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Study of fat compost from dairy industry wastewater as a new substrate for pepper (*Capsicum annuum* L.) crop

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ABSTRACT

The aim of this study was to analyze the impact of different doses of compost obtained from fats waste of dairy industry mixed with commercial peat-based substrate and with soil on pepper plants development. Furthermore, this investigation attempted to determine the effects of these mixtures on germination and composition of pepper fruits. Fat compost (FC) was obtained by aerobic composting from dairy industry wastewaters mixed with wood chips and green grass. FC extract resulted free of phytotoxicity for seed germination, obtained 99.71% the relative seed germination and 74.10% of germination index. Different growing media were prepared by mixing 0, 10, 20 and 40% of FC with commercial substrate (CS) or Soil in two different experiments. Results showed that the application of FC on CS and Soil respectively, increased plant dry matter. On pepper seedling FC 40 achieves 250 mg plant^{-1} DM. Moreover, higher doses of FC improved yield and several characteristics of fruit as dry matter, diameter and concentrations of some carotenoids. The incorporation of FC did not increase the heavy metals concentration of pepper fruit. At leaves, the highest concentrations of N were reached shown on treatment with FC (Soil: 1.46%, FC 10: 1.92%, FC 20: 2.00%, FC 40: 2.09%). Application of FC for germination and development of pepper plants improved the seedlings, fruit yield and quality.

1. Introduction

The province of Santa Fe, in the central region of Argentina, produces about one half of the milk all over Argentina. Then, dairy industries are a significant component of the regional economy and are involved in the manufacturing of various types of milk products such as fluid milk, butter, cheese, yogurt, etc. The dairy industry is one of the most polluting of industries, not only in terms of the volume of wastewater generated, but also in terms of its characteristics as well. Common techniques for treating dairy industry wastewaters include grease traps, oil water separators for separation of floatable solids, equalization of flow, and clarifiers to remove sewage sludge; biological treatment consists on the aerobic and anaerobic process. Dissolved Air Flotation (DAF) is a relatively simple technology that uses fine air bubbles to float attached solids particles (mostly fats) to the surface of a flotation cell for their removal from a wastewater stream. The management of the DAF biosolids is complicated because of its high water content that deteriorates the landfills and is not suitable for incineration. This type of waste has similar properties that sludge from grease traps and was expected to be suitable for biological treatment as aerobic composting using several co substrates as bulking agent and C–N sources (i.e., wood shavings and chips, green grass).

Nowadays, it is widely accepted that there is a need for finding new substrates which can replace peat in the formulation of plant growth substrates for horticulture and potted plant production (Paradelo et al., 2012; Restrepo et al., 2013). Additionally, organic materials are the safer sources of plant nutrients without any detrimental effect to crops and soil (Hasanuzzamanet al., 2010). The compost acts as a source of slow-release nutrients, available for plants (Chaoui et al., 2003; Raviv, 2005) and prevents nutrient losses into the environment (Giuffré et al., 2011). It is well know that the addition of compost to soils and growing media promotes the development and productivity of different horticultural crops such as tomato (Hernández et al., 2014) and pepper (Pascual et al., 2009; Shrestha et al., 2013). Compost products can be beneficial, supplying nutrients for plant growth, organic matter for soil improvement and agents for plant disease suppression. There are myriad of reasons to consider composting as an option for future management of organic waste: it is a technology that can be used

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on any scale; compost products can be sold on the soil market; it is more flexible than anaerobic digestion and incineration in terms of size, time for planning, construction and pay-back time for investment (Sundberg, 2005; Mehta et al., 2014).

The physical properties and chemical composition of fruits and vegetables have been shown to be highly influenced by agricultural management practices, and organic vegetables in particular are usually considered to have higher mineral content and to be more nutritious and safer than conventionally grown vegetables (Jolly, 1991). The changes in the elemental concentrations are due to internal and external reasons by interactions soil-plant. Among the external, the applied fertilizer, soil fertility level, soil and climatic conditions (rainfall, temperature, etc.,) are among the most important. On the other hand, the internal factors include the genotype, ageing of the plant organ, type of the organ, and yield and growth phase of the plant (Kalavrouziotis et al., 2009). Kalavrouziotis and Koukoulakis (2009) have shown that the interactions between heavy metals and essential elements may supply the plants with significant quantities of nutrient elements as well as heavy metals and may thereby affect plant growth positively or negatively, depending on the type of interaction (synergistic or antagonistic). Like other vegetable crops, peppers (*Capsicum annuum* L.) can be cultivated on conventional substrates such as perlite, rockwool, sand, and other soilless systems, which have replaced the traditional crop grown in agricultural soil in the greenhouse (Zhai et al., 2009; Díaz-Pérez and Camacho-Ferre, 2010). Increasing awareness of the adverse economic and environmental impacts of conventional substrates has stimulated the interest in using organic wastes and agricultural by-products as substrates in soilless culture (Del Amor and Gómez-López, 2009; Lin et al., 2009). On the other hand, there has not been found evidence on the utilization of fat compost to provide organic amendment. The present study focus on the impact of different doses of compost obtained from fats waste of dairy industry mixed with commercial peat-based substrate and with soil for pepper plants development. Furthermore, this investigation attempted to determine the effects of these mixtures on germination and composition of pepper fruits.

