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Insecticidal effects of *Ocimum sanctum* var. *cubensis* essential oil on the diseases vector *Chrysomya putoria*

[Efecto insecticida del aceite esencial de *Ocimum sanctum* var. *cubensis* sobre el vector de enfermedades *Chrysomya putoria*]

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Abstract

Context: The blowfly *Chrysomya putoria* is widely distributed throughout the Neotropical region and, besides transmitting pathogens; they could cause secondary myiasis. Botanical insecticides provide an alternative to synthetic pesticides because the excessive use of synthetic insecticides resulted in a progressive resistance of the pests to these chemicals, diminishing their effectiveness and generating consequences with negative environmental impact. The essential oil extracted from *Ocimum sanctum* (basil) has showed insecticidal activity against some insects but has no reported studies on the activity of this plant against flies.

Aims: To evaluate the insecticidal effects of *Ocimum sanctum* var. *cubensis* Gomes essential oil on the post embryonic development of *Chrysomya putoria*.

Methods: The colonies of *Chrysomya putoria* were established and maintained at the Laboratório de Entomologia Médica e Forense (FIOCRUZ), Rio de Janeiro, Brazil. The basil essential oil was tested in six concentrations (4.13, 8.25, 20.63, 41.25, 61.87 and 80,25 mg/mL). Mortality and changes in life cycle were recorded daily.

Results: β-caryophyllene, β-selinene and eugenol, were the main constituents of the basil essential oil. The experiments demonstrated that in all concentrations tested, this essential oil shortening the duration of all post embryonic stages having a direct impact in the viability of this fly estimating the LC_{50} in 7.47 mg/mL of concentration. In addition, the essential oil caused morphological alterations in abdomen, wings and ptilinum at lower concentrations.

Conclusions: This essential oil emerge as a good option for the control of the disease vector blowfly *Chrysomya putoria*.

Keywords: blowfly; holy basil; larvicidal effect; myiasis; sanitary importance.

Resumen

Contexto: La mosca *Chrysomya putoria* está ampliamente distribuida en toda la región neotropical y, además de transmitir patógenos, puede causar miiasis secundaria. Los insecticidas botánicos proporcionan una alternativa a los plaguicidas sintéticos porque el uso excesivo de los sintéticos resultó en una resistencia progresiva de las plagas a éstos, disminuyendo su efectividad y generando un impacto ambiental negativo. El aceite esencial de *Ocimum sanctum* (albahaca) ha mostrado actividad insecticida contra algunos insectos, pero no se han reportado estudios sobre la actividad de este contra moscas.

Objetivos: Evaluar el efecto insecticida del aceite esencial de *Ocimum sanctum* var. *cubensis* Gomes en el desarrollo post embrionario de *Chrysomya putoria*.

Métodos: Las colonias de *Chrysomya putoria* fueron establecidas y mantenidas en el Laboratorio de Entomología Médica y Forense (FIOCRUZ), Río de Janeiro, Brasil. El aceite esencial de albahaca se ensayó en seis concentraciones (4,13; 8,25; 20,63; 41,25; 61,87 and 80,25 mg/mL). La mortalidad y los cambios en el ciclo de vida se registraron diariamente.

Resultados: β-cariofileno, β-selineno y eugenol fueron los principales constituyentes del aceite esencial de *O. sanctum*. El aceite esencial acorta la duración de todas las etapas post-embrionarias que tuvieron un impacto directo en la viabilidad de esta mosca, estimando la CL_{50} en 7.47 mg/mL de concentración. Además, el aceite esencial causó alteraciones morfológicas en el abdomen, las alas y el ptilinum de los insectos a las concentraciones más bajas.

Conclusiones: Este aceite esencial emerge como una buena opción contra la mosca *Chrysomya putoria* para el control del vector de enfermedades.

Palabras Clave: albahaca santa; efecto larvicida; importancia sanitaria; miiasis; mosca.

