Effect of vermicompost, worm-bed leachate and arbuscular mycorrizal fungi on lemongrass (*Cymbopogon citratus* (DC) Stapf.) growth and composition of its essential oil

Elizabeth León-Anzueto¹ · Miguel Abud-Archila² · Luc Dendooven³ · Lucía María Cristina Ventura-Canseco² · Federico A. Gutiérrez-Miceli²

- 1 Instituto Tecnológico de Tuxtla Gutiérrez, División de Estudios de Posgrado e Investigación, Tuxtla Gutiérrez, Chiapas, México
- 2 Instituto Tecnológico de Tuxtla Gutiérrez, Departamento de Ingeniería Química y Bioquímica, Tuxtla Gutiérrez, Chiapas, México
- 3 Centro de Investigación y de Estudios Avanzados, Departamento de Biotecnología y Bioingeniería, Laboratorio de Ecología de Suelos, México

Corresponding author: fgmiceli@gmail.com
Received May 10, 2011 / Accepted August 30, 2011
Published online: November 15, 2011
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Abstract The objective of this study was to investigate the effect of vermicompost, worm-bed leachate (WBL) and *Glomus mosseae*, an arbuscular mycorrhizal fungi (AMF), on growth of lemongrass (*Cymbopogon citratus* (DC.) Stapf). A response surface methodology, *i.e.* a three-level Box Benhen design with three repetitions and three blocks, was applied to optimize biomass production, essential oil yield and its composition. Application rates of *Glomus mosseae* were 0, 1 or 2 g plant⁻¹, vermicompost 0, 5 or 10 g plant⁻¹ and WBL 0, 10 and 20%. The AMF had no significant effect on the variables tested, but vermicompost had a significant effect on essential oil yield and WBL on essential oil yield, myercene concentration and shoot dry weigh (p < 0.05). It was found that lemongrass fertilized with 2.0 g *G. mosseae*, 5.0 g vermicompost and 20% worm-bed leachate would yield 0.797% essential oil of which 62.6% was citral.

Keywords: citral, desirability function, lemongrass, myrcene, optimization, response surface methodology

INTRODUCTION

Lemongrass (*Cymbopogon citratus* (DC.) Stapf.) is cultivated in many countries around the world, e.g. Argentina, Brazil, Cuba, Ecuador, India, Singapore, China, France, Madagascar, Haiti, Puerto Rico, Mexico, Guatemala, Honduras and Salvador, for its essential oil rich in terpene compounds (Soto-Ortiz et al. 2002). Citral, the main constituent of lemongrass essential oil, is used in perfumery, confectionery and infusions, and used as raw material in the synthesis of ionone, aromatic substances, and vitamin A (Theaganajar and Kumar, 1995). In Europe, leaves are used in teas and infusions. In Mexico, lemongrass tea is used traditionally as a sleep aid, tranquilizer, digestive, anti-influenza and antispasmodic (Rauber et al. 2005). Recently, anticonvulsant activities of the essential oils (EOs) from *Cymbopogon winterianus* Jowitt and *C. citratus* (DC) Stapf. in mice have been reported (Blanco et al. 2009), which has stimulated the cultivation of lemongrass in Chiapas Mexico.

Different organic cultivation techniques have been used to increase plant yield and essential oil in lemongrass. Lemongrass has been inoculated with arbuscular mycorrhizal fungi (AMF), e.g. Glomus mosseae and G. fasciculatum, and biomass yield increased by 3-10% (Soto-Ortiz et al. 2002). Vermicompost and its derivates are often used in organic farming, but little information exists about a possible effect on lemongrass and its essential oil. Vermicomposting is the nonthermophilic biodegradation of organic material through the interaction between earthworms and microorganisms

(Arancon et al. 2003). Leachate derived from vermicomposting often called "worm tea" or worm-bed leachate is regarded beneficial to the crops. It can be used as a liquid fertilizer as it contains large amounts of plant nutrients (Oliva-Llaven et al. 2010). Additionally, worm-bed leachate might contribute to plant development as it contains large amounts of humic acids (Arancon et al. 2005). Humic acids are molecules that regulate many processes of plant development including macro and micro nutrient adsorption (Atiyeh et al. 2002).