2. Materials and methods

2.1. Compost: process and characterization

The compost used in this study was obtained from the aerobic co-processing of fat of wastewaters of dairy industry, wood chips and green grass. The fats wastes were obtained from a Dissolved Air Flotation process (DAF) from a local dairy industry in Rafaela, Santa Fe – Argentina. The composting experiment was carried out using green grass, fat and wood chips as mixture. Wood chips were used as bulking agent. The composting experiment was carried out using green grass, fat and wood chips as mixture. The proportions of mixture fat: wood chips: green grass were 53%:35%:12% (w/w/w). Green grass (*Lolium perenne* L.) and wood chips (*Jacaranda mimosifolia* D.Don and Acacia caven (Molina) Molina) were obtained from campus of CONICET in Santa Fe – Argentina. The composting process lasted 8-month until the maturation stage. During the first period when occurs increased microbial activity and augmented the temperature of mixtures, the homogenization of wastes was performed daily. When the product or composting process reached ambient temperature (stabilization stage), the windrows were turned weekly until maturation stage and subsequent utilization in this study. Deionized water was manually added to the composting mixture when necessary in order to maintain moisture content in the optimal range for composting (40–60%) (Haug, 1993). All composting process occurred at room temperature ranged between 15 °C and 25 °C.

Table 1
The main properties of fat compost (FC), commercial substrate (CS) and soil utilized.

Measurement	FC	CS	Soil
pH	7.68	6.25	6.79
EC (mS cm^{-1})	3.72	0.61	0.30
$N_{kjeldhal}$ (%)	2.77	0.92	0.18
K (g kg^{-1})	135.68	12.5	16.03
Mg (mg kg^{-1})	5.97	2.22	1.45
Na (g kg^{-1})	40.38	4.01	2.03
Ca (mg kg^{-1})	153.50	96.49	3.17
C (%)	43.35	25.41	1.69
OM (%)	78.03	45.73	3.00
C/N	15.65	27.62	9.40
Ash (%)	21.96	54.27	97.00

EC: electric conductivity; OM: organic matter.

The main properties of fat compost (FC), commercial substrate (CS) and soil (S) utilized in both experiments are shown in Table 1. The pH and electrical conductivity (EC) were measured in an aqueous solution (1:10 w/v) (Laos et al., 2002). Nitrogen content was determined in dried samples by using the Kjeldahl method. Potassium (K), Magnesium (Mg), Calcium (Ca) and Sodium (Na) were extracted following EPA Method 200.2 and then analyzed by atomic absorption spectrometry (AAS) (Martin et al., 1994). The mixture samples from the end of seedbed and pots experiments were analyzed by AAS by flame. Organic matter (OM) and percentage of ashes were determined by dry combustion at 550 °C during 4 h. The total organic carbon was determined from OM using a correlation factor (Barrington et al., 2002). Heavy metal content in fat compost was not a significant issue since concentrations in pots were below the limits established by SENASA (Resolución 264/2011), Cd 0.72 mg kg^{-1} DM; Cr 7.03 mg kg^{-1} DM; Cu 2.58 mg kg^{-1} DM; Ni 3.46 mg kg^{-1} DM; Pb 8.88 mg kg^{-1} DM; Zn 137.49 mg kg^{-1} DM.

Phytotoxicity test on FC and CS consists in a germination test utilizing radish (*Raphanus sativus* L.) seeds. The germination test was performed for 5 days at 25 °C in the dark with 15 radish seeds placed on a 9 mm filter paper (Whatman #1) soaked with 7 ml of FC extract or CS extract (Bertran et al., 2004), and placed in a Petri dish. Deionized water was used as a control. A 5 mm primary root was used as the operational definition of germination. The percentages of relative seed germination (RSG%) = (number of seeds germinate in treatment/number of seed germinate in control) × 100, relative root growth (RRG%) = (average root length of seedling in treatment/average root length of seedling in control) × 100, and germination index (GI%) = (RSG X RRG)/100 were calculated (Zucconi et al., 1981; Kumar et al., 2010).

2.2. Plants experiments: growth and development

2.2.1. Seedbed experiments

Four growing media were tested. The commercial growing media used routinely at nursery was used as control. The growing media were prepared by mixing 0–10–20–40% of FC with CS. Composition of growing media mixtures are shown in Table 2. Nursery trays of 36 cells of 0.4 L capacity were used for the experiment. Three replicate trays were filled with each substrate (treatment). One seed of *Capsicum annuum* L. cv Paco, hybrid F1 (sweet variety) per cell was sowed. The trays were distributed randomly in three blocks, each containing all the treatments. The trays were moistened periodically with tap water. Over the 40-day growth-period in the nursery, no fertilizer was applied; seedling nutritional requirements were thus met entirely by the substrates. Seedlings were grown in a glasshouse at 29 °C/18 °C and 50%/70% RH (day/night). The photoperiod was 14 h under natural daylight.

Emergence of pepper plants was checked on alternate days. Seedlings were counted 20 days after sowing. Rates of emergence

Table 2
Composition of growing media mixtures.