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INTRODUCTION

Since ancient times, flies have been in contact with humans. The species of the genus *Chrysomya* (Robineau-Desvoidy, 1830) (Diptera: Calliphoridae) are commonly known as blowflies and, like other blowflies, *Chrysomya putoria* (Wiedemann, 1818) is now distributed through the New World (Guimarães et al., 1978). Blowflies are of medical and veterinary importance worldwide due to their feeding habits, such as on animal and human excrement, urban garbage, decomposing meat and fresh food (Tomberlin et al., 2017). These flies are synanthropic and could act as important physical carriers of various pathogens to man and domestic animals such as bacteria (*Escherichia coli*, *Proteus mirabilis*, *Citrobacter* sp., *Klebesiella* sp., *Morganella* sp., *Pseudomona* sp., *Enterobacter* sp., *Salmonella* sp.), viruses (West Nile Virus), protozoa (*Entamoeba coli, Entamoeba histolytica*, *Giardia lamblia* and *Cryptosporidium* sp.) and helminths (*Ascaris lumbricoides*, *Trichuris trichiura*, *Strongyloides stercoralis*, *Taenia* spp.) (Getachew et al., 2007; Johnson et al., 2010; Nayduch and Burrus, 2017). *Chrysomya putoria* are commonly found in latrines and larvae can cause secondary myiasis in animals and humans (Barbosa and Vasconcelos, 2015).

The current use of chemical insecticides could cause untold toxicological damage to humans and animals, in the short and long terms. Moreover, insecticide resistance has become an increasing problem in agriculture, in the economy and in public health. Botanical insecticides provide an alternative to synthetic pesticides because the excessive use of synthetic insecticides resulted in a progressive resistance of the pests to these chemicals, diminishing their effectiveness and generating consequences with negative environmental impact (Valente et al., 2007).

In the last decades, thousands of plants have been studied to detect activity as repellents and insecticides. Secondary metabolites often have a potential to control insects, because they are naturally feeding deterrents, toxic agents or developmental disruptors. It has been shown that numerous plant species growing in different geographical areas around the world cause lethal and sub-lethal effects on various insects (Cabral et al., 2007; Mendonça et al., 2011).

Ocimum sanctum L. (Lamiaceae) also known as *Ocimum tenuiflorum* L., holy basil, tulasi or tulsi is an aromatic plant native to the Indian subcontinent and cultivated throughout the tropical Southeast Asia, cultivated and naturalized also in other tropical areas (Al Rashid et al., 2013). The plant is less known in western countries where a lack of information on cultural practices exists (Sims et al., 2014). In Cuba, it is cultivated in yards and gardens like aromatic herb. Its seeds easily propagate by air and then could be found spontaneously in the neighborhoods of the towns and the peasant dwellings (Roig, 1998).

The extracts of this species have potential in treatments for coughs, dysentery, diarrhea, worms and kidney malfunction, bronchitis, liver diseases, genitourinary disorders, catarrhal fever, otalgia, lumbago, hiccough, ophthalmia, painful eye disease, gastric disorders, diabetes, skin diseases, various forms of poisoning and psychosomatic stress disorders, arthritis, chronic fever and insect sting (Al Rashid et al., 2013; Gradinariu et al., 2015; Jamal et al., 2015).

Previous reports sign the essential oil of *O. sanctum* as insecticide, but those studies were developed only against mosquitoes' species (*Anopheles stephensi* Liston, *Aedes aegypti* Linnaeus and *Culex quinquefasciatus*) under laboratory conditions (Bhatnagar et al., 1993), but in the literature reviewed there is not reports regarding their actions over flies.

The aim of this study was to evaluate the insecticidal effect of the essential oil of *Ocimum sanctum* L. var. *cubensis* Gomes on the post-embryonic development of the blowfly *Chrysomya putoria*.

