Article

Agronomic components and diversity in the pattern of fatty acids in advanced coriander lines

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Abstract

Cilantro in Mexico is consumed fresh and the cultivated varieties are of foreign origin, and there are no reports of germplasm with high oil content that can be used in the industry. The objective was to evaluate phenotypically, morphologically and biochemically seven advanced coriander lines with desirable agronomic characteristics for production of fresh biomass and oil. Under a design in complete blocks at random the lines L9-CB, L13-CB, L17-CB, L21-CB, L25-CB, 29-CB and INIFAP-17 were sown. The agronomic management was carried out according to Gonzalez et al. (2017). The phenotypic, morphological and biochemical characters in plant and seed were determined. The best material for fresh production was the INIFAP-17 line for its greater number of basal leaves (31), fresh biomass (221 g) and long vegetative period, while for seed production the best lines were L9-CB (1.12 g pta⁻¹) and L13-CB (1.16 g pta⁻¹) are an option. The oil content of the advanced lines analyzed (6.21 to 11.23%) was higher than that reported in the literature (3-5%). Petroselinic acid was the majority with a concentration between 68.95 and 73.51 g 100 g^{-1} of oil, followed by linoleic acid (14.2 - 18.55 g 100 g⁻¹ of oil). From a nutraceutical point of view, coriander oil presented an oleic/linoleic (O/L) ratio of less than 1. It is of interest to identify germplasm with a higher content of linoleic acid because the O/L ratio greater than 1 helps to prevent certain diseases.

Keywords: coriander, fatty acids, fresh biomass, genetic improvement, oil.

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Introduction

Cilantro (*Coriandrum sativum* L) is an aromatic plant native to the eastern Mediterranean region (Ayanoglu *et al.*, 2017) that is grown in many parts of the world. The main producing countries are India, Morocco, Bulgaria, Romania, Canada, China, Syria and Mexico. While Malaysia, Pakistan, Saudi Arabia, United Arab Emirates and England are the main importing countries (FAO, 2016; Sharma *et al.*, 2014). In Mexico, more than 7 000 hectares are planted per year (SIAP, 2017), where Puebla (3 312 ha) and Baja California Norte (1 227 ha) sow more than 60% of the area destined for this crop. All production is destined for the fresh market for both national consumption and export.

During the last five years, the export of fresh cilantro has grown by 100%, with the United States of America being the main buyer (98%), followed by Canada (1.8%) and another 15 countries intermittently (SIAP, 2017). The yield of fresh biomass ranges between 6 and 30 t ha⁻¹, and is mainly associated with the planting season, variety, population density, type of irrigation and fertilization. While low yields are associated with the rustic management of gravity irrigation, manual seeding, low fertilization rates and the use of seed that is harvested.

In contrast, the highest yield is achieved with drip irrigation, certified seed, integrated pest and disease management and a fertilization program supported by soil and plant analysis (González *et al.*, 2017). In Mexico, cilantro is consumed fresh as a spice, so work on its production has focused on the health and yield of fresh biomass. However, information on the production of dry biomass and seed is scarce, so that most of the seed used for planting is imported. The seed is also an underutilized resource that for its oil has economic importance thanks to its medicinal properties (Bhat *et al.*, 2014) and to the use of the seed itself as a condiment in foods including the curry of Indian food (Laribi *et al.*, 2015). The seed has also been used in the treatment of digestive problems such as indigestion, nausea and dysentery (Al-Mofleh *et al.*, 2006; Maroufi *et al.*, 2010).

The oil is used in several fields of the industry for its high content of petroselinic acid (C18:1n-12), (up to 80 g 100 g⁻¹ of oil) (Kleiman and Spencer, 1982, Camilo-Manríquez, 2008), followed by linoleic acid (C18:2n-6). Petroselinic acid is an isomer of oleic acid and can be used in the production of medium chain fatty acids since it can be easily transformed to lauric acid (C12:0) and adipic (C6). The lauric acid is used as a softener, emulsifier and in the soap industry, while the adipic acid is used for the production of high grade plastics (specialized use polymers) (Khodadadi *et al.*, 2016). In addition to the industrial importance of oil, it is also attractive to the pharmaceutical industry; it has been reported to have antimicrobial, antioxidant, antidiabetic, anti-epileptic, antimutagenic, anti-inflammatory and anti-hypertensive properties, among others (Ullagaddi and Bondada, 2011; Sahib *et al.*, 2013). Coriander oil can be consumed as food by healthy adults at a dose of up to 600 mg per day (Agostoni *et al.*, 2013).

