

Antioxidant activity of two varieties of *Ocimum basilicum* L. for potential use in phytocosmetics



Actividad antioxidante de dos variedades de *Ocimum basilicum* L. para uso potencial en fitocosmética

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ABSTRACT

Key words:

Antioxidant capacity Ocimum basilicum L. var. cinammom var. album Volatile oils This investigation aimed to evaluate two varieties of *Ocimum basilicum* L., known as Basil, as potential raw material for the cosmetic industry, assessing their antioxidant properties, considering their industrial use in phytocosmetics. The antioxidant activity of essential oils (EOs) for the species *Ocimum basilicum* var. *cinammom* and var. *album*, were obtained by distillation steam using a Clevenger-type device. The antioxidant capacity was evaluated by the method of bleaching radical 1,1-diphenyl-2-picryl hydrazyl (DPPH) and the method of linoleic acid peroxidation (ferric thiocyanate). The EOs of the two species had significant antioxidant properties. The method of DPPH facilitated the evaluation of the antioxidant capacity versus the concentration of EOs, showing an efficient concentration at 10 ppm. On the other hand, the ferric thiocyanate method displayed an efficient inhibition up to 360 h (15 d). The obtained results demonstrated the antioxidant capacity of EOs in the investigation. The capacity was related to their chemical composition (phenylpropane and oxygenated monoterpenes). Therefore, EOs can be considered as a potential source in the field of phytocosmetics.

RESUMEN

Palabras claves:

Capacidad antioxidante Ocimum basilicum L. var. cinammom var. album Aceites volátiles El presente estudio tuvo como finalidad valorar dos variedades de *Ocimum basilicum* L.; conocida con el nombre de Albahaca, como materia prima potencial para la industria cosmética, evaluando su propiedad antioxidante, con miras a un aprovechamiento industrial en fitocosmética. La actividad antioxidante de los aceites esenciales (AEs) de las especies *Ocimum basilicum* var. *cinammom* y var. *album*, se obtuvieron por destilación de arrastre de vapor tipo Clevenger. La capacidad antioxidante se evaluó por el método de la decoloración del radical 2,2-difenil-1-picril hidrazilo (DPPH) y el método de peroxidación del ácido linoleico (tiocianto férrico). Los AEs de las dos especies en estudio, presentan propiedades antioxidantes representativas. El método del DPPH permitió evaluar la capacidad antioxidante frente a la concentración de los AEs, demostrando una concentración eficiente a 10 ppm. Con el método del tiocianto férrico se encontró un máximo de inhibición eficiente a las 360 h (15 d). Los resultados obtenidos demuestran la capacidad antioxidante de los AEs en estudio, capacidad que está relacionada con la composición química (fenilpropanos y monoterpenos oxigenados) y que permite considerar los AEs en estudio como fuente potencial en el campo de la fitocosmética.

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he country of Colombia counts with a great biological diversity. Nevertheless, the richness and utility of the different species is at some point unknown. It is considered that there are about 250,000 vegetal species in the world, which around 80,000 of them can be found in Latin America. An approximate of 40,000 species are found in Colombia, Brazil and Peru (Forero, 1987). An approximate of 5,000 of these species have been used by indigenous communities and local farmers to treat the wide range of diseases and in facial, body and decoration activities. As a result, Colombia has a huge potential as a source for new active ingredients that can be used as a therapeutic and cosmetic alternative (Fonnegra and Jimenez, 2007).

In Risaralda, some of the most harvested species are certain varieties of basil (*Ocimum basilicum* L.) such as the *O. basilicum* L. var *cinammom* and the var *album*. The little information found in relation to the use of these species regards to traditional uses in order to stimulate the scalp which evolves in the growth of hair. This property is attributed mainly to the white basin *Ocimun basilicum* L (Atehortua, 1992; Chaves and Arango, 1998).

The cosmetics industry has been continuously seeking for phytoingredients as bioactive components. Fields such as the phytocosmetics apply ingredients taken from plants for the preparation of all types of cosmetics as a way to reduce the use of chemical substances in cosmetology. These natural raw materials are starting to be more implemented, especially when there is report of adverse reactions that are provoked by chemical substances used in cosmetology (Álvarez and Bague, 2012). The particular characteristics of the phytoingredients are given by the diverse metabolites of plants, and the pharmacological and ethnobotanical laboratories are the ones who weigh and analyze the different phytochemicals that are suitable for cosmetic usage. Some of the most used phytoingredients in the phytocosmetics are the essential oils (Ferraro et al., 2012).