Response surface methodology (RSM) is a collection of mathematical and statistical techniques widely used to determine the effects of several variables and to optimize different biotechnological process (Georgakopoulos et al. 2011). The objective of the present investigation was to optimize the use of vermicompost, worm-bed leachate, and the arbuscular mycorrhizal fungus *G. mosseae* in the organic cultivation of lemongrass. Plant yield in terms of shoot dry weight, essential oil yield and its composition were monitored.

MATERIALS AND METHODS

Vermicompost and worm-bed vermicompost

Eight beds (1.5 m x 6.6 m and 1 m deep) were used to obtain worm-bed leachate. Each bed was covered with a plastic sheet to protect the vermicompost against sun and rain. A total bed area of 79 m² was available. Cow manure was composted thermophilically for two months while mechanically being turned over every 15 days. The composted cow manure, adjusted to 80% moisture content, was placed in the beds to a depth of 0.5 m and earthworms (*Eisenia fetida*) were added at a rate of 25 g earthworms/kg cow manure or 2.5 kg earthworms/m² bed. The mixture was left for two months. Each bed contained a leachate drainage and collection system. Leachate from each bed was collected in a separate 200 L tank, pumped into a central collection 1500 L tank and characterized.

Table 1. Experimental design with code levels and real values for evaluating AMF, vermicompost and wormbed leachate on growth, essential oil yield and composition of essential oil of *Cymbopogon citratus* (DC) Stapf. plants.

	AMF		Vermic	ompost	Worm-bed leachate	
	Code levels	Real values	Code levels	Real values	Code levels	Real values
Treatment	X ₁	(g plant ⁻¹)	X_2	(g plant ⁻¹)	X ₃	(ml plant ⁻¹)
1	0	1	0	5	0	10
2	-1	0	0	5	+1	20
3	0	1	-1	0	+1	20
4	+1	2	-1	0	0	10
5	0	1	-1	0	-1	0
6	-1	0	+1	10	0	10
7	0	1	+1	10	-1	0
8	0	1	0	5	0	10
9	+1	2	+1	10	0	10
10	-1	0	-1	0	0	10
11	0	1	+1	10	+1	20
12	-1	0	0	5	-1	0
13	+1	2	0	5	-1	0
14	+1	2	0	5	+1	20
15	0	1	0	5	0	10

Table 2. Effect of different treatments for evaluating AMF, vermicompost and worm-bed leachate on growth, essential oil yield and composition of essential oil of Cymbopogon citratus plants.

Treatment	AMF	Vermicompost	Worm-bed leachate	Essential oil content	Aerial dry weight	Citral content	Linalool content	Myrcene content
	(g plant ⁻¹)	(g plant⁻¹)	(ml plant ⁻¹)	(%)	(g)	(%)	(%)	(%)
1,8,15	1	5	10	0.18 ± 0.04	29.2 ± 4.9	59.7 ± 5.7	1.02 ± 0.23	12.6 ± 2.3
2	0	5	20	0.42 ± 0.04	33.4 ± 2.4	60.3 ± 0.1	1.14 ± 0.04	16.4 ± 0.7
3	1	0	20	0.68 ± 0.00	34.9 ± 2.7	61.6 ± 0.6	1.04 ± 0.03	13.6 ± 0.2
4	2	0	10	0.32 ± 0.18	25.7 ± 1.3	60.7 ± 3.3	0.92 ± 0.3	10.1 ± 2.0
5	1	0	0	0.63 ± 0.01	26.3 ± 1.1	60.6 ± 1.3	0.88 ± 0.38	26.4 ± 1.1
6	0	10	10	0.65 ± 0.04	30.0 ± 5.4	60.2 ± 1.2	1.15 ± 0.31	12.4 ± 3.2
7	1	10	0	0.26 ± 0.01	40.7 ± 2.2	64.0 ± 1.4	0.88 ± 0.17	10.7 ± 1.5
9	2	10	10	0.70 ± 0.07	32.1 ± 1.1	60.5 ± 0.5	1.15 ± 0.04	15.8 ± 0.5
10	0	0	10	0.14 ± 0.08	34.1 ± 2.2	60.7 ± 0.3	1.18 ± 0.01	15.4 ± 0.5
11	1	10	20	0.69 ± 0.09	28.9 ± 6.2	60.8 ± 2.1	1.10 ± 0.04	15.2 ± 1.5
12	0	5	0	0.79 ± 0.00	39.7 ± 1.5	63.5 ± 0.2	0.95 ± 0.14	10.2 ± 2.7
13	2	5	0	0.16 ± 0.08	35.2 ± 1.6	61.1 ± 1.3	0.98 ± 0.16	13.5 ± 3.7
14	2	5	20	0.68 ± 0.02	44.1 ± 2.8	62.7 ± 2.8	1.09 ± 0.32	12.1 ± 1.7