MATERIAL AND METHODS

Plant material

The leaves of *Ocimum sanctum* were collected in September 2016, in the municipality of San Luis (20°11'13.88" N; 75°50'51.31" O), province of Santiago de Cuba, Cuba. The plants were in a vegetative state. To collect the plant a region was selected where a population of more than 20 plants of the species existed. The collected plants were randomly selected and taxonomically identified by Mr. Jainer Costa Acosta, specialist at the Eastern Center for Ecosystems and Biodiversity (BIOECO) in the prov-

ince of Santiago de Cuba. A voucher specimen was checked and confirmed with a previous one archived at the BSC Herbarium of the same center under No. 3247.

Essential oil extraction

The extraction of the essential oil from *Ocimum sanctum* L. fresh leaves was carried out until exhaustion by hydrodistillation, using a conventional Clevenger apparatus. The essential oil chemical composition was determined in a Gas Chromatography Mega 2 series coupled to a mass spectrometer (GC/MS) Hewlett Packard model 5890 (USA), using the same conditions declared in a previous work conducted by the research group. A DB-5 MS capillary column (Agilent Technologies, USA) and helium as carrier gas were used. The program temperature condition was 60°C (2 min), with an increment of 3°C/min until 110°C, of 15°C/min until 150°C and finally with an increment of 17°C/min until 290°C (Chil Núñez et al., 2017). Electron impact ionization at -70 eV was used to characterize the compounds that were identified comparing their mass spectral data with the National Institute of Standards and Technology mass spectrometry library and according with their Kovats retention indexes.

Colonies of Diptera

The flies were obtained from the Collection of flies of the laboratory "Laboratório de Entomologia Médica e Forense, Instituto Oswaldo Cruz/Fundação Oswaldo Cruz", Rio de Janeiro, RJ, Brazil. The adult flies were kept in wooden cages with nylon screens on the sides and a front opening containing a black fabric sleeve for insect handling. The volume of the cage was approximately 2.7 m^3 (30 x 30 x 30 cm).

Bioassays

For the topical application, four groups of 50 newly-hatched larvae were placed in Petri dishes and get in contact with 1 µL/larva applied on the larvae bodies using an automatic pipette. Six level of concentration of the essential oil were tested and defined as experimental groups A, B, C, D, E and F: (A = 4.13 mg/mL, B = 8.25 mg/mL, C = 20.63 mg/mL, D = 41.25 mg/mL, E = 61.87 mg/mL and F = 80.25 mg/mL) were prepared using dimethylsulfoxide (DMSO, ACS reagent ≥ 99.9%; 472301 Sigma-Aldrich) as solvent. Three control groups were considered: solvent control (just with pure DMSO), classic or pure control where no substances were added and a natural insecticide essential oil as positive control. With this purpose, *Cymbopogon citratus* essential oil at 83,3 mg/mL was used according to previous experiences at laboratory with this substance on this biological model (Pinto et al., 2015). Bioassays were performed in quadruplicate.

After application of the different concentrations of the essential oil, the larvae were transferred to a vessel containing 50 g of bovine putrefied meat. These 50 mL vials were then placed in 500 mL vials (Copaza, Brazil) containing vermiculite (Vermiculite Expanded Medium, expansion volume 0.1 m^3, Brazil) as substrate for pupation and covered with a nylon web clamped with a rubber band. The experiments were maintained in acclimatized chambers setting the temperature at 27 ± 1°C, 70 ± 10% of relative humidity and with a 12:12 light cycle (light/darkness). Mortality and changes in life cycle were recorded daily. After reaching maturity, the larvae spontaneously abandoned the diet and were collected. These larvae were individually weighed and transferred to glass tubes containing vermiculite and sealed with cotton plugs. After emergence, adults were separated by gender selected by the distance between the eyes (Dübendorfer et al., 2002). The bioassays were performed in quadruplicate with the second laboratory generation. Corrected mortality and duration of each developmental period (larval, pupal and newly-hatched larvae to adult) were analyzed. With this intention, a corrected mortality parameter was calculated using the Abbot equation (equation 1). The weight of mature larvae and the sex ratio (equation 2) were also considered.

$$\text{Corrected Mortality} = \frac{(\text{treated group mortality (\%)} - \text{Control group mortality}) \times 100}{100 - \text{Control group mortality (\%)}} \quad (1)$$

$$\text{Sex ratio} = \frac{\text{Number of females flies emerged}}{(\text{Number of females flies emerged} + \text{Number of male flies emerged})} \quad (2)$$

Morphological alterations

All insects were observed under an optical stereoscope (Zeiss, model Stemi SV 6, Germany) to investigate possible morphological alterations in

head, wings and abdomen. Pictures were taken using a digital camera (Canon, model PowerShot A630, Japan) attached to the stereoscope.