The concentration of primary and secondary metabolites of the phytoingredients is not uniform in all the plant's cycle of life. It varies according to intrinsic and intrinsic factors such as geographical factors, weather, harvest time, the part of the plant that is being used, origin, and postharvest treatment among others (Ferraro *et al.*, 2012). In regard to the species of this investigation (Ocimum basilicum), it belongs to the Lamiaceae family. It is one of the most used families in the world as a source of spices and extracts with antibacterial and antioxidant properties (Hirose et al., 1986). The interest in the investigation of antioxidant activity of EOs species basil (O. basilicum) and extracts of different polarities has grown (Beltrán et al., 2010; Fernández et al., 2007). A preliminary investigation on the chemical composition, conducted to the same plant varieties that were collected in the same zone, found that EOs of O. basilicum L. var. cinammom and var. album. reported high content in phenylpropane and oxygenated monoterpenes. The var. cinammom presented as major components: Eucaliptol (24.06%) and eugenol (37.60%), and the var. album: methyl (E)-cinnamate (28.20%) and linalool (18.16%) (Beltrán et al., 2010).

The EOs which are rich in monoterpenes are called monoterpenoid EOs (eg, peppermint, basil, sage, etc.). The EOs which are rich in phenylpropane are called phenylpropanoid EOs (eg, cloves, cinnamon, anise, etc.) (Rojas et al., 2008; Mahecha, 2010). Different investigations have identified phenylpropane as compounds with significant biological activity. The high amount of methyl (E)-cinnamate, eugenol and linalool, are characterized by antimicrobial, antifungal, antiseptic and antioxidant activity (Reyes et al., 2007; Romero et al., 2004). Given the chemical complexity of AEs, antioxidant activity assay results can display dispersed results depending on the method used. Therefore, it is advisable an approach with multiple attempts (Granados et al., 2012).

Antioxidants are compounds which may inhibit or retard oxidation of other molecules, inhibiting the initiation and/or propagation of chain reactions of free radicals, preventing the formation of undesirable colors and flavors. This is why they are added to cosmetic products and food for human and animal consumption (Maestro and Borja, 1993; Murillo *et al.*, 2007; Muñoz and Gutiérrez, 2009). Artificial antioxidants are widely used in industry. However, due to the carcinogenicity of these synthetic antioxidants, a growing interest on natural antioxidants has arose. These natural antioxidants can be found in plant products, since the presence of various chemical compounds gives them the property to act as anti-radical or antioxidant. This capacity has been demonstrated at a laboratory level and it is now widely researched and is a global trend in preference consumer (Muñoz and Gutiérrez, 2009; Granados *et al.*, 2012; Rincón *et al.*, 2011).

Natural products have shown the presence of phenolic compounds (tocopherols, flavonoids and phenolic acids), nitrogen compounds (alkaloids, chlorophyll derivatives, amino acids and amines) and carotenoids which have antioxidant properties. These features have favored its inclusion in cosmetic formulations (Muñoz and Gutiérrez, 2009; Rincón et al., 2011). In order to prevent the accumulation of free radicals, the body has different mechanisms of antioxidants defense produced endogenously. Nevertheless, as one ages and/or conditions of strong prooxidatives aggressions preveal, such as intense and repeated exposure to sun rays, these mechanisms start to be insufficient to protect the body. Based on this fact, it has been evidenced that organisms age because the cells accumulate the damage of free radicals in time, so that antioxidants are commonly used in skin care to prevent this aging (Rincón et al., 2011).

Natural antioxidants in the field of cosmetics are increasingly used for their ability to cancel or reduce these oxidative processes carried out uncontrollably into the skin tissue. The skin is the most exposed organ to the environment. It is especially vulnerable to damage caused by the free radicals. Whereby, it has a number of requirements for maintenance. If these needs are met satisfactorily, it will show a healthy skin. Whereas if this does not happen, its structure and metabolic activity can be compromised, causing a rough, prematurely aged and dull skin (Gajardo *et al.*, 2011).

There are diverse antioxidants which are implemented in the treatment of problems caused by free radicals. They are capable of damaging the connective tissue, cell membranes and the DNA. The collagen is the most abundant fibrous protein of the connective tissue. It is responsible for the maintenance of texture and elasticity of the skin. The polyphenols are capable of reactivating the harmed collagen and of protecting it from the attack of free radicals and the enzymes (elastase, collagenase). They attach to the fibers of collagen and help to rebuild the connecting links that are damaged by the free radicals. As a result, there is an improvement in the skin flexibility. The tissues with mayor catchment (affinity) are the richer in glycosaminoglycans. This implies that there is activity at epidermal basal level. The damages produced in the skin by these unstable molecules are premature aging and cancer. In general, all the polyphenols have antioxidant capacity (Ferraro *et al.*, 2012).