In Mexico, the varieties of cilantro that exist in the market are of foreign origin, so, to make the improvement of genotypes of high yield coriander leaf and fruit is highly desirable. At the same time, it is necessary that the new varieties produce quality fruit in terms of the content and balance of the fatty acids contained in the oil (Giridhar *et al.*, 2016). Although the bio-

active compounds of the oil have been characterized as well as the chemical composition, the data are scattered and fragmented. Therefore, it is not only important to develop new materials with outstanding characteristics and properties from the point of view of health and for the industry, it is also necessary to characterize them chemically in order to find their own niche. Based on the above, the objective was to evaluate phenotypically and biochemically seven advanced coriander lines with desirable agronomic characteristics for production of fresh biomass and oil.

Materials and methods

Biological material and agronomic management

Seven advanced coriander lines were planted in August 2017 in the Experimental Bajio field of INIFAP, in Celaya, Guanajuato, located at the geographic coordinates of 20° 32' 05" north latitude and 100° 48' 49" west longitude, at 1 750 meters above sea level, during the fall-winter season, 2017, under the conditions of environmental temperature, precipitation, relative humidity and evapotranspiration that are described (Table 1).

Month	Tempera	ture (°C)	Precipitation	RH means	Eto accumulated	
WOIT	Maximum	Minimum	(mm)	(%)	(mm)	
August	30.2*	11.1	122.4	69.7	148.6	
September	30.7	6.8	91.6	73.3	109.6	
October	29.9	3.5	2	65.4	116.9	
November	30.3	0.6	0	50.9	93.6	
December	28.1	-1	0	51	83.8	
January	26.7	-0.5	0	50.5	84.3	

Table 1. Environmental variables prevailing	n the Bajio Experimental Field during the crop
cycle. Fall-winter, 2017.	

*= data from the National Modeling and Remote Sensing Laboratory, 2018. Bajio Experimental Field, INIFAP. RH= relative humidity; Eto= evapotranspiration.

The experimental lot used was 500 m² with clay loam soil (Table 2). Cilantro was planted under a randomized complete block design with three replications (Table 3). The plantation arrangement was three bolillo with separation between bushes and lines of 25 and 20 cm, respectively. At each planting point, five seeds were placed at 1.5 \pm 0.5 cm depth, to achieve a planting density of 500 000 plants per hectare. The crop was managed according to what was reported by González *et al.* (2017). Drip irrigation was used with 8 000 gauge strip of 16 mm diameter with drippers every 20 cm with flow rate of 1 L h⁻¹. During the crop cycle, a total irrigation sheet of 38 cm (3 800 m³ ha⁻¹) was handled. In the first 20 days after sowing (DDS) irrigation was applied every two days with volumes of 15 to 20 m³ ha⁻¹ to favor the emergence. Subsequently, irrigation management was based on soil moisture tension at 20 centibars, measured with a TDR device placed between the bushes. The fertilization dose (kg ha⁻¹) applied consisted of $160N-70P_2O_5-100K_2O-120Ca-90Mg$. For the control of pests and diseases, organic substances were used as repellents, natural extracts, beneficial microorganisms, among others allowed in organic production (González *et al.*, 2017).

Parameter	Value	Nutriment	Concentration (mg L ⁻¹)	Nutriment	Concentration (mg L ⁻¹)
pН	8.55	Nitrogen	10.55	Iron	17.3
DA (g cm ⁻³)	0.99	Phosphorus	18.42	Zinc	0.42
MO (%)	1.68	Potassium	399.56	Manganese	7.74
CE (dS m ⁻¹)	0.9	Calcium	4209.39	Copper	1.04
		Magnesium	792.65	Boron	0.11
		Sodium	1066.81		

Table 2. Condition of experimental soil fertility before establishing cilantro cultivation. BajioExperimental Field, 2017.

Table 3. Advanced coriander lines obtained by the mass selection method and stabilized during eight cycles.