Statistical analysis

The duration of development periods of this fly species and larval weight were analyzed by ANOVA ($p \leq 0.05$), mean values were compared with the Newman-Keuls test at a significance level of 0.05, while sex ratio and mortality were analyzed with the chi-square test. Mortality was corrected using Abbot's formula (Abbott, 1995). Probit analysis of concentration-mortality data was conducted to estimate the LC_{50} value using the SPSS for Windows 18.0/2009 statistical package. For this intention, data normalization was done using the square root of the concentrations tested.

RESULTS

The chemical composition showed β-caryophyllene, β-selinene and eugenol as the main compounds (Table 1). These results are similar to those obtained in previous studies matching two of the three main compounds (β-caryophyllene and eugenol) according to previous experiences of the research group (Chil Núñez et al., 2017). The compound bicyclogermacrene (that appears as majority in the previous study with 20.38%) in this case is present in 7.81% as fourth place after the three-main declared. In total, seven compounds are common between both samples representing more than the fifty percent (Table 1).

Table 1. Chemical composition of essential oil of *Ocimun sanctum* var. *cubensis* Gomes.

Compound	Area (%)	Retention index (experimental)	Retention index (reported)*
Elemene <delta->	2.84	1326	1327
Eugenol	12.34	1365	1367
Eugenol <methyl->	3.71	1387	1383
Caryophyllene	19.28	1424	1423
Elemene <gamma->	2.37	1428	1430
Farnesene <(E)-, beta->	1.77	1451	1454
Humulene <alpha->	3.6	1455	1454
Alloaromadendrene	1.64	1458	1457
Gurjunene <gamma->	7.17	1475	1475
Selinene <beta->	16.14	1493	1496
Bicyclogermacrene	7.81	1500	1500
Bisabolene <beta->	5.8	1507	1509
Spathulenol	3.32	1578	1577
Caryophyllene oxide	1.06	1581	1582
Eudesmol <epi-gamma->	1.16	1624	1622
Unidentified compound	1.31	1634	-
Hinesol	1.9	1640	1640
Eudesm-7(11)-en-4-ol	2.78	1644	1646
Cadin-4-en-10-ol	2.81	1658	1656
Phytol	1.19	2106	2105

*Retention index reported by Gas Chromatographic Retention Data- the NIST WebBook.

The six concentrations evaluated affected the life cycle of flies. It is noted that all concentrations of the essential oil *O. sanctum* showed a significant decrease in the duration of the larval period of *C. putoria* when compared to control group and control with DMSO, but not significant regarding to the positive control *C. citratus* essential oil (Table 2). The duration of the pupal period was also significantly lower for insects treated with all concentrations of essential oil when compared to the control group and control with DMSO, except for the group treated with 20.63 mg/mL. On the other hand, these times reduction were different statistically than those obtained for the positive control same tendency of time reduction with statistical differences is described when analyzing the newly hatched larvae to adult period (Table 2).

Larval weight was also directly affected by basil essential oil. According to Fig. 1, larvae from pure control group (36.5 ± 10.17 mg) and DMSO control (34.7 ± 5.24 mg) were lighter than the larvae treated with *O. sanctum* essential oil in all concentrations (all above 50 mg), with highly significant differences ($p < 0.001$) as well happen with the positive control with which not statistical behavior is found. Therefore, the use of holly basil essential oil besides reducing the larval period (Table 2) also caused the larvae to gain more weight in a shorter time.