Growing media	Formulation (v:v%)
Nursery trays	
CS	100% Commercial substrate (control)
FC 10	10% fat compost: 90% CS
FC 20	20% fat compost: 80% CS
FC 40	40% fat compost: 60% CS
Pots	
Soil	100% Soil (control)
FC 10	10% fat compost: 90% S
FC 20	20% fat compost: 80% S
FC 40	40% fat compost: 60% S

Percentage of each component on a volume basis.

were calculated using a modified Timson's emergence velocity index: $\Sigma G/t$, where G is the number of seeds emerged at 2-day intervals, and t is the total time of emergence (Khan and Ungar, 1998). The plants were harvested 45 days after sowing, when the seedlings reached approximately the commercial transplanting size. At the end of the nursery seedling growth period, the seedling height and stem diameter were recorded. Plant organs were carefully separated, weighed and stored at −80 °C until analysis. Leaf chlorophylls were extracted in 95% (v/v) ethanol and their concentration was quantified in a spectrophotometer (PerkinElmer Lambda 35. UV/VIS, Spectrometer). Calculations were made using the equations of Lichtenthaler (1987). Total soluble proteins (TSP) were analyzed by the protein dye-binding method using bovine serum albumin as a standard (Bradford, 1976). Total soluble sugars (TSS) in leaves and roots were analyzed by reacting 0.25 ml of the extracts with 3 ml of freshly prepared anthrone solution and placing in boiling water for 10 min. After cooling, the absorbance at 620 nm was determined spectrophotometrically (Yemm and Willis, 1954).

The stem height/diameter (H/D) ratio, leaf area per seedling; specific leaf area (SLA); and leaf area ratio (LAR) were obtained as follows. The SLA was calculated as the ratio of the leaf area to leaf dry weight. LAR was calculated as the ratio of the leaf area to plant dry matter (DM). DM was obtained by drying samples at 85 °C to constant mass. The experiment was repeated twice with similar results. Data from both repetitions were analyzed pooled (Total of nine plants per treatment).

2.2.2. Pots experiments

One pepper seedling (*Capsicum annuum* L. cv Paco, hybrid F1, sweet variety) (two or three-leaf stage) was transplanted into each pot with a capacity of 4 L. Four different growing media were used as shown in Table 2. The plants were irrigated only with water until harvest. Plants were grown up under identical conditions that those above specified for the seedbed experiment. Plants were harvested until maturity stage (plants with red fruits) for the determination of growth and yield parameters. The experiment was repeated twice with similar results. Data from both repetitions were analyzed pooled (total of six plants per treatment).

Leaf and fruit Nitrogen content was determined in dried samples by using the Kjeldahl method. Potassium (K), Magnesium (Mg), Calcium (Ca) and Sodium (Na) and heavy metals were extracted following EPA Method 200.3 and analyzed by AAS by flame. Leaf chlorophylls, TSP and TSS were measured as explained before. Fresh samples of pepper fruit were homogenized using a pestle and mortar in the presence of liquid N_2. Lycopene and β-carotene contents were calculated according to the Nagata and Yamashita equations: Lycopene (mg 100 ml^{-1} of extract) = −0.0458 × A663 + 0.204 × A645 + 0.372 × A505 − 0.0806 × A453. β-carotene (mg 100 ml^{-1} of extract) = 0.216 × A663 − 1.22 × A645 − 0.304 × A505 + 0.452 × A453. Lycopene and β-carotene were finally expressed as mg kg^{-1} DM (Navarro et al., 2006). Dry matter (DM) was determined by drying samples at 85 °C to constant mass. To obtain yield, all fruits from each plant (including red, green and breaking-point) were detached. The numbers of fruits were recorded and fruit DM was obtained after drying at 60 °C for 45 days. Morphological traits of fruits (length, thickness and diameter) were recorded in commercially ripe fruits (reds).

Table 3
Main properties of commercial substrate (CS) (seedbed experiments), of soil (control) (pots experiments) and different growing media prepared with fat compost (FC) at the end of experiment period.

Measurement	CS	FC 10	FC 20	FC 40
Seedbed experiments				
$N_{kjeldhal}$ (%)	1.02 ± 0.03c	1.19 ± 0.02c	1.54 ± 0.04b	1.97 ± 0.07a
K (g kg^{-1})	12.27 ± 2.02bc	9.26 ± 0.32c	15.92 ± 0.07b	40.12 ± 1.02a
Mg (mg kg^{-1})	4.27 ± 0.15c	10.33 ± 0.25a	6.03 ± 0.19b	7.16 ± 0.76b
Na (g kg^{-1})	7.73 ± 0.73b	7.34 ± 0.07b	7.62 ± 0.12b	14.22 ± 0.23a
Ca (mg kg^{-1})	136.74 ± 17.66b	136.03 ± 7.48b	166.97 ± 4.74ab	195.86 ± 4.72a
C (%)	24.03 ± 1.44b	26.96 ± 3.48b	35.71 ± 0.76a	37.88 ± 0.04a
OM (%)	43.23 ± 2.57b	48.53 ± 6.26b	64.3 ± 1.38a	68.20 ± 0.06a
Cu (mg kg^{-1})	29.17 ± 0.87ab	31.09 ± 1.19a	33.27 ± 0.48a	25.85 ± 1.37b
Cr (mg kg^{-1})	35.27 ± 0.91b	43.93 ± 1.64ab	39.12 ± 4.25ab	50.97 ± 3.62a
Ni (mg kg^{-1})	34.01 ± 1.30b	47.89 ± 1.47a	45.43 ± 1.07a	51.27 ± 2.07a
Pb (mg kg^{-1})	0.83 ± 0.06a	0.91 ± 0.04a	0.88 ± 0.01a	0.99 ± 0.09a
Zn (mg kg^{-1})	1.66 ± 0.14d	43.33 ± 0.86c	54.71 ± 2.3b	83.58 ± 1.68a
Pots experiments				
$N_{kjeldhal}$ (%)	0.16 ± 0.02c	0.22 ± 0.01bc	0.30 ± 0.03b	0.53 ± 0.05a
K (g kg^{-1})	18.96 ± 0.27b	18.90 ± 1.07b	28.44 ± 1.36ab	35.99 ± 4.21a
Mg (mg kg^{-1})	2.03 ± 0.04a	1.72 ± 0.09a	2.27 ± 0.11a	2.07 ± 0.26a
Na (g kg^{-1})	3.09 ± 0.22c	5.30 ± 0.3b	5.26 ± 0.00b	7.01 ± 0.54a
Ca (mg kg^{-1})	5.06 ± 0.48c	8.81 ± 1.02c	35.52 ± 2.61b	51.29 ± 6.49a
C (%)	2.22 ± 0.06d	3.86 ± 0.33c	4.77 ± 0.08b	7.09 ± 0.21a
OM (%)	4.00 ± 0.11d	6.96 ± 0.61c	8.57 ± 0.14b	12.80 ± 040a
Cu (mg kg^{-1})	0.44 ± 0.06c	0.55 ± 0.00c	1.09 ± 0.07b	2.91 ± 0.21a
Cr (mg kg^{-1})	0.52 ± 0.06a	0.59 ± 0.07a	0.74 ± 0.09a	0.58 ± 0.00a
Ni (mg kg^{-1})	18.21 ± 0.71a	17.68 ± 0.37a	17.79 ± 0.73a	16.70 ± 0.18a
Pb (mg kg^{-1})	0.83 ± 0.04a	1.25 ± 0.19a	0.83 ± 0.00a	0.83 ± 0.01a
Zn (mg kg^{-1})	30.12 ± 0.95b	27.49 ± 1.25b	39.22 ± 1.88ab	43.46 ± 5.23a

