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Effect of vermicompost on the growth and health of *Impatiens wallerana*

(With 2 Tables & 3 Figures)

Efecto del compost de lombriz sobre el crecimiento y sanidad de Impatiens wallerana

(Con 2 Tablas y 3 Figuras)

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Abstract. *Rhizoctonia solani* damping-off and root rot diseases are frequent in impatiens crops in Buenos Aires. Their control is based in fungicide use. In a sustainable nursery production, beneficial microorganisms that develop in organic amendments may suppress diseases and promote plant growth. The aim of this work was to evaluate the effect of different proportions of vermicompost on the growth and health of patience-plant (*Impatiens wallerana*). The experiment was carried out in a polyethylene greenhouse. Treatments were defined as follows: infested substrate, substrate, sterilized substrate, vermicompost, and vermicompost mixed with infested substrate at 75, 50 and 25% by volume. Seeds of *I. wallerana* were sown in plugs containing the different substrates. Percentage of healthy seedlings was evaluated since emergence, and growth parameters were recorded at day 51. The concentration of pathogen in the different treatments was estimated at the beginning and end of the experiment. Treatments with 100-75% of vermicompost showed important increases of leaf area, plant height and fresh and dry weight of aerial and subterranean organs. Root length was not modified by compost addition. Vermicompost at 75% provided slight control of damping-off caused by *R. solani*.

Key words: vermicompost, growth, damping-off, *Impatiens wallerana*

Resumen. El mal de los almácigos y la pudrición de raíces causadas por *Rhizoctonia solani* son enfermedades frecuentes en los cultivos de alegría del hogar en Buenos Aires. Su control se basa en aplicación de fungicidas. En una producción de vivero sustentable, los microorganismos benéficos que desarrollan en las enmiendas orgánicas pueden ser supresores de enfermedades, como así también promotores del crecimiento de

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las plantas. El objetivo de este trabajo fue evaluar el efecto de diferentes porcentajes de compost de lombriz sobre el crecimiento y la sanidad de las plantas de alegría del hogar (*Impatiens wallerana*). El ensayo se realizó en un invernáculo de polietileno. Los tratamientos fueron sustrato infestado, sustrato, sustrato esterilizado, compost de lombriz y compost de lombriz mezclado con sustrato infestado al 75, 50 y 25% en volumen. Se sembró *I. wallerana* en multimacetas plásticas con los diferentes sustratos. Se registró el porcentaje de plántulas sanas desde la emergencia y parámetros de crecimiento al día 51. Se estimó la concentración del patógeno en los distintos tratamientos al inicio y final del ensayo. Los tratamientos con 100 y 75% de compost de lombriz mostraron incrementos importantes en área foliar, altura de planta y pesos fresco y seco de órganos aéreos y subterráneos. El largo de la raíz no fue modificado por la adición de compost. El compost de lombriz al 75% controló en forma leve el mal de los almácigos causado por *R. solani*.

Palabras clave: compost, crecimiento, mal de los almácigos, alegría del hogar.

INTRODUCTION

Impatiens or patience-plant (*Impatiens wallerana* J.D. Hook.; Balsaminaceae) is an herbaceous ornamental, with colorful flowers, widely grown as a bedding plant (Daughtrey et al., 1995). *Rhizoctonia solani* Kühn causes damping-off and root rot on this host (Farr et al., 1989). The disease was reported in Buenos Aires in 2000 (Grijalba et al., 2000) and its control is currently dependent on chemicals.

Indiscriminate use of fungicides for disease control and their effect on the environment have influenced public interest in the search for natural innocuous products (Lange, 1992). This is especially important for greenhouse workers, who are continually exposed to harmful fungicides (Sirjusingh & Sutton, 1996). The physical and chemical properties of vermicomposts (Albanell et al., 1988; Orozco et al., 1996) make them appropriate to be used as plant growing media (Brinton et al., 1996) and disease suppressive substrates (Szczech, 1999; Rodríguez Navarro et al., 2000). In Argentina, the effect of vermicompost on damping-off caused by *R. solani* was evaluated on Cucurbitaceae (Wright et al., 1999; Rivera et al., 2004) and its influence on plant growth and incidence of *Rhizoctonia* damping-off was studied on Solanaceae (Rivera et al., 2000, 2001). The aim of this work is to evaluate the effect of different proportions of vermicompost on the incidence of *Rhizoctonia* disease and the growth of patience-plant.

MATERIAL AND METHODS

The experiment was carried out in CETEFFHO-INTA-JICA (Ituzaingó, Buenos Aires), in a polyethylene greenhouse. Medium air temperature was set at 21°C.