Glomus mosseae was obtained from the microbial collection of Cinvestav-Irapuato (Mexico). Rhodes grass (*Chloris gayana* Kunth (L.'t Mannetje and S.M.M. Kersten)) was inoculated with them and cultivated in a sand:soil (1:1) mixture. Spores of *G. mosseae* were retrieved from the rhizosphere of Rhodes grass. Each *C. citratus* plant was inoculated with 200 spores (1 g) or 400 spores (2 g) of *G. mosseae*.

Characterization of vermicompost and worm-bed leachate

The pH was measured in 1:2 vermicompost- H_2O (w:w) solution using a 716 DMS Titrino pH meter (Metrohm Ltd. CH.-9101 Herisau, Switzerland) fitted with a glass electrode. The Electric conductivity (EC) was determined in 1:5 vermicompost/ H_2O (w:w) solution. The cation exchange capacity (CEC) was measured with the 1 N ammonium acetate method (pH 7.0) (Renault et al. 2009). Total P was determined by digestion with $HNO_3/HCIO_4$ mixture 1:3 (v:v) with sodium carbonate fusion. Extractable P in the 0.5 M $NaHCO_3$ extract was determined by the antimony-potassium-tartrate method. Organic matter was determined by chromic acid titration method. Total N was measured by the semi-micro Kjeldhal method, Apparent density was determined by the method with the cylinders of 100 cm³ (Da, q/cm^3). Humic and fulvic acids were determined by the Walkley and Black (1934) method.

Lemongrass slips were obtained from healthy adult plants. One lemongrass slip was planted in a plastic pot (25 cm diameter and 30 cm) containing 5 L of the different mixtures of vermicompost and peat moss (Table 1). Spores of *G. mosseae* were placed 8 cm from the bottom of the pots. Plants were watered with 250 mL worm-bed leachate diluted at different concentrations every 48 hrs for 4 months (Table 1). No additional fertilizer was applied and the plants were harvested after four months.

Experimental design and statistical analysis

A response surface 3-level Box Benhen design with 3 repetitions of central point (in each block) was applied to optimize yield, essential oil yield and its composition. The experiment was done with 4 blocks. Each plant was fertilized with 0, 1 g (200 spores of *G. mosseae*) or 2 g (400 spores of *G. mosseae*), 0, 5 or 10 g vermicompost and 0, 10 or 20 % worm-bed leachate as shown in Table 1.

All data were analyzed by multiple regressions with the least square method:

$$Y = \beta_0 + \beta_1 A + \beta_2 B + \beta_3 C + \beta_4 A^2 + \beta_5 A * B + \beta_6 A * C + \beta_7 B^2 + \beta_8 B * C + \beta_9 C^2$$

[Equation 1]

Where β : the coefficients to be defined, A: amount of G. mosseae per plant, B: the amount of vermicompost per plant, and C: the % worm-bed leachate. R^2 values were calculated for each equation.

All results were subjected to a one-way analysis of variance to test for significant differences between the treatments using PROC GLM (SAS statistical package) with the Tukey's Studentized Range test (P < 0.05) (SAS, 1989). The response surface graphs and desirability function analysis were made with Statgraphics plus software (Statgraphics, 1999).

Dry weight and essential oil content

Shoot plant dry weight was determined by drying the plant in an oven at 60°C under vacuum for 24 hrs. Total essential oil content was measured by hydrodistillation using a Clevenger type apparatus. A 40 g sub-sample of lemongrass leaves was cut in 3 cm long strips, added to a 500 mL flask containing 200 mL distilled water and boiled for 30 min (Szumny et al. 2009). The vapours were condensed by a condenser. The oils were dried with anhydrous sodium sulphate and stored in amber glass jars at 4°C.