O. basilicum L. (basil), having the chemical composition polyphenols (phenylpropane) presents a high biological activity as a natural antioxidant that could be considered as ingredients in the field of phytocosmetics (Muñoz and Gutiérrez, 2009; Rincón *et al.*, 2011). There are various methods to evaluate antioxidant activity, either *in vitro* or *in vivo*. One of the most implemented strategy in the *in vitro* measurement in the total antioxidant capacity of a compound, mixture or food; involves determining the antioxidant activity against chromogenic substances of radical nature. Color loss occurs in proportion to the concentration. However, the determinations of the antioxidant capacity *in vitro* only gives us a rough idea of what happens in complex situations *in vivo* (Madrigal *et al.*, 2013).

MATERIALS AND METHODS Plant material

Plant species were worked: *O. basilicim* var. *cinammom* and var. *album*, regionally known as cinnamon basil and white basil, respectively. These plants were obtained in La Florida, in the municipality of Pereira, Risaralda department (Colombia) at an altitude of 1440 m and average temperature of 27 °C. The plant species worked, were characterized taxonomically in previous investigations (Beltrán *et al.*, 2010).

Treatment of plant material

Once the fresh plant material was collected, it was dried at a room temperature (25 $^{\circ}$ C) for 8 days. Some leaves and flowers that were considered healthy were selected and a reduction in size was performed by a hand mill in order to increase the contact surface (Lima, 2005). The content of moisture of the vegetal material was determined by the AOAC 934.91 method.

Distillation of EOs

The extraction of the EOs was done by steam distillation using a Clevenger-type device. The process was

conducted with 100 g of plant material of grounded vegetal material and they were subjected to hydrodistillation with 500 mL of distilled water for 3 h. After obtaining the EOs, they were dried with anhydrous sodium sulfate and finally were stored in airtight containers protected from light and under refrigeration at 4 °C until use. The percent yield of extraction was calculated with the amount of EOs obtained (Murillo *et al.*, 2004).

The EOs sensory characteristics (appearance (oily), color (bright light yellow) and odor (similar to the raw material)) were determined according to the NTC 3925, consisting of an assessment of the samples through the sensations perceived by the sense organs (ICONTEC, 1996).

Determination of antioxidant capacity

It was evaluated by two spectrophotometric methods, both procedures were performed in duplicate and all reagents were analytical grade.

DPPH method

The method was performed according to Cotelle *et al.* (1996). The free radical DPPH method reduces the radical 2,2-diphenyl-1-picrylhydrazyl (DPPH) in 2,2-diphenyl-1-picryl-hydrazine by the antioxidant action of compounds containing -OH groups that discolor the reagent DPPH. The color changes from purple to yellow after reduction, can be quantified by the decrease in absorbance at 517 nm (Muñoz and Gutiérrez, 2009; Rincón *et al.*, 2011; Delgado *et al.*, 2010).

Ascorbic acid (vitamin C) was used as the reference standard (Carhuapoma *et al.*, 2005). A solution standard of ascorbic acid 1000 ppm (100 mg in 100 mL ethanol) was prepared and dilutions of 50, 30, 20, 18, 16, 16, 14, 12, 10 and 8 ppm.

From the EOs of *O. basilicum* var. *cinnamon* and var. *album*, four dilutions in ethanol: 10, 20, 30 and 50 μ L mL⁻¹ were prepared.

DPPH solution was prepared at a concentration of 100 mM in ethanol, taking 3.9 mg of DPPH in 100 mL of solvent (Fernández *et al.*, 2007; Delgado *et al.*, 2010).

The neutralization reaction of the radical was carried out in test tubes. To each test tube, 250 μ L of dilutions

of ascorbic acid and EOs was added. Subsequently, 750 μ L of the solution of DPPH radical (as white was used DPPH solution without sample) were added. The reaction was stirred at room temperature for 30 minutes. Finally, the absorbance was measured (Kuskoski *et al.*, 2005; Delgado *et al.*, 2010).

The antioxidant activity is expressed as percent inhibition, which corresponds to the amount of DPPH radical neutralized by the extract at a concentration determined according to the equation 1 (Muñoz and Gutiérrez, 2009):

$$\%Inhibition = \frac{(A - A_1)}{A} * 100 \tag{1}$$

A = Control absorbance $A_1 = Sample absorbance$

Ferric thiocyanate method

Based on a linoleic acid peroxidation, using ammonium thiocyanate and ferrous chloride (Rincón *et al.*, 2011). A reference standard α -tocopherol (vitamin E) solution in ethanol of 1000 ppm was used (Murillo *et al.*, 2007). It were prepared simultaneous samples of α -tocopherol pattern diluted in ethanol to 10 ppm, samples of EO *O. basilicum* var. *album* and var. *cinammom* to 40 ppm and control samples (Murillo *et al.*, 2007).