Mortality effects of holly basil oil were observed in all concentrations tested (Fig. 2). In the larvae stage, the most effective concentration of the essential oil was 4.13 mg/mL and the least effective was a concentration of 61.87 mg/mL. The mortality in the pupal period was higher than in the larval and newly hatched larvae to adult periods for five of six concentrations tested as well as happen with positive control. The only exception was for the group treated with 80.25 mg/mL essential oil (Fig. 2).

Regarding to the sexual ratio parameter, no statistical difference was found, according to the chi-square test, similar as happen with all control groups including the positive one. By this way, the use of this natural extract has not a measured influence on this biological variable.

Besides all the modifications of the postembryonic cycle, *O. sanctum* also caused morphological alterations in adults whose larvae where treated with 4.13 mg/mL and 8.25 mg/mL concentrations (Fig. 3A-B).

Table 2. Effects on the postembryonic development period of *Chrysomya putoria* when topically treated with *Ocimum sanctum* var. *cubensis* Gomes essential oil.

Treatment	Larval period (days)	Pupal period (days)	Newly hatched larvae to adult (days)
Control	3.16 ± 0.37^{c}	5.16 ± 0.45^{a}	8.31 ± 0.58^{b}
Control with DMSO	3.78 ± 0.54^{a}	5.12 ± 0.41^{a}	8.86 ± 0.52^{a}
Cymbopogon citratus (pure oil)	2.99 ± 0.28^{d}	4.51 ± 0.33^{c}	7.50 ± 0.70^{d}
Ocimum sanctum (mg/mL)			
4.13	3.03 ± 0.17^{d}	4.70 ± 0.45^{b}	7.72 ± 0.51^{d}
8.25	3.00 ± 0.01^{d}	4.57 ± 0.58^{b}	7.55 ± 0.55^{d}
20.63	3.00 ± 0.03^{d}	4.94 ± 0.22^{a}	7.94 ± 0.22^{c}
41.25	3.00 ± 0.04^{d}	4.73 ± 0.64^{b}	7.74 ± 0.70^{d}
61.87	3.00 ± 0.01^{d}	4.71 ± 0.73^{b}	7.71 ± 0.73^{d}
80.25	3.39 ± 0.49^{b}	4.68 ± 0.53^{b}	8.16 ± 0.82^{c}

Data represent mean ± SD of n = 200 larvae per each group. Values with different upper letters were significantly different (Newman-Keuls test; $p < 0.05$). Dimethylsulfoxide (DMSO) was used as vehicle and *Cymbopogon citratusessential* oil as a reference compound.

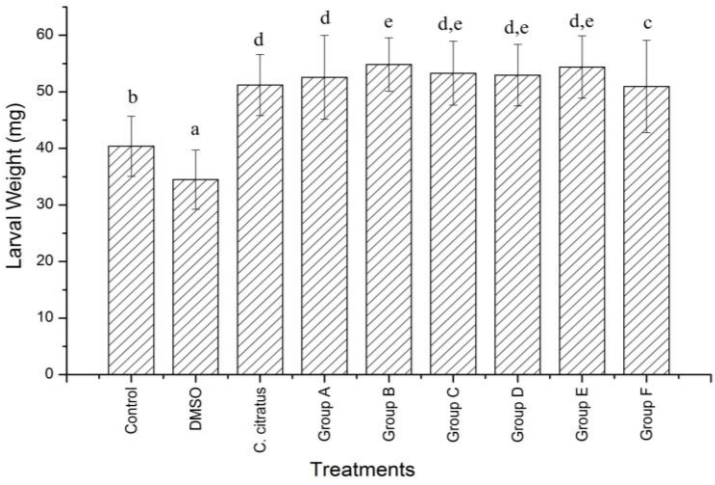

Figure 1. Effects on *Chrysomya putoria* larval weight when topically treated with *Ocimum sanctum* var. *cubensis* Gomes essential oil. Data represent mean ± SD of n = 200 larvae per each group. Values with different upper letters were significantly different (Newman-Keuls test; $p < 0.05$).