OM: organic matter. Within each file means followed by different letter are significantly different ($p < 0.05$) according to Tukey's test. Values represent means ± S.E. ($n = 9$ pepper seedlings and $n = 6$ pepper plants).

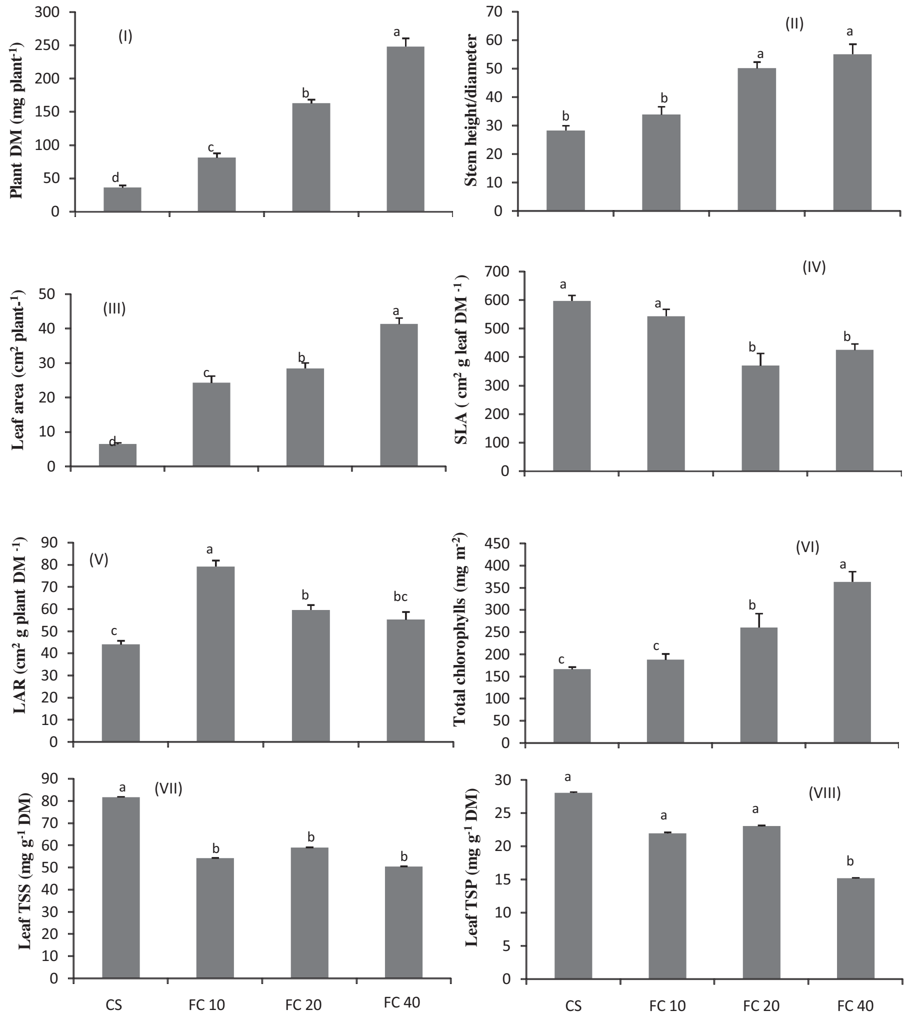

Fig. 1. Main characteristics of pepper seedlings and leaves of pepper seedlings grown in commercial substrate (CS) and different growing media prepared with fat compost (FC). (I) Total DM (dry matter), (II) stem height/diameter, (III) leaf area, (IV) specific leaf area (SLA), (V) leaf area ratio (LAR), (VI) total chlorophylls, (VII) total soluble sugars (TSS) and (VIII) total soluble proteins (TSP). Values represent means ($n = 9$); bars indicate standard error (S.E.) of the mean. Different letters indicate significant differences ($P \leq 0.05$) treatments according to a Tukey's test.

Finally, the harvest index was calculated as the ratio between fruit DM and plant DM.