According the method adapted by Osawa and Namiki (1981), 1 mL sample pattern (10 ppm) and EOs (40 ppm) to a test tube cap screw were transferred individually, which were dissolved in 4 mL of ethanol 95%, then 4.1 mL of linoleic acid (2.51% v/v in ethanol 96%), 8 mL of 0.05 M phosphate buffer (pH 7.0) and 3.9 mL of distilled water were added. This solution was incubated at 40 °C temperature in the dark (Murillo *et al.*, 2007; Huang *et al.*, 2005; Solanilla *et al.*, 2011).

From this solution, 0.1 mL was taken daily and 9.7 mL of ethanol (75% v/v) and 0.1 mL of ammonium thiocyanate (30% w/v) were added. To this reaction mixture 0.1 mL of ferrous chloride (20 mM) in hydrochloric acid (3.5% v/v) was added, and exactly 3 min later, the absorbance of the resulting mixture (Fe(SCN)₃, red color) was read at 500 nm. The reading was repeated every 24 h for 30 d to evaluate the behavior over time (Huang *et al.*, 2005).

The percentage of inhibition of peroxidation of linoleic acid was calculated according to the equation 2 (Huang *et al.*, 2005; Solanilla *et al.*, 2011).

$$\%Inhibition = 100 - \left(\frac{A_1}{A} * 100\right)$$
(2)

 $A_1 =$ Sample absorbance at 500 nm

A = Control absorbance at 500 nm

RESULTS AND DISCUSSION

The worked ground plant material reported moisture content of $12.63 \pm 0.34\%$ for the var. *cinamom* and $13.31 \pm 0.26\%$ for the var. *album*. High moisture content can affect negatively the extraction or alter the chemical quality of the oil, increasing acidity. A high acidity, according to the purity of the oil, indicates that there has been alteration causing changes in the aroma and flavor (Martínez, 2010; Zumbado, 2004).

The extraction yield was completed in triplicate by steam distillation Clevenger – type device, resulting in a percent yield of $0.324 \pm 0.06\%$ for var. *cinamom* and $0.158 \pm 0.01\%$ for var. *album*. The extraction yield percentages of EOs vary according to the plant, and are generally

low values. Research indicates that basil contains EOs between 0.04 to 0.7%, and are responsible for its aroma and flavor (Díaz, 2010).

The organoleptic characteristics of EOs extracted from the var. *cinammom* and var. *album* displayed a bright light yellow and oily aspect and with a similar odor to the raw material. The vast majority of plant essences are colorless. Some have colorations modifiable by oxygen in the air. The observed modifications in color were caused by the action of light, which implies the presence of aliphatic compounds with little unsaturations or monoannular aromatics (Murillo *et al.*, 2004).

The antioxidant activity of ascorbic acid at different concentrations in relation to the DPPH indicated that at higher doses, the concentration of the radical 2,2-diphenyl-1-picrylhydrazyl decreases, which is reduced and decolourised by the action of ascorbic acid. This reaction is recorded by the decrease in absorbance; therefore, a greater percentage of inhibition. The inhibition of ascorbic acid to 30 ppm was 96.8%, and almost complete inhibition. Consequently, the increase to 50 ppm was more stable, indicating a value of 97.3 \pm 0.24% (Figure 1).

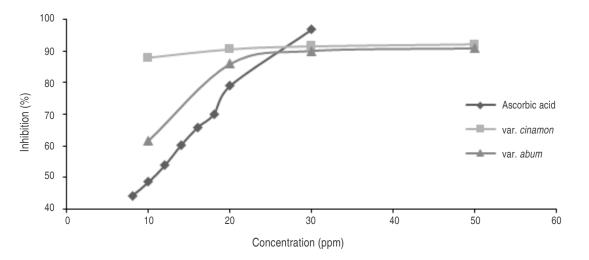


Figure 1. Evaluation of the antioxidant activity by DPPH method.

It was found that the higher the concentration of EOs of *O. basilicum* var. *cinammom* and var. *album*, the higher the inhibition activity, reflected by the increase of the inhibition percentage.

The EOs of *O. basilicum* var. *cinammom* presented a inhibition of 87.9% at a concentration of 10 ppm, an inhibition of 91.4% at 30 ppm and 92.1% up to 50 ppm. EOs of *O. basilicum* var. *album* obtained a 61.8% inhibition at 10 ppm and 90% inhibition at 30 ppm. Inhibition remained constant at 50 ppm.

The above data verify that by the DPPH method the antioxidant activity is dependent on the concentration of extract (Kuskoski *et al.*, 2005). This behavior was shown by the ascorbic acid, indicating that a percentage of inhibition increased until a concentration of 30 ppm. It was also manifested by samples of the EOs, where the % inhibition increased to 30 ppm, and where the inhibition concentration began to stabilize.