The pathogen *R. solani* AG-4-HG-II strain Ri, isolated from diseased impatiens plants grown in Buenos Aires, was used for this experiment. Inocula was obtained from pure cultures on potato dextrose agar (PDA) and multiplied by 6-day incubation on 10 ml of PDA contained in Petri dishes. Pieces of 1-cm^2 of mycelial growth were cut off, mixed with the substrate Germinating Mix (Fafard) and incubated in 4.4-dm^3 metal containers at room temperature for 10 days (Rivera et al., 2000). When required, the substrate was sterilized by autoclaving at

121ºC and air pressure of 1 bar for 1 h. One-year-old vermicompost (Biogreen) was tested for disease suppressiveness and plant growth promotion. Treatments were defined as follows: infested substrate (IS), substrate (S), sterilized substrate (SS), vermicompost (VC), and vermicompost mixed with infested substrate at 75, 50 and 25% by volume (75VC-25IS, 50VC-50IS and 25VC-75IS, respectively). Mixtures of infested substrate with vermicompost were incubated at room temperature during 10 days. Inoculum applied to the substrate was the necessary to reach 0.1% by volume in all the treatments.

Seeds of *I. wallerana* cv. Accent Burgundy were sown in plugs (66 cells/plug) containing the different substrates. A Randomized Block Design with 10 replicates was used for the experiment. Percentage of healthy seedlings was observed since emergence. Leaf area, plant height, root length, aerial and root weight were recorded for 4 randomly selected plants, at day 51. The concentration of pathogen in the different treatments was estimated by the beet-seed colonization method described by Ko & Hora (1971), as number of colony forming units per gram of dried substrate (cfu/g). For each of 5 replicates per treatment, 50 sugarbeet (*Beta vulgaris* L.) glomerules (baits for *R. solani*) were randomly distributed on 32 g of substrate (field capacity) in a Petri dish and covered with additional 32 g of substrate. After 48-h incubation at 26ºC, the glomerules were recovered, placed in a colander and washed for 5 min with running tap water. Excess water was blotted with paper towels. The glomerules were then plated on acidified PDA pH 4 (10 glomerules per plate) for incubation at the same temperature during 24 h and examined at 40x for *R. solani* hyphal emergence. The pathogen was identified by the distinctive morphological characteristics of its mycelium. Pathogen concentration was calculated as mean number of infected glomerules per gram of dried substrate. After additional 3-day incubation, data on colony diameter were registered as an additional parameter emerging from this technique. Estimations of inoculum of *R. solani* by cfu and colony diameter were done after sowing and at the end of the experiment. Results on inocula concentration and plant growth were studied by analysis of variance. Plant health data over time were analyzed by mixed models methodology. The covariance structure was selected by Akaika criterion (Littell et al., 1996).

RESULTS

Pathogen-free commercial substrate produced the highest number of healthy seedlings from day 17 onwards (Table 1). At the end of the assay, the number of healthy plants in inoculated substrate was 28% lower than plant stand in sterilized substrate. Among amendments, a tendency to control disease was observed for 75% vermicompost, from day 17. Plant stand diminished along time for infested substrate and for all vermicompost treatments, even at 100%, with no infestation of pathogen. At day 51, the number of plantlets for vermicompost and 75% vermicompost mixed with infested substrate, were higher compared with the rest of inoculated treatments.

Pathogen concentration measured as cfu/g did not differ among *Rhizoctonia* treatments at the beginning and at the end of the experiment (Table 2). However, a decrease in diameter was observed for colonies emerging from substrate mixes with 75% of vermicompost at the end of the assay.

Table 1. Number of healthy seedlings 7 to 51 days from sowing

	Mean number of healthy seedlings*						
Treatment	At day 7	At day 17	At day 22	At day 29	At day 36	At day 43	At day 51
VC	8 a	51 b	51 b	53 b	49 bc	48 bcd	47 b
75VC - 25IS	5 ab	54 b	53 b	52 b	51 b	49 bc	45 b
50VC - 50IS	4 bc	49 b	49 b	47 c	46 bc	42 de	38 cd
25VC - 75IS	5 ab	49 b	48 b	46 c	44 c	39 e	35 d
SS	1 c	60 a	61 a	60 a	61 a	61 a	60 a
IS	3 bc	53 b	52 b	50 bc	48 bc	44 de	43 c
S	2 bc	60 a	58 a	58 a	56 a	53 b	55 a

*different letters within columns indicate significant differences (α : 0.05)

VC: vermicompost; 75VC - 25IS: 75% (vol.) vermicompost + 25% (vol.) infested substrate
50VC - 50 IS: 50% (vol.) vermicompost + 50% (vol.) infested substrate
25VC - 75 IS: 25% (vol.) vermicompost +75% (vol.) infested substrate
SS: sterilized substrate; IS: infested substrate; S: substrate

Table 2. *R. solani* colony forming units and colony diameter for the different treatments