Groups A-F: *Ocimum sanctum* var. *cubensis* Gomes essential oil at different concentrations: A = 4.13 mg/mL, B = 8.25 mg/mL, C = 20.63 mg/mL, D = 41.25 mg/mL, E = 61.87 mg/mL and F = 80.25 mg/mL dissolved in dimethylsulfoxide (DMSO). *Cymbopogon citratus* essential oil (*C. citratus*) was used as a reference compound.

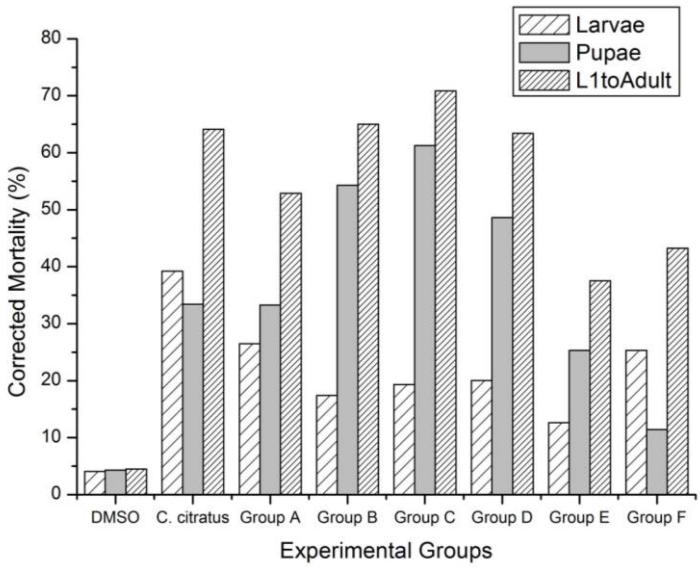

Figure 2. Mortality (%), corrected using Abbot's formula (1925), of *Chrysomya putoria* (Diptera: Calliphoridae) topically treated with different concentrations of essential oil extracted from *Ocimum sanctum* var. *cubensis* Gomes.

Groups A-F: *Ocimum sanctum* var. *cubensis* Gomes essential oil at different concentrations: A = 4.13 mg/mL, B = 8.25 mg/mL, C = 20.63 mg/mL, D = 41.25 mg/mL, E = 61.87 mg/mL and F = 80.25 mg/mL dissolved in dimethylsulfoxide (DMSO). *Cymbopogon citratus* essential oil (*C. citratus*) was used as a reference compound. L1 to adult → Newly hatched larvae to adults.

DISCUSSION

As can be observed in Table 1, similarities and differences can be appreciated in *O. sanctun* essential oil. The fact that the main compounds remains almost unaltered in both studies could mean that they can be considered as important chemical patterns of the *Ocimum sanctum* var. *cubensis* essential oil that grow up in Cuba, giving them an important chemotaxonomic role. This is in agreement with other studies within the American continent presenting eugenol and and β-caryophyllene as the major components (Machado et al., 1999; Sims et al., 2014). On the other hand, the different composition regarding the minor compounds are in agreement with the suggestion that aromatic character of each type of basil depends on the genetics of the vegetable, its state of vegetative development, agroclimatological factors and chemical compounds in the synthesis of essential oils as is well described in the literature (Sims et al., 2014; Jamal et al., 2015).

Eugenol and β-caryophyllene are frequently found in high concentrations in essential oils with insecticidal properties (Jantan and Zaki, 1998). Eugenol is repeatedly described as a good insecticide compound in many kinds of insects and also in flies, such as *Musca domestica* L. and *Drosophila melanogaster* (Meigen, 1830), as well as sesquiterpenes (Koul et al., 2008).