The antioxidant activity when implementing the DPPH method, allowed us to observe that the EOs of *O. basilicum* var. *cinammom* presents greater % inhibition (87.8%) at lower concentration compared to the EO of var. *album* (61.5%). Although both EOs reached similar inhibitions at higher concentrations (30 and 50 ppm), the greatest potential inhibition at 10 ppm of EO var. *cinammom* indicates that lower concentration of EO is required to

take free radicals; hence, it proves to be more efficient as antiradical (Rincón *et al.*, 2011).

The antioxidant potential of EOs was complemented assessing their ability to inhibit the oxidation of linoleic acid. In this case, the same concentration of α -tocopherol (10 ppm) and EOs (40 ppm) was used, evaluating their behavior over time. It was assessed for a total of 720 h (30 d).

Through this method, EOs of *O. basilicum* var. *cinammom* and var. *album* inhibited the linoleic acid peroxidation between 48.2 and 90.3% for the case of var. *cinammom* and between 38.1 and 84.4% for var. *album*. This showed, in both cases, the maximum potential at 360 h (15 d).

The α -tocopherol pattern showed a minimum percentage of inhibition of 55.4% (48 h) and peaked at 84.6% to 360 h (15 d), as well as the worked EOs. The through the ferric thiocyanate method was of +/- 0.8. Figure 2 displays the results of the pattern of inhibition and the EOs.

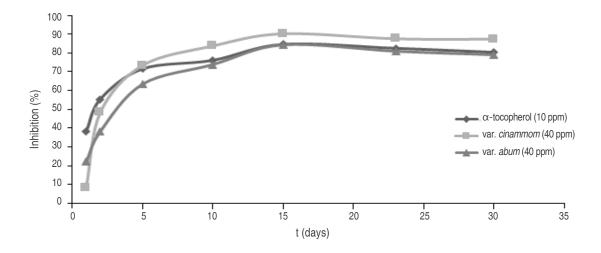


Figure 2. Evaluation of the antioxidant activity of a system of linoleic acid using by ferric thiocyanate method.

There was a similar behavior of α -tocopherol and EOs, for both cases, a maximum inhibition percentage at 360 h (15 d) was obtained. This similarity in the inhibitory power of the samples with α -tocopherol could be apparent if one takes into account the difference in concentrations between EOs (40 ppm) and standard (10 ppm). However, the varieties of *O. basilicum* showed the ability to prevent formation of peroxides as a result of deterioration of linoleic acid. Despite the degree of dilution worked, their potential was significant, even at 48 h (2 d), which reached a inhibition of about 40% (Murillo *et al.*, 2007).

The s of the results of moisture and extraction yield obtained for the plant material and of antioxidant capacity by DPPH and ferric thiocyanate methods are low (≤ 0.1), indicating that the methods used and the results obtained are repeatable and dependable (Sánchez and Santa, 2009). The results of the two methods are comparable for

the determination of the antioxidant activity, because in both cases the EOs var. *cinammom* revealed a higher potential activity compared to the var. *album*. Nevertheless, the antioxidant activity of var. *album* is representative (Rincón *et al.*, 2011).

Prior investigation suggest that an efficient antioxidant activity is mainly due to the presence of phenolic compounds (Rincón et al., 2011; Kuskoski et al., 2005; Juliani and Simon, 2002; Quiroga, 2013). This would support the high antioxidant activity of EOs worked; considering that in a previous investigation where the plant material was collected in the same location, it was found that var. cinammom worked contains eugenol (37.60%) and eucalyptol (24.06%) as major components, and var. album contains methyl (E)cinnamate (28.20%) and linalool (18.16%), in which the eugenol and methyl (E)-cinnamate belongs to the group of the phenylpropanes, and the eucalyptol and linalool to the group of oxygenated monoterpenes, chemical compounds that are directly related to the antioxidant capacity of worked O. basilicum varieties (Beltrán et al., 2010).

In a research conducted by Ramírez *et al.* (2013) the species *Ocimum basilicum* L., showed as major components phenylpropane (eugenol) and oxygenated monoterpenes (linalool and eucalyptol), demonstrating that the highest influence in the behavior of the antioxidant activity is the content of eugenol; therefore, when there is a higher content of eugenol, there is a better antioxidant activity (Juliani and Simon, 2002; Ramírez *et al.*, 2013; Wang *et al.*, 2010). This statement makes sense if it is consider that the var. *cinammon* has a 37.60% of eugenol, while the var. *album* worked this compound was not present, and evidently by both methods var. *cinammon* had a higher activity inhibition.