Citral, linalool and myrcene identification and quantification

A 1 μ L sample of essential oil was analyzed by gas chromatography coupled with mass spectrometry (GC-MS) (Agilent 5975C) to identify and quantify citral, b-myrcene and linalool. The separation was achieved with a fused silica capillary DB-WAX column (60 m x 0.25 mm i.d., film thickness 0.25 μ m) (J & W Scientific, Folsom, CA, USA). Temperature of the column was increased from 70 to 250°C at 16°C/min up to 150°C and at 25°C/min up to 250°C. The injector and detector temperatures were maintained at 250°C. The carrier gas was helium (He) flowing at 1.0 mL/min. All analyses were done in duplicate.

Table 3. Effect of AMF, vermicompost and worm-bed leachate on aerial dry weight (g), and concentrations essential oil, citral, linalool and myrene of *Cymbopogon citratus* plants.

	Essential oil content (%)	Aerial dry weight (g)	Citral content (%)	Linalool content (%)	Myrcene content (%)
AMF					
0	0.437 ^a	61.50 ^a	1.11 ^a	13.61 ^a	34.76 ^a
1	0.529 ^a	61.09 ^a	1.00 ^a	12.64 ^a	31.56 ^a
2	0.563 ^a	61.58 ^a	1.03 ^a	12.90 ^a	34.86 ^a
Vermicompost (%)					
0	0.496 ^{ab}	61.27 ^a	1.01 ^a	12.66 ^a	31.74 ^a
5	0.403 ^b	61.19 ^a	1.04 ^a	12.89 ^a	35.14ª
10	0.630 ^a	61.72 ^a	1.07 ^a	13.59 ^a	34.30 ^a
Worm bed leachate (%)				
0	0.506 ^{ab}	62.45 ^a	0.94 ^a	11.52 ^b	35.70 ^a
10	0.360 ^b	60.26 ^a	1.08 ^a	13.16 ^{ab}	29.97 ^b
20	0.662 ^a	61.46 ^a	1.10 ^a	14.47 ^a	35.52 ^a
MSD (p < 0.05)	0.151	2.25	0.15	2.03	3.88

Values with a different letter are significantly different between the treatments (p < 0.05).

RESULTS

Characterization of vermicompost and worm-bed leachate

Vermicompost had pH 8.25, electrical conductivity 1.44 dS m⁻¹, cation exchange capacity (CEC) 98.6 Cmol kg⁻¹ total phosphorus 341.4 mg kg⁻¹, extractable phosphorus 472 mg kg⁻¹, organic matter 20.17% and total nitrogen 132 mg kg⁻¹. The apparent density was 0.69 g mL⁻¹, the humic acid concentration 4.34% and the fulvic acid concentration 3.99% of the total C content of the vermicompost. The humic to fulvic acid ratio was 1.08.

The worm-bed leachate (mean of three samples collected every 8 weeks) had pH 7.5, electrical conductivity 9.8 dS m^{-1} , and total phosphorus 0.916 g L^{-1} and nitrogen content of 15.3 g L^{-1} . Humic acid concentration was 0.07% and the fulvic acid 0.22% of the total C content of the worm-bed leachate. The humic to fulvic acid ratio was 0.32.

Essential oil content

Essential oil content varied between 0.14 and 0.79% in the different treatments (Table 2). The AMF had no significant effect (p < 0.05) on the essential oil content. However, plants amended with 10 g vermicompost plant⁻¹ had 0.13% more essential oil than control plants without addition of vermicompost (Table 3). The plants amended with 20 ml WBL plant⁻¹ had 0.16% more essential oil content than control plants (Table 3). Studying the individual effects showed that higher vermicompost and WBL concentrations resulted in higher essential oils contents (Table 3). Combining those two factors showed that a maximum essential oils content was obtained using 5% vermicompost and 20%

WBL. This could be due to synergistic effect between the nutrients and humic substances of vermicompost and WBL.

The analysis of variance (Table 4) for b coefficients of the model (Equation 2) showed that vermicompost, AMF, AMF-WBL interaction and quadratic term of vermicompost had a significant effect on essential oil content in lemongrass leaves (P < 0.05). The Fisher's F test showed that the model applied was significant. The coefficients of the second-order model calculated with a least square fitting (Equation 2) were:

 $Y = 1.39 - 0.529A + 0.479B - 0.096C - 0.113A^{2} - 0.033AB + 0.066AC - 0.039B^{2} + 0.0007BC + 0.002C^{2}$

[Equation 2]

with Ythe essentials oil content.