2.3. Statistical analyses

Statistical analyses were conducted using Statistical Product and Service Solutions for Windows, version 15.0 (SPSS Inc., Chicago, USA). Data was analyzed using analysis of variance (ANOVA), and the differences between the means were tested using a Tukey's *t*-test ($P < 0.05$).

3. Results

3.1. Compost properties and germination assay

Results presented in Table 3 showed that in both experiments. FC addition improves the fertility of growing media and soil due to the increased concentrations of available main macronutrients and organic matter in relation to the increment of FC doses. FC application also led to increased concentrations of heavy metals (Cr, Ni and Zn) in growing media (seedbed experiments) (Table 3). In contrast, FC application only increases Cu and Zn concentrations in soil (pots experiments) (Table 3). The outcomes of the germination test are given in Table 4 and show that 100% relative seed germination was achieved for FC extract and 99.17% for CS extract. The value of relative root growth and germination index in CS extract was approximately double than in FC extract. When the percentage of germination and emergence rate of pepper seedlings were analyzed, not significant differences between the treatments (CS, FC 10, FC 20 and FC 40) were observed (data not shown).

Table 4
Outcomes of germination test of fat compost (FC) and commercial substrate (CS).

Parameter	Control test	FC	CS
pH	5.85	7.68	6.25
Total seeds	135	135	135
Germinated seeds	120	124	120
Mean root length (cm)	7.29	4.28	12.08
RSG (%)	–	100.00	99.17
RRG (%)	–	73.35	165.67
GI (%)	–	74.10	165.04

RSG: relative seed germination. RRG: relative root growth. GI: germination index.

3.2. Seedlings development

Growth characteristics of pepper seedlings are shown in Fig. 1. Results showed that the addition of increasing amounts of FC to substrate improved leaf area, plant dry matter (DM) and the stem

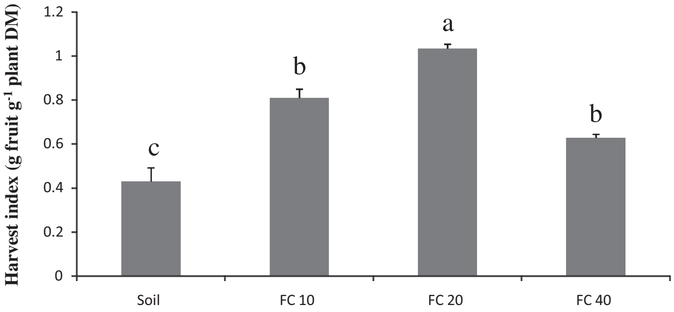

Fig 2. Harvest index of pepper plants grown in pots with different growing media: soil (control), FC 10, FC 20 and FC 40. Values represent means ($n = 6$); bars indicate standard error (S.E.) of the mean. Different letters indicate significant differences ($P \leq 0.05$) treatments according to a Tukey's test.

height/diameter ratio of pepper seedlings (Fig. 1 (I–III)). By contrast, SLA and LAR of seedlings tended to decrease in FC 20 and FC 40 (Fig. 1 (IV) and (V)). On the other hand, leaf chlorophylls content increased in pepper seedlings as increasing the doses of FC applied (Fig. 1 (VI)). However, the concentrations of total soluble sugars (TSS) in leaves significantly decreased in all treatments amended with FC, and the total soluble proteins (TSP) decreased in seedlings grown in FC 40 (Fig. 1 (VII) and (VIII)).

3.3. *Plants growth and yield*

Table 5 shows main characteristics of plants grown in pots with different culture media. Results showed that leaf, stem and root DM production was significantly improved with increasing doses of FC. Similarly, leaf area and leaf chlorophylls were increased after FC addition to soil. TSP concentrations on leaves of pepper plants were increased in FC 20 and FC 40 treatments. Leaf TSS concentrations were increased in pepper plants treated with FC, being higher in plants grown in FC 20. On the other hand, yield and fruit DM were significantly improved regardless the dose of FC applied (Table 5). A similar response was obtained for length and diameter of fruits. However, the increase of number of fruits per plant was dependent on FC doses, reaching the highest values for FC 40. Also, our results show that increasing doses of FC significantly improved the Lycopene and β-carotene contents in pepper fruits (Table 5). Finally, harvest index calculated from data of yield and plant DM showed that application of FC improved this parameter in all FC doses applied, but the maximum values were obtained in the FC 20 treatment (Fig. 2).

3.4. *Elemental analysis*

The N concentrations in leaves increased with the addition of increasing amounts of FC to substrate. In contrast, not significant differences between the treatments were observed on pepper fruits (Table 6). Furthermore, it can be seen from the data in Table 6 that the concentrations of heavy metals were accumulated at leaves compared with pepper fruits.