Mahecha (2010) indicated that in general, the antioxidant capacity is proportional to the presence of phenylpropanoid derivatives, phenols or proton donor substances in the composition of the OEs. This is why the two EOs from the plant material studied presented efficient inhibition.

On the other hand, it is well known that the high antioxidant activity of the phenolic monoterpenes

behavior can also be related to the antioxidant potential of EOs of *Ocimum basilicum* L. worked (Quiroga, 2013).

These chemicals compounds are part of the main source of natural antioxidants from fruits and vegetables, which are generally phenolic compounds in abundance. These compounds are closely associated with the color and flavor of plant origin, as well as its nutritional quality for its antioxidant properties. In several investigations, researchers have evaluated the antioxidant capacity of plant species, concluding that polyphenolic compounds are primarily responsible for the antioxidant activity "*in vitro*" (Rodas *et al.*, 2010).

These antioxidant compounds are able to inhibit oxidation and that is why they can be added to pharmaceutical or cosmetic products that are continuously exposed to deterioration by oxidative processes such as rancidity in oils and fats. They are also significant components foranti-aging preparations (Genaro, 2003; Rodas *et al.*, 2010).

The antioxidant capacity reflected by phenylpropane and oxygenated monoterpenes present in EOs of var. *cinammon* and var. *album*, can be considered as natural antioxidant compounds that could be used as additives in cosmetic products and thus reduces the use of synthetic antioxidants (Rincón *et al.*, 2011; Quiroga, 2013).

There are certain requirements to be met by antioxidants when being implemented in cosmetics. Some worth to be mentioned are that the concentrations used should not be irritating or allergenic, must not cause discoloration or odor in the preparation and should be sufficiently liposoluble to develop its effect. Besides the antioxidant must be stable and effective over a wide pH range, and be soluble in its oxidized form, and their products reaction should be colorless and odorless. Other essential and obvious requirements are that they should not be toxic; they must be stable and compatible with the product ingredients and composition of packaging (Genaro, 2003; Rodas *et al.*, 2010).

Aromatic and medicinal plants are an excellent alternative to be use in the cosmetics industry because

of their antioxidants properties. Therefore, they provide an opportunity to explore their potential in different growing zones, strengthening the productive chain and the cosmetic industry, a sector considered world class, given the possibility of natural raw materials in the manufacture of cosmetics (Ferraro and Martino, 2012).

Parameters such as geographical, climate, time of harvest, part of the plant used, origin, post-harvest treatment, etc. are important to take into account in order to optimize the concentration of primary and secondary metabolites of phytoingredients (Ferraro and Martino, 2012).

CONCLUSIONS

The EOs of *O. basilicum* L. var. *cinammom* and var. *album* showed significant antioxidant properties by the two spectrophotometric methods (DPPH and ferric thiocyanate). The DDPH method demonstrated the upward antioxidant potential according to the concentration, being a significant inhibitory power at 10 ppm of EOs.

The ferric thiocyanate method confirmed the antioxidant activity of EOs, worked at 40 ppm. It showed a maximum inhibition at 360 h (15 d), this inhibition potential is substantial compared to the reference standard (α -tocopherol) that we worked at lower concentration (10 ppm).

The effectiveness of the antioxidant activity to the essential oils is related with composition phenylpropane and oxygenated monoterpenes, which were previously studied. These chemicals compounds have been attributed the antioxidant behavior present in various varieties of *O. basilicum*. This anti-radical action may result as an excellent raw material for the production of natural antioxidants and be utilized in cosmetic formulations in order to prevent oxidative processes or as an active ingredient in anti-aging.

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REFERENCES

Álvarez N and Bague A. 2012. Fitocosméticos. Primera edición. AMV Ediciones, Madrid, España. 330 p.

Atehortua L. 1992. Banco de Germoplasma de Plantas Medicinales una prioridad nacional. In: Memorias I Symposium on Medicinal Plants. Universidad Javeriana. Santafé de Bogotá.

Beltrán M, Peláez E, Estrada J, Escobar J, Serna L and Ríos D. 2010. Estudio farmacognósico para el cuidado de la salud a partir de aceites esenciales obtenidos por destilación de arrastre de vapor. Investigaciones Andina 12(20): 8-18.

Carhuapoma M, Bonilla P, Suárez S, Vila R and López S. 2005. Estudio de la composición química y actividad antioxidante del aceite esencial de *Luma chequen* (Molina) A. Gray "arrayán". Ciencia e Investigación 8(2): 73-79.

Chaves M, Arango N. 1998. Informe nacional sobre el estado de la biodiversidad 1997-Colombia. Tomo III. Instituto de Investigación de Recursos Biológicos Alexander von Humboldt, Bogotá, Colombia. 133 p.