Citral, linalool, and myrcene contents

Citral contents varied from 59.7 to 64.0%; linalool from 0.88 to 1.18% and myrcene from 10.14 to 26.4% (Table 2). The AMF and vermicompost had no significant effect on citral, linalool and myrcene contents (Table 3). Worm-bed leachate only had a significant effect on myrcene contents in lemongrass leaves (p < 0.05). The response surface graph indicated that 20% of WBL resulted in maximum citral content (Figure 1b). The analysis of variance (ANOVA) show that β coefficients for WBL had a significant effect on citral and myrcene contents (P < 0.05) but not for linalool content (Table 4). The coefficients of the second-order model for citral (Equation 3), myrcene (Equation 4) and linalool contents (Equation 5) were calculated by least square fitting:

$$Y = 50.34 - 6.06A - 0.81B + 1.23C + 1.83A^{2} + 0.95A * B - 0.27A * C - 0.017B^{2} + 0.004B * C - 0.018C^{2}$$

[Equation 3]

$$Y = 12.42 - 1.83A - 0.52B + 0.32C + 0.61A^{2} + 0.43A * B - 0.19A * C + 0.009B^{2} + 0.009B * C - 0.001C^{2}$$

[Equation 4]

$$Y = 1.02 - 0.21A - 0.012B + 0.02C + 0.067A^2 + 0.013A * B - 0.002A * C + 0.0002 B^2 + 0.0003 B * C - 0.0006 C^2$$

[Equation 5]

Where, Y = citral, myrcene or linalool contents.

The response surface graph indicated that WBL at 20% resulted in maximum myrcene content in the lemongrass leaves (Figure 1c). The ANOVA demonstrated that WBL, AMF-vermicompost and AMF-WBL interaction were significant and Fisher's F test demonstrated that the model applied was significant (Table 4).

Plant yield

The aerial dry weight varied between 25.7 and 44.1 g. Vermicompost and AMF had no significant effect on shoot dry weight, but the effect of WBL was significant (p < 0.05) (Table 3). The response surface graph (Figure 1d) indicated that vermicompost at 5 g plant and AMF at 0.2 g plant resulted in maximum shoot dry weight. The ANOVA for b coefficients of the model (Equation 6) showed that AMF and the quadratic term of vermicompost were significant and the Fisher's F test showed that the model applied was significant (Table 4). The coefficients of the second-order model were estimated (Equation 6).

 $Y = 78.5 + 1.17A + 22.81B + 2.6C - 10.81A^{2} - 0.80AB - 0.70AC - 1.90B^{2} + 0.25BC - 0.10C^{2}$

[Equation 6]

Where, Y was the dry weight.

Table 4. Regression coefficients and variance analysis of the second-order models to determine the effect of variables on essential oil, citral, myrcene, and linalool contents, and aerial dry weight.