4. Discussion

The most frequently cited problems regarding the use of compost in the growing medium for vegetable transplants include unstable or immature compost, high soluble salt concentrations and poor water-holding capacity (Carmona et al., 2012). The principal requirement of compost for it to be safely used in soil is a high degree of stability or maturity, which implies a stable OM content and the absence of phytotoxic compounds and plant or animal pathogens (Bertran et al., 2004). Maturity is associated with plant growth potential or phytotoxicity (Iannotti et al., 1993), whereas stability is often related to the compost's microbial activity. However, both stability and maturity usually go hand in hand, since phytotoxic compounds are produced by the microorganisms in unstable composts (Zucconi et al., 1985). The germination rate and germination index (GI) are commonly used to assess the phytotoxicity of compost since it is a problem associated with immature composts. Zucconi et al. (1981, 1985) reported that the compost with GI values greater than 80% was phytotoxin-free and it is considered as having completed maturity, GI values lower than 60% would indicate incomplete stabilization of the organic matter and consequently the presence of toxic substances. In the present study, GI of FC treated substrate was slightly lower than the values reported by Zucconi et al. (1981, 1985) as optimum but the germination rate was almost similar than in commercial compost (Table 4). The lower RRG could be result from higher electric conductivity on the fat compost (Table 1). No significant differences in the percentage of germination (Table 5) and in the rate of emergence (data not shown) were detected on seedlings grown at nursery trays with CS and with FC. Our data agree with those reported by Pérez-Murcia et al., 2006; Carmona et al., 2012 and Restrepo et al. (2013). Both germination and root development would be influenced by any toxic substances, and root development was usually more sensitive than germination (Ko et al., 2008). The fat compost utilized at this study did not present any substances to cause inhibition or retard germination or root development of pepper seedlings.

Composts have shown to enhance plant growth in several occasions and these growth enhancements have been attributed to an improvement of the physical, chemical and biological properties of the growing substrate. Generally, the replacement of peat with moderate amounts of composts produces beneficial effects on plant growth due to the increase on the bulk density of the growing media and the decrease on total porosity and amount of readily available water in the pots (Papafotiou et al., 2005; Grigatti et al., 2007). Such changes in the physical properties of the substrates might be responsible for the better plant growth with the lower doses of compost as compared to the peat-based substrate. In spite that the amount of nutrients in these amendments varies depending on the parent material from where they are originated, compost constitute a slow release source of nutrients that supply the plants with the nutrients when they are needed (Nevens and Reheul, 2003). Further, several examples in the literature show that composts are able to enhance the growth of horticultural plant species what can be expected because of the supply of nutrients (Grigatti et al., 2007; Killi and Kavdir, 2013; Restrepo et al., 2013; Shrestha et al., 2013). Composts are potential substitutes of mineral fertilizers for sustainable agriculture. They are commonly applied to compensate organic matter loss and nutrient depletion. Some authors (Chandra et al., 2008; Scaglia et al., 2015) suggest that the effectiveness of organic amendments could be due not only to nutrients and their availability to plants, but also certain substances that facilitate plant growth, such as hormones and growth-promoting substances for plants. Such substances could come from waste used in the composting process as raw materials. Xu et al., (2012) support that compost also have bio-stimulating effects on plant growth because they contain hormones, hormone-like molecules and other growth promoting substances. Compost aqueous extracts, for example, have been reported to stimulate plant growth because of the presence of hormones, vitamins and other biostimulant components (Pedurand and Reynaud, 1987; Du Jardin, 2012). The phytohormones can be present in organic raw material (Arthur et al., 2007) or they can be produced during biomass transformation. On the other hand, several articles have reported the ability of biomass to stimulate plant growth independently of the presence of identified hormones, ascribing this effect to the presence of specific

Table 5
Main characteristics of pepper plants and pepper fruits grown in Soil (control) and different growing mixtures prepared with fat compost (FC).

Measurements	Soil	FC 10	FC 20	FC 40
Pepper plants				
Leaf DM (g planta $^{-1}$)	2.51 ± 0.14c	5.97 ± 0.27b	5.87 ± 0.53b	13.19 ± 1.02a
Stem DM (g planta $^{-1}$)	2.07 ± 0.19c	5.82 ± 0.35b	6.95 ± 0.25b	10.72 ± 0.54a
Root DM (g planta $^{-1}$)	1.48 ± 0.10c	2.05 ± 0.15bc	2.33 ± 0.26a	3.14 ± 0.24a
Leaf area (cm^2 planta^{-1})	900.34 ± 94.21c	1998.33 ± 125.96b	2492.98 ± 99.20a	2938.14 ± 193.35a
Stem height/diameter	85.09 ± 4.11a	97.91 ± 4.73a	96.62 ± 6.21a	91.19 ± 5.66a
Total chlorophylls (mg m^{-2})	142.69 ± 8.07c	196.89 ± 13.88c	389.78 ± 47.47b	510.29 ± 45.57a
Leaf TSP (mg g^{-1} DM)	23.63 ± 0.73 b	22.75 ± 1.13 b	31.30 ± 2.06 a	35.56 ± 0.833 a
Leaf TSS (mg g^{-1} DM)	35.56 ± 4.17 c	69.91 ± 4.01 b	106.13 ± 9.55 a	73.38 ± 7.66 b
Pepper fruits				
Yield (g fruit DM plant^{-1})	2.44 ± 0.41b	12.91 ± 0.94a	14.17 ± 0.46a	15.04 ± 1.55a
Fruit number (number plant $^{-1}$)	1.00 ± 0.00c	2.26 ± 0.17b	2.87 ± 0.10ab	3.14 ± 0.31a
Fruit DM (g DM fruit^{-1})	1.14 ± 0.08b	5.14 ± 0.42a	5.31 ± 0.35a	5.10 ± 0.29a
Fruit length (cm)	3.52 ± 0.32b	6.91 ± 0.33a	6.55 ± 0.46a	5.85 ± 0.40a
Fruit diameter (cm)	3.65 ± 0.33b	5.37 ± 0.11a	5.74 ± 0.4a	4.97 ± 0.17a
Fruit thickness (cm)	0.52 ± 0.03a	0.51 ± 0.02a	0.55 ± 0.05a	0.47 ± 0.02a
Carotenoids				
Lycopene (mg kg^{-1} DM)	352.76 ± 27.71c	244.35 ± 35.43c	671.83 ± 44.64b	930.78 ± 40.57a
β-carotene (mg kg-^{1} DM)	661.25 ± 43.73b	534.99 ± 43.08b	1035.20 ± 105.05a	1065.57 ± 101.73a