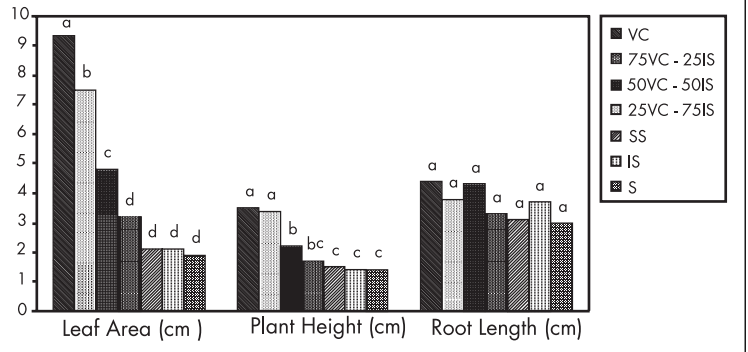
Treatment	Inicial inoculum density		Final inoculum density	
	cfu/g dried substrate*	colony diameter (cm)*	cfu/g dried substrate*	colony diameter (cm)*
VC	0	0	0	0
75VC - 25IS	1.0 a	1.5 a	0.9 a	1.8 a
50VC - 50IS	1.1 a	1.6 a	1.2 a	2.6 b
25VC - 75IS	1.2 a	1.6 a	1.2 a	2.3 b
SS	0	0	0	0
IS	1.2 a	1.7 a	1.2 a	2.8 b
S	0	0	0	0

*different letters within columns show significant differences (α : 0.05)

VC: vermicompost; 75VC - 25IS: 75% (vol.) vermicompost + 25% (vol.) infested substrate
50VC - 50 IS: 50% (vol.) vermicompost + 50% (vol.) infested substrate
25VC - 75 IS: 25% (vol.) vermicompost +75% (vol.) infested substrate
SS: sterilized substrate; IS: infested substrate; S: substrate

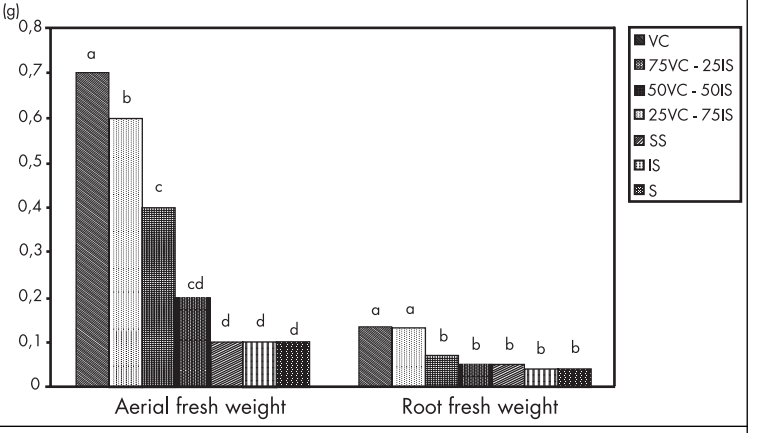
Plantlets grown in 100% vermicompost had the highest values for leaf area and aerial fresh weight (Figs. 1 and 2). Plant height, aerial dry weight and root fresh weight were higher for plots with 100% and 75% of vermicompost (Figs. 1-3). Root length was not affected by either substrate or pathogen (Fig. 1). Vermicompost incorporation at 75% increased leaf area, plant height and aerial fresh weight and at 50% only increased leaf area (Figs. 1 and 2).

Fig. 1. Growth parameters for patience- plants grown in different treatments



Different letters within each group of bars indicate significant differences (α : 0.05)

Fig. 2. Patience-plants fresh weight 51 days from sowing

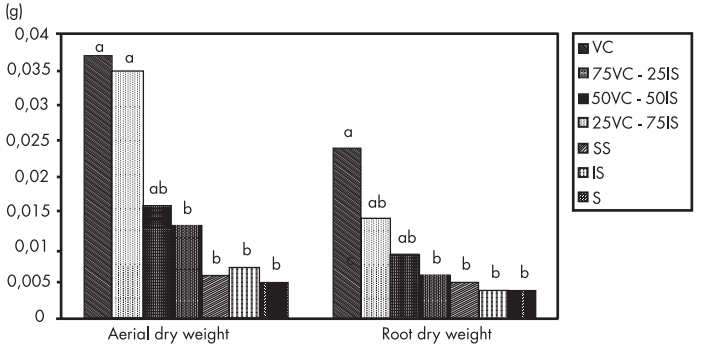


Different letters within each parameter indicate significant differences (α : 0.05)

DISCUSSION

Vermicompost addition to pathogen-infested substrate influenced disease expression at a low level. Although with the same tendency, a more efficient disease control was reported on different horticultural species (Weltzien, 1989; Rivera et al., 2004). Crop protection was dose-dependent in our experiment; only 75% vermicompost diminished the incidence of