		Essential oil content			
Source	Coefficients	Sums of Squares	P-values		
Constant	1.39415				
A:AMF	-0.528722	1.6485	0.0408*		
B:Vermicompost	0.478847	2.12118	0.0214*		
C:worm-bed leachate	-0.0955181	1.4099	0.0575 ^{ns}		
AA	-0.113264	0.142103	0.5370 ^{ns}		
AB	-0.0331667	0.330008	0.3486 ^{ns}		
AC	0.0659	5.21137	0.0006**		
BB	-0.0393272	10.7074	0.0000**		
BC	0.000705	0.0149108	0.8411 ^{ns}		
CC	0.00251653	0.701492	0.1747 ^{ns}		
Total error		12.7945			
R ²	63.93%				
	Citral conter	t			
Constant	50.3418				
A:AMF	-6.06325	2.76828	0.8680 ^{ns}		
B:Vermicompost	-0.814459	0.0791202	0.9776 ^{ns}		
C: worm-bed leachate	1.23962	915.592	0.0044**		
AA	1.83302	37.2181	0.5434 ^{ns}		
AB	0.95845	275.588	0.1039 ^{ns}		
AC	-0.273467	89.7408	0.1033 0.3472 ^{ns}		
BB	-0.273407	2.08539	0.8853 ^{ns}		
BC	0.004105	0.505531	0.8833 0.9434 ^{ns}		
CC		37.712	0.9434 0.5407 ^{ns}		
Total error	-0.0184514		0.5407		
R ²	20.220/	3458.95			
ĸ	28.33%	amt			
Constant	Myrcene cont	ent			
Constant	12.4256	0.00004	0.4570NS		
A:AMF	-1.83862	3.03384	0.4573 ^{ns}		
B:Vermicompost	-0.5274	5.15597	0.3338 ^{ns}		
C: worm-bed leachate	0.322654	52.3301	0.0036**		
AA	0.616417	4.20889	0.3820 ^{ns}		
AB	0.432583	56.1385	0.0027**		
AC	-0.191267	43.8995	0.0071**		
ВВ	0.00914	0.578351	0.7447 ^{ns}		
BC	0.00961167	2.77152	0.4772 ^{ns}		
CC	-0.00158917	0.279742	0.8208 ^{ns}		
Total error		187.914			
R ²	47.09%				
	Aerial weight	dry			
Constant	78.4977				
A:AMF	1.17629	7329.69	0.0074**		
B:Vermicompost	22.8164	79.792	0.7686 ^{ns}		
C: worm-bed leachate	2.59973	1.71735	0.9656 ^{ns}		
AA	-10.8115	1294.76	0.2403 ^{ns}		
AA					
AB	-0.80682	195.288	0.6456 ^{ns}		

BB	-1.90839	25213.6	0.0000**
BC	-0.2561	1967.62	0.1498 ^{ns}
CC	-0.102324	1159.77	0.2660 ^{ns}
Total error		31764.9	
R^2	53.36%		
	Linalool conte	nt	
Constant	1.02791		
A:AMF	-0.216619	0.030317	0.4069 ^{ns}
B:Vermicompost	-0.0127406	0.023375	0.4659 ^{ns}
C: worm-bed leachate	0.0201506	0.167334	0.0565 ^{ns}
AA	0.0669972	0.0497202	0.2897 ^{ns}
AB	0.01355	0.0550807	0.2655 ^{ns}
AC	-0.00206667	0.00512533	0.7320 ^{ns}
BB	0.000253222	0.000443918	0.9197 ^{ns}
BC	0.00029	0.002523	0.8100 ^{ns}
CC	-0.000559194	0.0346374	0.3756 ^{ns}
Total error		1.5054	
R ²	19.55%		

P-values for β coefficients of second-order model. * significant to p < 0.05, ** significant to p < 0.01 and ns not significant.

Desirability function approach (DFA)

The maximal values of shoot dry weight, essential oil, citral, linalool and myrcene contents were calculated with RSM. However, it is necessary to determine a biofertilization protocol related to the optimal responses. The DFA was used to obtain a biofertilization scheme for maximizing essential oil yield and citral content. A value of 1.0 for the DFA is the optimum. Results indicated that optimal values were 2.0 g plant⁻¹, 5.0 g plant⁻¹ and 20% of AMF (*G. mosseae*), vermicompost and WBL, respectively to obtain a desirable value of 0.92 (Figure 1e). As such, lemongrass could yield 0.797% essential oil with 62.6% of citral.

DISCUSSION

Essential oil content

The essential oil content of *C. citratus* in the various treatments varied between 0.14 and 0.79%. Similar high essential oil content, *i.e.* between 0.68 and 0.73%, were reported by Kanko et al. (2004) and Kasali et al. (2001). Linares et al. (2005) obtained 0.40% essential oil in lemongrass using both chemical and organic fertilizer (poultry litter dried Fertipollo®). The essential oil yield increased up to 27% when biofertilizers were applied to lemongrass compared to chemical fertilizer. Serrato-Castillo and Moreno-Berrocal (2003) found that applying decomposed coffee pulp as biofertilizer increased essential oils in lemongrass with 25%. Studying the individual effects showed that higher vermicompost and WBL concentrations resulted in higher essential oils contents (Table 3). Combining those two factors showed that a maximum essential oils content was obtained using 5% vermicompost and 20% WBL. This could be due to synergistic effect between the nutrients and humic substances of vermicompost and WBL.