Num.	Accession	Origin [†]	Latitude [‡]	Longitude	Elevation *
1	L9-CB	Mextiquic de Carmona, San Luis Potosi	22°15'03"	101°07'36"	2 050
2	L13-CB	Tekax, Yucatan	20°12'42"	89°16'34"	37
3	L17-CB	Tizimin, Yucatan	21°08'10"	88°08'44"	21
4	L21-CB	San Juan Teposcolula, Oaxaca	17°33'03"	97°25'29"	2 302
5	L25-CB	San Juan Teposcolula, Oaxaca	17°33'03"	97°25'29"	2 302
6	L29-CB	Ixtacuixtla de Mariano	19°19'31"	98°22'44"	2 282
7	INIFAP-17	Matamoros, Tlaxcala Roque, Celaya, Guanajuato	20°35'50"	100°54'18"	1 760

[†]= place where the original material was collected; [‡]= estimated geographic coordinates based on the place of collection; ^{*}= meters above sea level.

Characters evaluated

The phenological, morphological and biochemical characteristics were evaluated (Table 4) based on what was suggested by Diederichsen and Hammer (2003); González *et al.* (2017). The results of each character were the average of four individual plants chosen at random within each population.

Character *	Description
Phenological	
Days to emergency (DE)	Days from sowing until the appearance of more than 50% of plants.
Days to emission of floral scape (DEEF)	Days from sowing until the appearance of the floral scape in more than 50% of plants.
Days between emergence and emission of the floral scape (DEEEF)	Days between the emergence and the appearance of the floral scape.
Days at the beginning of	Days between planting date and the beginning of
flowering (DCF)	flowering in each plant.
Days to seed harvest (DCS)	Days from harvest to harvest day.
Morphological	
Height of the plant in cm (AP)	Measure from the surface of the soil to the apex of the highest leaf.
Number of basal leaves (NHB)	Total, of basal leaves counted until the release of the escape.
Number of umbels (NU)	Total, of umbels counted in the harvest.
Production of fresh biomass per plant (BFP)	Weight of the aerial part determined at the end of the vegetative period.
Seed production per plant in g (PSP)	Estimated average weight of four plants.
Weight of 1000 seeds in g (P1000S)	Estimated weight of four repetitions of 1000 seeds.

Table4.	Agronomic	importance	characters	evaluated	in	seven	advanced	coriander	lines.
	Autumn-win	nter, 2017.							

*= characters based on Diederichsen and Hammer (2003); González et al. (2017).

Oil content and fatty acid profile

The oil content in the samples was determined with the 920.85 method of the AOAC (2000), while the fatty acid profile was determined by the proposed Agilent Industries method for the mass-coupled gas chromatograph (AOCS, 2013) (Agilent Technologies, Inc. Santa Clara CA, USA, Models 6890N and 5973). An HP-88 column (100 m x 0.25 mm ID, 0.2 μ m, 250 °C, 1 μ L injection volume) was used, with hydrogen as carrier gas A and helium as carrier gas B, with a constant flow of 2 mL min⁻¹ and oven conditions A: 120 °C for 1 min, 10 °C min⁻¹ at 175 °C for 10 min, 5 °C min⁻¹ at 210 °C for 5 min, 5 °C min⁻¹ at 230 °C for 5 min and from oven B: 175 °C for 10 min, 3 °C min⁻¹ at 220 °C for 5 min.

The gas flow in the detector was of hydrogen 40 mL min⁻¹, of air 450 mL min⁻¹ and of helium 30 mL min⁻¹. The temperature detector was at 280 °C. A standard of a mixture of 21 fatty acids (Sigma) at a known concentration was used to identify and quantify fatty acids. The mixture was injected into the chromatograph to generate a pattern of peak height (concentration) and retention time of each fatty acid. The peaks of each sample were compared with the retention

times for their identification, and with the height of the peak for their quantification by extrapolating with the algorithm provided by the program of the equipment itself (ChemStation). The data obtained were subjected to an analysis of variance (Anova) and comparison of means by Tukey ($p \le 0.05$) with the statistical package SAS (SAS, 2008).

Results and discussion

Phenotypic and morphological diversity

The behavior of the coriander lines showed significant differences in some phenological characters ($p \le 0.05$). The emergency was presented between 12 and 15 days after sowing (DDS). For the DEEF, line INIFAP-17 was late at the beginning of the emission of the floral scape at 86 days, while the other lines started this stage at 58 DDS. The precocity shown by these lines leaves a margin of five days for the fresh harvest, unlike the INIFAP-17 line, which showed a margin of 25 days (Table 5). The period of vegetative growth as well as the days to emergence (DE) and days to emission of the floral scape (DEEF) was higher in the line INIFAP-17, followed by the L17-CB.