Cotelle N, Bernier J, Catteau J, Pommery J, Wallet J and Gaydou E. 1996. Antioxidant properties of hydroxy-flavones. Free Radical Biology and Medicine 20(1): 35-43. doi:10.1016/0891-5849(95)02014-4

Delgado Y, Báez J, Núñez M, García C, Amaya C and Pimentel D. 2010. Determinación de la actividad antioxidante del aceite esencial de orégano (*Poliomitha longiflora* Gray). pp. 1-7. In: Memorias XII National Congress of Food Technology. Universidad de Guanajuato. Guanajuato, México.

Díaz P. 2010. Efecto del tiempo de secado y de la variedad en las características físico-químicas de la albahaca (*Ocimun basilicum*) seca. Tesis en Ingeniería y Tecnología de Alimentos. Universidad Zamorano. Honduras. 18 p.

Fernández K, Viña A, Murillo E and Méndez J. 2007. Actividad antioxidante y antimicrobial de los volátiles de cuatro variedades de albahacas cultivadas en el departamento del Tolima. Scientia et Technica Año XIII (33): 401-403.

Ferraro G, Martino V, Bnadoni A and Nadinic J. 2012. Fitocosmética. Fitoingredientes y otros productos naturales. Primera edición. Editorial Eudeba, Buenos Aires, Argentina. 272 p.

Fonnegra R and Jiménez, S. 2007. Plantas medicinales aprobadas en Colombia. Second edition. Editorial Universidad de Antioquia, Medellín, Colombia. 368 p.

Forero N. 1987. La taxonomía, el herbario y la investigación etnobotánica. p. 251-255. In: Memorias I Colombian Symposium on Ethnobotany. Santa Marta, Colombia.

Gajardo S, Benites J, López J, Burgos N, Caro C and Rojas M. 2011. Astaxantina: antioxidante de origen natural con variadas aplicaciones en cosmética. Biofarbo 19(2): 6-12.

Genaro A. 2003. Remington: Farmacia. Volumen 1. Editorial Médica Panamericana, Argentina. 1388 p.

Granados C, Yáñez X and Santafé G. 2012. Evaluación de la actividad antioxidante del aceite esencial foliar de *Calycolpus moritzianus* y *Minthostachys mollis* de Norte de Santander. Bistua: Revista de la Facultad de Ciencias Básicas 10(1): 12-23.

Hirose M, Hagiwara A, Masui T, Inoue K and Ito N. 1986. Combined effects of butylated hydroxyanisole and other antioxidants in induction of forestomach lesions in rats. Cancer Letters 30(2): 169-174. doi: 10.1016/0304-3835(86)90085-6

Huang D, Chen H, Lin C and Lin Y. 2005. Antioxidant and antiproliferative activities of water spinach (*Ipomoea aquatica* Forsk) constituents. Botanical Bulletin of Academia Sinica 46(2): 99-106.

ICONTEC. 1996. NTC 3925. Análisis sensorial. Metodología. Guía general. Instituto Colombiano de Normas Técnicas y Certificación, Santa Fe de Bogotá. 25p.

Juliani H and Simon J. 2002. Antioxidant Activity of Basil. pp. 575–579. In: Janick J, Whipey A. (eds.). Trends in new crops and new uses. First edition. ASHS Press, Alexandria. 599 p.

Kuskoski E, Asuero A, Troncoso A, Mancini-Filho J and Fett R. 2005. Aplicación de diversos métodos químicos para determinar actividad antioxidante en pulpa de frutos. Food Science and Technology (Campinas) 25(4): 726-732. doi: 10.1590/S0101-20612005000400016

Lima S. 2005. Análisis de los rendimientos obtenidos de dos especies de eucalipto trabajados en seco a nivel laboratorio y a nivel planta piloto en la extracción de su aceite esencial. Thesis in Chemical Engineering. Faculty of Engineering. Universidad de San Carlos de Guatemala. Guatemala. 76 p.

Madrigal E, García F, Morales J, Vászquez P, Muñoz S, Zuñiga C, Sumaya M, Madrigal E and Hernández A. (2013). Antioxidant and anticlastogenic capacity of prickly pear juice. Nutrients. 5(10): 4145–4158. doi: 10.3390/nu5104145

Maestro R and Borja R. 1993. Actividad antioxidante de los compuestos fenólicos. Grasas y Aceites 44(2): 101-106. doi:10.3989/gya.1993.v44.i2.1105

Mahecha C. 2010. Actividad antioxidante y antibacteriana de aceites esenciales extraidos de hojas y frutos de *Siparuna sessiliflora*. Tesis de Maestría en Ciencias Biológicas. Departamento de Química. Facultad de Ciencias. Pontificia Universidad Javeriana. Santa Fe de Bogotá. 115 p.

