability and increasing the N₂-fixation performance, absorption of nutrients, and metabolism of photosynthates and improve the productivity and quality of many legumes. Finally, it is of interesting to mention that even there is no significant difference between the suggested compost formulations, but importantly, no significant difference also was detected when compared with the recommended chemical fungicide.

Table 6. Effect of compost and their teas on lupine yield and its component

Treatment	Measurement						
	No. of pods/pl.	Seed weight /pl. (g)	No. of seeds/pl.	100 seed wt. (g)	Seed protein content (%)	Seed yield (kg/fed)	Straw yield (kg/fed)
Untreated soil (Control)	15.0 ± 1.00 ^c	23.27 ± 0.35 ^d	57.0 ± 1.0 ^d	40.82 ± 0.11 ^c	34.63 ± 0.07 ^d	1164.1 ± 43.95 ^d	2338.8 ± 150.90 ^d
Compost	20.0 ± 2.00 ^b	25.30 ± 1.93 ^{cd}	61.0 ± 2.70 ^{cd}	41.44 ± 1.43 ^c	36.44 ± 0.75 ^c	1225.5 ± 26.80 ^{cd}	2737.8 ± 134.55 ^c
Compost with sour pomegranate peels	22.0 ± 1.70 ^b	27.30 ± 1.30 ^c	65.0 ± 2.00 ^c	41.99 ± 0.71 ^c	38.79 ± 0.16 ^b	1260.8 ± 45.90 ^c	2883.7 ± 192.45 ^c
Compost tea (alkal)	27.0 ± 1.70 ^a	37.90 ± 2.50 ^b	81.0 ± 4.00 ^b	46.76 ± 0.78 ^{ab}	41.89 ± 1.10 ^a	1392.7 ± 60.30 ^b	3307.9 ± 153.20 ^b
Compost/sour pomegranate tea	28.0 ± 1.70 ^a	43.20 ± 2.00 ^a	90.0 ± 3.00 ^a	47.99 ± 0.62 ^a	42.32 ± 0.43 ^a	1626.8 ± 58.70 ^a	3783.6 ± 197.20 ^a
Aqueous extract of sour pomegranate peels	26.0 ± 1.00 ^a	37.63 ± 0.81 ^b	82.0 ± 3.00 ^b	45.91 ± 0.72 ^b	41.97 ± 1.28 ^a	1353.3 ± 37.39 ^b	2898.0 ± 117.25 ^c
Rizolex-T	28.3 ± 1.50 ^a	44.07 ± 1.85 ^a	92.0 ± 2.00 ^a	47.88 ± 0.97 ^a	42.32 ± 1.07 ^a	1630.2 ± 74.65 ^a	3704.3 ± 211.15 ^a
F value	29.96 ^{***}	79.29 ^{***}	85.24 ^{***}	41.68 ^{***}	44.75 ^{***}	38.91 ^{***}	29.50 ^{***}

Means followed by the same letter are not significantly different at * P < 0.05 ** P < 0.01 *** P < 0.001.

CONCLUSION:

Since the excessive use of pesticides for plant disease control had high risk for environmental pollution and residual effects, therefore all farmers everywhere all over the world are looking for minimizing dependence on chemicals for pest management. The utilization of organic agricultural wastes in this respect is considered as a safe non-chemical control method. The present investigation has demonstrated that blended composted agricultural wastes with botanical plant material (rich in bioactive ingredients)

have potential and can be used as alternative to reduce the use of chemicals for soil-borne disease control. This favor bio-protection of plants against soil-borne pathogens

CONFLICT OF INTEREST:

Authors declare that there is no conflict of interest.

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التأثير الكيحي لشاي (مستخلص) الكومبوست وكومبوست قشر الرمان الجاف على نمو فطري الفيوزاريوم والرايزوكتونيا سولاني كبديل لاستخدام المبيدات الفطرية المصنعة

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معنوي لشدة المرض في نبات الترمس في جميع مراحل النمو (معايير النمو- تراكم النيتروجين - الفوسفور - الخطي بينما في الظروف الحقلية اوضحت النتائج اختزال البوتاسيوم ومكونات المحصول) وكان أكثرهم فعالية هو المستخلص القلوي لقشور الرمان المالح مع شاي الكومبوست. وعلي ذلك استخدام مستخلص قشور الرمان وشاي الكومبوست يعتبر من المعاملات الواعدة للتحكم في الأمراض الفطرية التي تصيب النباتات وتعتبر من المبيدات الفطرية الطبيعية المؤثرة حتى لو كانت بدون فروق معنوية مقارنة بالمبيد الفطري الكيميائي ويمكن أن يكون بديل آمن.

تحدث الأمراض النباتية تدمير لمعظم المحاصيل الزراعية والذي يؤدي إلى نقصا كبيرا لكل من الإنتاج ونوعية هذه المحاصيل. وتسبب المخلفات الزراعية بالإضافة الي ذلك مشكله حقيقيه يجب التخلص منها، ونتيجة لذلك صُممت هذه الدراسة لمعالجة البقايا النباتية بتحويلها إلى كومبوست جيد التهوية، وتم استخدام شاي الكومبوست مع شاي مسحوق قشر الرمان لتقييم إمكانية إستخدامهما (منفردين أو مجتمعين) للمعالجة البيئية الآمنة كبديل للمبيدات الفطرية المصنعة لمقاومة مرضي الخناق والذبول والتي تحدثها فطره رايزوكتونيا سولاني وفيوزاريوم. تم عزل هذه الفطريات من نباتات ترمس مصابه وأظهرت الدراسة المعملية تثبيط للنمو