The beginning of flowering in the early lines started at 78 DDS and in the late line 20 days later. Seed harvest in early lines was at 100 DDS, intermediate at 105 DDS and late at 125 DDS. It was observed that the INIFAP-17 line presented a higher cycle compared to the rest, which are early and intermediate lines, this line can be used for fresh production, while the early ones are suggested for areas with temperate climate and for production of oil.

In studies carried out in cilantro it has been determined that the precocity of the materials is determined by the emission of the floral scape or punctation which is influenced by the photoperiod, the temperature and the change in the endogenous hormonal concentration (González *et al.*, 2017). Bashtanova and Flowers (2011), when evaluating 90 accessions of cilantro from the Caucasian region, found that the vegetative period of cilantro lasts from 31 to 60 days (from germination to emission of the floral scape) and that time is specific for each genotype, but in general all materials behave the same when grown in winter. On the contrary, the vegetative period in summer lasts from 22 to 45 days because it is affected by the length of the day (light hours) and temperature. Ghobadi and Ghobadi (2010), mention that during the first 40 DDS the growth is slow and that between 40 and 60 DDS the speed of growth increases and that after 60 DDS is slow again. Similar results were found in the lines with short crop cycle (early) (Table 5).

The materials evaluated can be classified into sensitive genotypes with the exception of the INIFAP-17 line, which can be considered a neutral genotype according to the classification proposed by Bashtanova and Flowers (2011). The results obtained in the present work show two events in the senescence of the plants: when the rosette matures completely and when the flowering finishes. These results could indicate independent events and show a relation by the change in vegetative stages during the ontogeny of the plant (Guiboileau *et al.*, 2010; Bashtanova and Flowers, 2011).

Character	L9-CB	L13-CB	L17-CB	L21-CB	L25-CB	L29-CB	INIFAP-
							17
Days to emergency (DE)	12 *‡ a	12 a	14 a	12 a	12 a	12 a	15 a
Days to emission of floral scape (DEEF)	58 a	58 a	65 b	58 a	58 a	58 a	86 c
Days between emergence and emission of the floral scape (DEEEF)		36 a	51 b	36 a	36 a	36 a	71 c
Days at the beginning of flowering (DCF)	78 a	78 a	84 b	78 a	78 a	78 a	98 c
Days to seed harvest (DCS)	100 b	100 b	105 b	100 b	100 b	100 b	125 ab
Height of the plant in cm (AP)	51.6 c	71.1 a	53.5 c	53.7 c	58.4 bc	63.4b	76.1 a
Number of basal leaves (NHB)	17 c	17 c	24 b	26 b	19 bc	24 b	31 a
Number of umbels (NU)	14.2 a	14 a	21 b	16 a	14 a	14 a	16 a
Production of fresh biomass (g)	91.8 c	91.5 c	141.6 b	149.2 b	98.3 c	139.8 b	221.7a
Seed production per plant in g (PSP)	1.12 a	1.16 a	0.57 bc	0.68 b	0.78 b	0.77 b	0.97ab
Weight of 1000 seeds in g (P1000S)	10.7 b	9.7 b	14.1 a	11.2 b	10.5 b	10.9 b	11.2b

Table 5.	Comparison	of	agronomic	characters	in	seven	advanced	coriander	lines.	Fall-winter,
	2017.									

*= averages within the row followed by the same letter are not statistically different Tukey ($p \le 0.05$); \ddagger = mean values of three repetitions.

Regarding the morphological variables, significant differences were found ($p \le 0.05$), since the highest plant height (AP) was registered in the INIFAP-17 and L13-CB lines with 76.1 and 71.1 cm, respectively (Table 5). Similar results were reported by Ayanoğlu *et al.* (2002) who when evaluating 43 coriander lines recorded AP from 68.3 to 127 cm and Ghobadi and Ghobadi (2010) from 69.6 to 77.7 cm in planting densities of 70 ptas m⁻². While Hnamte *et al.* (2013) in coriander grown in winter reports AP from 40.8 to 63.9 cm, values below those obtained in the present work.