Fig. 3. Patience-plants dry weight 51 days from sowing



Different letters within each growth parameter indicate significant differences (α : 0.05)

Rhizoctonia disease, while 50% vermicompost did not differ from inoculated substrate. Twenty five percent vermicompost provided less healthy plants than inoculated substrate. Diab et al. (2003) controlled pre-emergence damping-off by growing impatiens in potting mix amended with 20% thermophilic compost. Rodríguez Navarro et al. (2000) reported that 20% vermicompost reduced Rhizoctonia disease incidence on *Gerbera jamesonii* Bolus. On the contrary, they found that disease was higher with vermicompost at 45%. In our work, 25 and 50% vermicompost were not enough to provide suppression in impatiens. Host-dependent response should be considered as well as compost quality in the analysis of substrate suppressiveness. Composts potential for disease management is based on their microbial activity. An overwhelming body of evidence indicates that microbial communities, stimulated as a result of compost amendments, are responsible for disease suppression (Nelson et al., 2000). Earthworms promote microbial activity and diversity in organic wastes to levels even greater than those in thermophilic composts (Edwards, 2004). The ability of the vermicompost used in this work to control pathogens can be attributed to its microflora, as suppressiveness disappeared after autoclaving (data not shown). Compost biological control can be explained by changes in the activity of pathogens and beneficial agents. The decomposition level of organic matter in compost-amended substrates has a major impact on disease suppression (Hoitink et al., 1997). *R. solani* is highly competitive as a saprophyte (Garret, 1962), but it can not colonize low-cellulose mature compost while i.e. *Trichoderma spp*. is capable of colonizing fresh as well as mature compost (Chung et al., 1988).

Regarding inocula concentration, it was not estimated in related reports. Pathogen concentration estimated as cfu/g was the same for all inoculated substrates, both at the beginning and at the end of this study. However, we detected differences among inocula concentration by quantifying the diameter of colonies of *R. solani* at day 51, which diminished at 75% vermicompost in the mixture. The measurement of colony diameter probed to be more sensitive to detect differences in pathogen population than the estimation of cfu/g of substrate. The reduction

of inoculum concentrations at the end of the experiment could explain our results on disease control for 75% vermicompost. Development of more sensitive methods for the detection of *R. solani* in soil is needed.

Vermicompost at 100% provided lower quantity of seedlings than Germinating Mix, which suggests that it may be somewhat toxic for impatiens growth. Composts provide acceptable to excellent container-growing media, often in amounts exceeding 50% and sometimes up to 100% by volume, even despite initially elevated and potentially toxic contents of soluble salts (Chong, 2005). These can be reduced to benign levels by either mixing with other substrates or leaching previous to planting (Healy, 1995) or with crop normal irrigation practices (Chong, 2005). The vermicompost used in this work has a content of soluble salt, expressed in terms of electrical conductivity, of 13.7 mmhos/cm, that is considered high (Healy, 1995). This could explain plant stand differences between compost and non inoculated substrate treatments.

Impatiens growth was improved by the use of vermicompost in the substrate. To our knowledge, there are few reports on growth promotion by vermicompost in ornamental plants, without the addition of fertilizers. Rodríguez Navarro et al. (2000) considered that 20% vermicompost was optimum for the nutrition of *G. jamesonii*. Atiyeh et al. (2002) detected significant growth promotion on *Tagetes patula* L. by the addition of 30 or 40% vermicompost. Earthworm biological degradation increases humification rate and degree and provides a higher humic acid to fulvic acid ratio, with the presence of small quantities of growth regulators (Senesi, 1989), that could explain impatiens growth promotion observed in our work.

Our results confirm an important activity of vermicompost as impatiens growth promoter and a lesser role as *Rhizoctonia* disease suppressor, which are encouraging in relation to its use in initial phases of the production of this flowering plant. However, Ben-Yephet & Nelson (1999) consider that, in spite of reported success, universal adoption of compost as substrates for biocontrol and growth promotion is threatened by suppression variability and a poor understanding of factors that affect it. The knowledge of compost mechanisms and soil factors that regulate its activity are critical to reduce their percentage in mixtures and increase its efficiency (Lazarovits et al., 2001). Temperature, humidity, compost dosage and target pathogen are among the sources of variability (Ben-Yephet & Nelson, 1999). In assays on the control of *R. solani* in vermicompost-amended substrates, a temperature-dependent response was observed (Rivera et al., 2004). Tendency was similar to that reported by Ben-Yephet & Nelson (1999). It is necessary to continue studies for increasing the knowledge of these systems, and the possibilities of their incorporation in environmentally friendly and sustainable ornamental crop productions.

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