Within each file, means followed by different letters are significantly different ($p \leq 0.05$) according to a Turkey's test. Values are means ± S.E. ($n = 6$).

organic molecules, named hormone-like compounds (Quilty and Cattle, 2011; Jindo et al., 2012). The addition of FC increased the biomass in pepper seedlings (Fig. 1 (I)) and pepper plants (Table 5), in agreement with data reported by other authors (García-Gómez et al., 2002; Pérez-Murcia et al., 2006; Díaz-Pérez and Camacho-Ferre, 2010); this situation has been attributed to the great input of nutrients provided by composts, especially N and K. In general, our results showed that the increasing rate of compost in the media induced an increase in the plant DM, stem height/diameter and leaf area on pepper seedlings (Fig. 1 (I–III)). Herrera et al. (2008) reported that the height/diameter ratio in tomato seedlings was higher in those who grew up in a culture medium of white peat, perlite and solid municipal waste compost. In contrast, Bustamante et al. (2008) showed that increasing rates of composts produced a decrease in the plant growth, despite the increase in the nutrient concentration of these media, probably due to the increase in EC.

Previous studies have noted the importance of the increase in biomass of seedlings grown in culture media with different composts compared to those grown in peat, since them exhibited better quality and suitability for transplanting (Herrera et al., 2008). Seedling resistance to transplant stress is directly related to DM content, which improves seedling establishment in the soil or growth substrate (Pimpini and Gianquinto, 1991). In nursery-produced tomatoes, seedling leaf area appears to be directly related to fruit production per plant, although Leskovar et al. (1991) report that beyond a certain maximum leaf area per seedling there is no appreciable increase in fruit production, and that increasing this index may therefore not be appropriate. Our results agree with Herrera et al. (2008), showing an increase on leaf area on pepper seedlings grown with major doses Fat Compost (Fig. 1 (III)) according with an increment of the number of fruits per plant (Table 5). Lazcano et al. (2009) indicated that substitution of peat by compost increased the aerial biomass and the root biomass of the tomato plants as compared to the pure peat-based substrate. Their data showed that the increase on plant aerial biomass were different in each substrate depending on the dose. Substitution of the peat-based substrate by 10 and 20% compost produced significant increases in aerial biomass, while 50% substitution did not produce any significant difference as compared to peat. In contrast, our results also indicate an increase on biomass with the higher doses (FC 40).

The SLA index, i.e., the ratio of leaf area to dry leaf weight reflects the relative proportion of assimilator tissues to conductor and mechanical tissues. This value serves to assess transplant stress resistance, which increases as SLA decreases. The LAR index is defined as the ratio of leaf area to seedling dry matter, and represents the relationship between photosynthetic material and respiratory material in the plant. It is also used to evaluate seedling resistance at transplant (Herrera et al., 2008). In this research, the pepper seedlings and plants were affected with the doses of FC on growing media (Fig. 1). The SLA and LAR were lower in pepper seedlings grown with FC 40 and FC 20 (Fig. 1 (IV) and (V)). Herrera et al. (2008) mentioned that addition of municipal solid waste compost did not produce significant differences on specific leaf area (SLA) or leaf area ratio (LAR). Furthermore, Díaz-Pérez and Camacho-Ferre (2010) reported the increase of SLA with the

Table 6
Principal elements and heavy metals of pepper plants grown in Soil (control) and different growing media prepared with fat compost (FC) at the end of experiment period.

Treatment	$N_{kjeldhal}$ (%)	K (g kg^{-1})	Mg (mg kg^{-1})	Na (g kg^{-1})	Ca (mg kg^{-1})	Cu (mg kg^{-1})	Cr (mg kg^{-1})	Ni (mg kg^{-1})	Zn (mg kg^{-1})
Leaves									
Soil	1.5 ± 0.0b	496.1 ± 10.3a	5.6 ± 0.4a	22.7 ± 0.4a	154.8 ± 12.2b	196.4 ± 2.3b	8.7 ± 0.0a	7.8 ± 0.4a	85.7 ± 3.0a
FC 10	1.9 ± 0.0a	476.4 ± 15.9a	5.9 ± 0.0a	11.2 ± 21.0b	188.1 ± 2.9a	244.0 ± 14.0a	7.1 ± 0.1b	7.7 ± 0.0a	70.2 ± 0.2b
FC 20	2.0 ± 0.0a	473.6 ± 24.0a	6.1 ± 0.1a	10.3 ± 0.9b	177.0 ± 4.3ab	252.6 ± 8.5a	7.5 ± 0.4b	7.9 ± 0.1a	57.8 ± 0.1c
FC 40	2.1 ± 0.0a	537.9 ± 0.3a	5.6 ± 0.0a	4.7 ± 0.0c	121.0 ± 0.1c	261.3 ± 0.8a	7.4 ± 0.0b	6.3 ± 0.6a	45.7 ± 0.0d
Fruit									
Soil	1.6 ± 0.1a	374.8 ± 24.6a	2.6 ± 0.0a	4.9 ± 0.6a	24.6 ± 0.5a	12.3 ± 1.0a	6.4 ± 0.8a	6.7 ± 0.5a	14.2 ± 0.0a
FC 10	1.2 ± 0.1a	290.2 ± 29.1ab	2.1 ± 0.1ab	3.3 ± 0.1b	17.8 ± 1.1b	6.8 ± 0.7b	2.9 ± 0.0b	4.9 ± 0.2b	14.3 ± 0.4a
FC 20	1.2 ± 0.1a	230.6 ± 20.7b	1.6 ± 0.0 a	2.9 ± 0.2b	11.1 ± 0.8c	4.8 ± 0.8b	1.4 ± 0.2c	4.1 ± 0.2b	10.9 ± 1.4a
FC 40	1.4 ± 0.0a	228.4 ± 39.5b	1.9 ± 0.3 ab	2.8 ± 0.4b	9.0 ± 1.4c	5.5 ± 0.5b	1.0 ± 0.1c	4.2 ± 0.5b	11.5 ± 1.3a