The highest number of basal leaves (NHB) was shown by the INIFAP-17 line with 31 basal leaves, followed by the L17-CB, L21-CB and L29-CB with 25 basal leaves on average each. This number of leaves is greater than those reported by Ayanoğlu *et al.* (2002) that in 43 lines of coriander registered a maximum of 8.7 basal leaves. The largest number of umbels (UN) was recorded in L17-CB with 21 umbels, while the rest presented 15 umbels on average. These results are within the values reported by Ayanoğlu *et al.* (2002) (5.8 to 27.4 umbels per plant) and Ghobadi and Ghobadi (2010) (6.7 to 50.7 umbels per plant).

In relation to fresh biomass (BF), significant differences were observed ($p \le 0.05$) since, as expected, line INIFAP-17 recorded the highest accumulation of BF with 221.07 g pta⁻¹, followed by L17-CB, L21-CB and L29-CB that on average obtained 35.4% less than BF. While the other lines L13-CB and L25-CB recorded less than 100 g pta⁻¹ of BF (Table 5). Cruz *et al.* (2017) when evaluating different concentrations of NPK in the variety Pakistan cultivated in hydroponics record 155 to 325 g of BF after 90 days, values above those found in this study.

The differences with the results of the present work can be explained by the number of seeds per pot (3 seeds germinated per pot) used by Cruz *et al.* (2017), which on average produced between 51.6 and 108 g pta⁻¹ of BF, while in this work there was 91.5 to 221.7 g pta⁻¹ of BF, which would mean higher production of BF on the INIFAP-17 line. On the other hand, Rashed and Darwesh (2015) report values of 355 to 860 g of BF in autumn-winter, values higher than those reported in this study, but influenced by the date of sowing and by the morphological characteristics of the genotypes used and number of seeds per bush (between 6 and 8 seeds).

In seed production per plant (PSP) there were no differences between the lines ($p \le 0.05$), but the highest production was recorded in L9-CB and L13-CB with 1.12 and 1.16 g pta⁻¹, respectively, while the L17-CB recorded the lowest seed production with 0.57 g pta⁻¹ (Table 5). The highest weight of 1 000 seeds was registered in the L17-CB with 14.1 g, followed by the L21 and INIFAP-17 with 11.2 g, while the other lines presented an average of 10.2 g. Similar results were reported by Ghobadi and Ghobadi (2010) who, when evaluating different genotypes, registered a weight of one thousand seeds from 9.36 to 10.5 g while Ayanoğlu *et al.* (2002) indicate weights of 1 000 seeds from 7.05 to 17.55 g, values higher than the one found in this study.

Oil content

The oil content of the fruit of the materials under analysis was presented in the range of 6.21 to 11.23% in lines L17-CB and L25-CB, respectively (Table 6). The oil levels found are higher than reported by the literature; For example, Camilo-Manriquez (2008) reported a 3-3.1% content of coriander purchased in the Santiago de Chile market. Khodadadi *et al.* (2016) reported oil levels of up to 5% in unhusked fruit and up to 40% in peeled fruit. Comparing these values of oil content with what was found, there are apparently promising advanced lines regarding the oil content. However, Nguyen *et al.* (2015) report that the content of oil in coriander increases significantly depending on the time that the fruit remains in the plant, 10 days after flowering the oil content was 10.5% while at 53 days it was 25.1%.

These authors did not perform any tests to evaluate the fatty acid profile and other characteristics of the oil as was done in this study. The seed used in this work was harvested after 100 DDS (Table 6).

Line	Oil content (%)
L9-CB	$6.95 \pm 0.05^* \mathrm{f}$
L13-CB	10.21 ±0.055 b
L17-CB	6.21 ±0.005 g
L21-CB	9.24 ±0.06 c
L25-CB	11.23 ±0.1 a
L29-CB	7.06 ±0.08 e
INIFAP-17	7.73 ±0.15 d

Table 6. Oil content of six advanced coriander lines.

*= means within column followed by same letter are not statistically different Tukey ($p \le 0.05$).