Citral, linalool and myrcene contents

The quality of C. citratus essential oil was related to its citral content, i.e. a mixture of isomeric aldehydes, neral and geranial. Essential oils with a citral content > 75% are considered of high quality (Combrinck et al. 2011). In our study, citral was the most important component of the essential oil, with concentrations ranging from 59.7 to 64.0%. Another factor that affects the quality of the essential oil of C. citratus is β -myrcene. Oils with β -myrcene are poorly soluble in ethanol and form opalescent solution, this property is a limitation for their use in the perfume industry (Chingin et al. 2008).

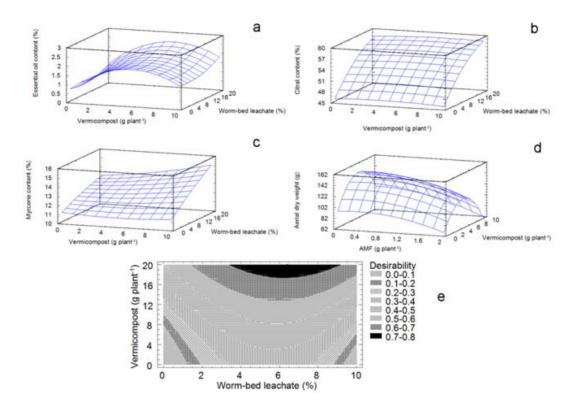


Fig 1. Response surface plot to investigate the effect vermicompost, worm-bed leachate and arbuscular mycorrizic fungi on essential oil content (a), citral content (b) and myrcene content (c), plant growth (d) and desirability response (e) in lemongrass (*Cymbopogun citratus* Stapf.).

Plant yield

Vermicompost can affect plant growth in different ways. Its high nutrient and humic acid contents stimulate plant growth, while its high organic matter improves soil physical and biological properties beneficial for plant development. Humic substances are known stimulators of plant growth (Dell'Amico et al. 1994). They increase membrane permeability, facilitate transport of essential elements within roots, and favour respiration. Canellas et al. (2010) extracted humic acids (HA) from vermicompost and separated them in different fractions. They found that all fractions promoted root growth in *Arabidopsis* and maize seedlings, but the effect depended on the fraction and plant species. In *Arabidopsis*, HA induced an auxin-like exogenous response, *i.e.* the principal root axis was shortened and lateral roots induced, but less biomass was produced, however, in maize the effect of HA resembled a plant with low auxin concentration, *i.e.* principal root elongation and more plant biomass. Lemongrass plants followed the same pattern as the maize plants. The stimulation of root development could be for the H⁺-ATPase activation that was induced by humic acid (Zandonadi et al. 2007). Lateral root development also was induced by humic acid with nitric oxide as a messenger (Zandonadi et al. 2010). Thus, mycorrhizal inoculation has the potential to enhance the production of lemongrass, particularly when farming systems are based on organic inputs (Gaur and Adholeya, 2002).

CONCLUDING REMARKS

Vermicompost increased the essential oil content of lemongrass leaves. On the other hand, worm-bed leachate increased the essential oil content, myrcene and shoot dry weigh of lemongrass leaves. *Glomus mosseae* had no effect on plant characteristics studied. Vermicompost and WBL can be used in organic cultivation of crops with higher plant yield and essential oil content, while conserving the essential oil quality in terms of the citral content.

ACKNOWLEDGMENTS

For Lujan Hidalgo C. for technical assistance for essential oils determination.

Financial support: This research was funded by Dirección General de Educación Superior Tecnológica (DGEST) project "Efecto de la biofertilización de plantas de *Cymbopogon citratus* sobre el rendimiento y composición del aceite esencial clave 3459.10-P".

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How to reference this article:

LEÓN-ANZUETO, E.; ABUD-ARCHILA, M.; DENDOOVEN, L.; VENTURA-CANSECO, L.M.C. and GUTIÉRREZ-MICELI, F.A. (2011). Effect of vermicompost, worm-bed leachate and arbuscular mycorrizal fungi on lemongrass (*Cymbopogon citratus* (DC) Stapf.) growth and composition of its essential oil. *Electronic Journal of Biotechnology*, vol. 14, no. 6. http://dx.doi.org/10.2225/vol14-issue6-fulltext-9