Within each element, means followed by different letters are significantly different ($P < 0.05$) according to a Tukey's test. Values are means ± S.E. ($n = 6$).

presence of compost compared with peat or habitual nursery substrate. In the present study, the decrease in SLA and LAR coincided with a higher concentration of total chlorophylls on these treatments (Fig. 1, (IV), (V) and (VI)). This pattern is contrary to those obtained by Carmona et al. (2012) concerning the chlorophylls concentration. They observed not substantial differences attributed to chlorophylls concentration on pepper plants grown on compost (mixture of dealcoholised grapevine marc and grape stalk) and peat.

Pascual et al. (2009) reported that the application of higher doses of sewage sludge and composted sludge to pepper plants increased leaf number, leaf area, and plant and fruit DM per plant. They concluded that composted wastes improve vegetative growth more than growth of reproductive organs. The increased growth and yield have been attributed directly to nutrient availability and indirectly to increased soil rhizosphere microorganisms activity (Pascual et al., 2008; Zayed et al., 2013), through the production of growth-stimulating plant hormones (Miransari and Simth, 2014). On the other hand, Vaughn et al. (2011) reported the increment on doses of ground tassel not affect the number and weight of tomatoes fruit per plant. But a decrease of number of fruits per plant and in yield at high doses of tassel was found. Compost is an excellent source of macro and micronutrients, especially N. In the present work FC increased the estimated available N in the amended substrates compared with control (CS and Soil), which may partially explain the increase in plant growth and fruit yield of these treatments (Table 5). Pepper was a good source of carotenoids, which could vary in composition and content owing to differences in genetics and maturation (Conforti et al., 2007; Serrano et al., 2010). Carotenoids are efficient antioxidants, quenching singlet oxygen and preventing lipid peroxidation in vitro (Institute of Medicine, National Academy of Sciences, 2000). Riahi and Hdider, (2013) reported that lycopene contents recorded for tomato plants grown with organic fertilizer confirm that genotype significantly affects carotenoid content in organic tomato but not between organic fertilizer treatments. By contrast, our data clearly showed higher concentrations of lycopene and b-carotene in fruits developed in FC 20 and FC 40, suggesting that fruit quality could be improved by application of compost to pepper crop.

Increasing metal concentrations and changes in the distribution of metals in soil amended with compost in the long-term are generally reported to increase the concentrations of heavy metals in the tissues of plants growing in the soil (Kabata-Pendias, 2004). This depends on the amount of metal in the soil, soil physico–chemical properties (e.g., soil pH), the strength of binding of the elements in soil and the ability of the plant to regulate the uptake of the elements. Thus, Zn is relatively labile and is readily transferred to plant tissues, and it is also usually present in larger concentrations in sludge and compost-amended soil compared with the other elements (Smith, 2009). Copper, on the other hand, tends to be more strongly sorbed in soil and plants regulate the uptake of this element more effectively than with Zn (Kabata-Pendias, 2004). Therefore, plant tissue concentrations and availability of Cu are usually much lower and less sensitive to inputs of this element to soil in compost or sludge compared with more mobile elements such as Zn (Zheljazkov and Warman, 2004). The application of FC to soil resulted in increased Cu and Zn concentrations (Table 3) but them always were lower than the permissible limits (Ley 24051, decreto n° 831/93). However, no significant accumulation of these elements was detected in leaves or fruits (Table 6), suggesting that translocation of Cu and Zn from roots to fruits was impaired. This effect was showed previously in barley grain (Antolín et al., 2005). The amounts of different concentrations of heavy metals on pepper fruits were lower than the permissible limits of ANMAT (2001).

5. Conclusions

This study shows that fat compost application on different doses to pepper plants provoked positive impacts. The main impact was the growth improvement of the seedlings on nursery trays because plants obtained all necessary nutrients from this compost. Specifically the application of FC increased plant DM, stem height/diameter, leaf area and total chlorophylls concentrations. All these parameters improved with increasing doses of FC. These findings suggest that it is possible to substitute commercial substrate (peat-based) by fat compost for the production of pepper plants in nurseries trays. In addition, the applied doses did not cause apparent damage or nutritional deficiency on pepper seedlings. Besides, the application of different doses of fat compost to soil increased the plant DM and improved fruit yield and quality. To our knowledge, this is the first study reporting physiological and nutritional responses of plants cultivated with a waste dairy residue. We consider important to continue the study of the effects of fat compost to successive applications on plants, as well as also under water stress conditions. On the other hand, this research shows the effectiveness of reusing of a residue as from composting process to obtain a product free of phytotoxicity effect and which enhance a crop of agronomic interest in the zone.

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