Fatty acid profile

In this work, the myristic, parlmitic, petroselinic, oleic and linoleic fatty acids were detected. Other peaks were observed that could not be detected based on the commercial standards that were used (Table 7). Of these, up to 80% were saturated (myristic, palmitic and petroselinic) (L13-CB) and the rest unsaturated (oleic and linoleic) (Table 7). The major fatty acid was petroselinic with a range of between 68.95 (INIFAP-17) and 73.51 mg 100 g⁻¹ of oil (L13-CB) followed by linoleic acid with a content of up to 17.07 g 100 g⁻¹ of oil (L25 -CB). The high content of petroselinic acid is normal in oil of this species; contents of this acid have been reported in cilantro of between 65.7 to 80.9 g 100 g⁻¹ of oil, followed by linoleic acid (13.05 - 16.7 g 100 g⁻¹ of oil) (Ramadan and Mörsel, 2002; Msaada *et al.*, 2009; Sriti *et al.*, 2009; Sriti *et al.*, 2009; Sriti *et al.*, 2010).

As can be seen in Table 7, the levels of both fatty acids reported here are within the range indicated by the literature, with the exception of sample L25-CB, which presented 17.07g 100 g⁻¹ of linoleic acid oil. Coriander oil can have different uses in addition to those indicated here. For example, diets supplemented with petroselinic acid given to fish increased the content of polyunsaturated fatty acids, improving the nutraceutical quality of meat (Teoh and Ng, 2013).

Line	Myristic	Palmitic	Petroselinic	Oleic	Linoleic	O/L
L9-CB	$2.96 \pm 0.065^{*} c$	$2.68 \pm 0.012 \text{ f}$	72.34 ±0.545 b	6.76 ±0.05 d	15.93 ±0.025 d	0.42
L13-CB	$3.46 \pm 0.05 \text{ b}$	3.68 ±0.015 e	73.51 ±0.152 a	$5.2 \pm 0.012 \text{ f}$	14.2 ±0.011 g	0.37
L17-CB	2.42 ±0.03 e	3.2 ±0.017 g	71.44 ±0.51 c	7.88 ±0.011 b	$15.05 \pm 0.024 \text{ f}$	0.52
L21-CB	3.82 ±0.075 a	4.45 ±0.11 b	$69.84 \pm 0.458 \text{ d}$	5.33 ±0.027 e	16.57 ±0.025 c	0.32
L25-CB	2.61 ±0.061 d	3.94 ±0.045 c	69.6 ±0.451 de	7 ±0.016 c	17.07 ±0.051 a	0.41
L29-CB	$2.35 \pm 0.01 \ f$	$3.84 \pm 0.024 d$	69.27 ±0.314 e	8.75 ±0.019 a	15.8 ±0.015 e	0.55
INIFAP-17	2.36 ±0.015 f	4.88 ±0.101 a	$68.95 \pm 0.147 \text{ f}$	6.88 ±0.013 d	$16.94 \pm 0.045 \text{ b}$	0.41

Table 7. Fatty acid profile (mg 100 L⁻¹ oil) of seven advanced coriander lines.

*= means within column followed by same letter are not statistically different Tukey ($p \le 0.05$).

From the nutraceutical point of view, in addition to the high content of saturated oils, it has been shown that when an oil contains an oleic/linoleic (O/L) ratio close to or greater than 1, it can have positive effects on health by reducing the possibility of suffering from cancer (Parry *et al.*, 2005, Martin *et al.*, 2011) and heart disease (Parry *et al.*, 2005). In this sense, the oils of the samples analyzed had an O/L ratio of less than 0.56, so it is presumed that their consumption does not contribute to the reduction of these diseases. However, in this apparently negative aspect, an area of opportunity is presented in relation to increasing the oleic content in cilantro oil. The immediate option is to look for varieties or lines whose oleic content allows the O/L ratio to be as close as possible to the value of 1, or to seek with the management of the crop to promote the increase of the oleic. In this regard, a very wide range was observed in the oleic acid content among the materials studied (5.2-8.75%) (Table 7).

Conclusions

From the study, INIFAP-17 was identified as the line with potential for the production of fresh biomass. This line has morphological attributes such as the greater number of basal leaves that allow the greater production of fresh biomass, in addition to having a longer vegetative cycle which gives the field greater shelf life, although its oil content is less than of other lines. For oil production lines L17-CB and L29-CB are an option. Future studies with the lines that showed the highest content of oleic oil and linalool should be made to obtain germplasm with industrial potential since it is of interest in the care of human health.

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