Martínez A. 2003. Aceites esenciales. Facultad de Química Farmacéutica. Universidad de Antioquia, Medellín. 34 p.

Martínez M. 2010. Extracción y caracterización de aceite de nuez (*Juglans regia* L.): influencia del cultivar y de factores tecnológicos sobre su composición y estabilidad oxidativa. Tesis Doctoral en Ciencias de la Ingeniería. Facultad de Ciencias Exactas, Físicas y Naturales. Universidad Nacional de Córdoba. Cordoba, Argentina. 141 p.

Muñoz M and Gutiérrez D. 2009. Determinación de actividad antioxidante de diversas partes del árbol *Nicotiana glauca*. Facultad de Química. Universidad Autónoma de Queretaro. Citado por : Alejandro M, Jaramillo X, Ojeda S, Malagón O and Ramírez J. 2013. Actividad antioxidante y antihiperglucemiante de la especie medicinal *Oreocallis grandiflora* (Lam.) R. Br., al sur del Ecuador. Boletín Latinoamericano y del Caribe de Plantas Medicinales y Aromáticas 12 (1): 59 – 68.

Murillo E, Fernández K, Sierra D, Viña A. 2004. Caracterización fisicoquímica del aceite esencial de albahaca. II. Revista Colombiana de Química 33(2):139-48. ISSN: 2357-3791

Murillo E, Fernández K, Viña A and Méndez J. 2007. Actividad antioxidante *in vitro* y antimicrobial de extractos metanólicos de cuatro albahacas cultivadas en Ibagué. Journal Tumbaga (2): 72-84. Osawa T and Namiki M. 1981. A novel type of antioxidant isolated from leaf wax of eucalyptus leaves. Agricultural and Biological Chemistry 45(3): 735-739.

Quiroga P. 2013. Evaluación de aceites esenciales y monoterpenos como agentes conservantes de las propiedades químicas y sensoriales de los alimentos. Tesis Doctoral en Ciencias Agropecuarias. Facultad de Ciencias Agropecuarias. Universidad Nacional de Córdoba. Córdoba, Argentina. 184 p.

Ramírez R, Angulo A, Olivero J and Santafé G. 2013. Relación entre la composición química y la actividad antioxidante del aceite esencial de *Ocimum basilicum* L. cultivado bajo diferentes tratamientos de fertilizante. Revista Cubana de Plantas Medicinales 18(1): 47-56.

Reyes J, Patiño J and Stashenko E. 2007. Caracterización de los metabolitos secundarios de dos especies de *Ocimum* (Labiatae), en función del método de extracción. Scientia et Technica Año XIII (33): 121-123.

Rincón A, Pérez M, Bou L, Romero A, Bucarito L and Padilla F. 2011. Métodos para la determinación de la actividad antioxidante de vegetales. Revista Facultad de Farmacia 74(1): 24-28.

Rojas D, Narvaéz E and Restrepo L. 2008. Evaluación del contenido de vitamina C, fenoles totales y actividad antioxidante en pulpa de guayaba (*Psidium guajava* L.) de las variedades pera, regional roja y regional blanca. p. 49-60. In: Memorias Red-Alfa Lagrotech. Comunidad Europea. Cartagena.

Rodas E, López K and Tul Y. 2010. Evaluación de la actividad antioxidante de extractos frutales como alternativa a los antioxidantes sintéticos en preparaciones cosméticas tipo emulsión. Seminario de Investigación en Química Farmacéutica. Facultad de Ciencias Químicas y Farmacia. Universidad de San Carlos de Guatemala. Guatemala. 86 p.

Romero C, Belisario Y and Blasco M. 2004. Extracción del aceite esencial de albahaca (*Ocimum basilicum* L.) con CO_2 supercrítico. Ciencia Scientific Journal from the Experimental Faculty of Sciences, Universidad del Zulia 12(4): 309-314.

Sánchez V, Santa J. 2009. Estudio de las antraquinonas presentes en extractos de mucílagos y hojas de *Aloe vera* de plantas cultivadas en la región cafetera. Tesis en Tecnología Química. Facultad de Tecnología. Universidad Tecnológica de Pereira. Pereira, Colombia. 60 p.

Solanilla J, Lombo O, Murillo E and Méndez J. 2011. Valoración del potencial antioxidante de *Mollinedia racemosa* (romadizo). Revista Cubana de Plantas Medicinales 16(2): 151-163.

Wang H, Yih K and Huang K. 2010. Comparative study of the antioxidant activity of forty-five commonly used essential oils and their potential active components. Journal of Food and Drug Analysis 18(1): 24-33.

Zumbado H. 2004. Análisis químico de los alimentos: métodos clásicos. Primera edtición. Editorial Universitaria, La Habana. 433 p.