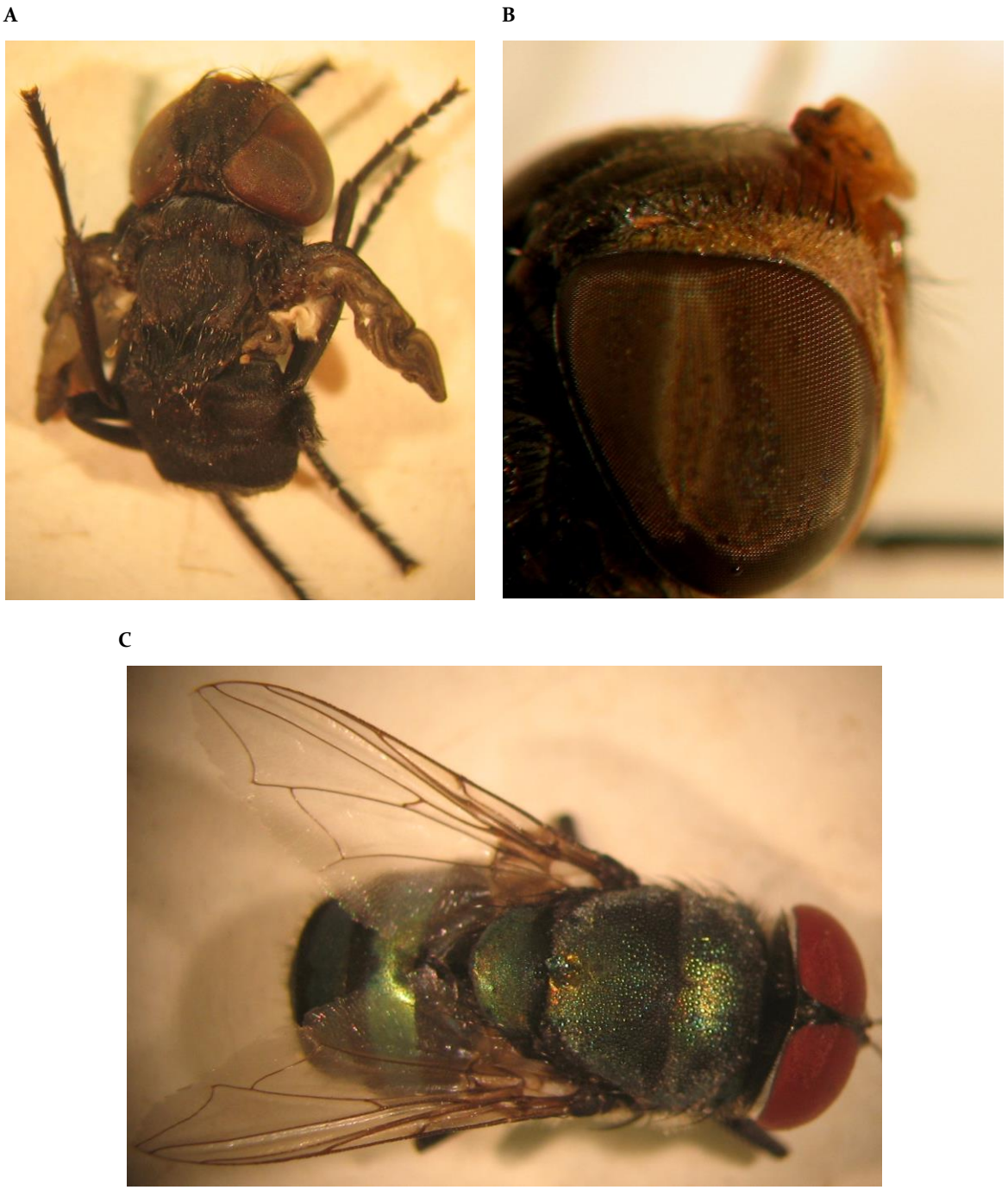

Figure 3. Morphological alterations of *Chrysomya putoria* (Diptera: Calliphoridae) adults topically treated with 5 and 10% concentrations of essential oil extracted from *Ocimum sanctum* var. *cubensis* Gomes.

A: Abdomen contracted, and wings twisted (x16). **B:** Ptilinum not retracted (x40). **C:** Insect from control group (x16).

It is internationally accepted that the larval period is the most important in the life cycle of the blowfly. This is the period where the larvae get to eat as much food as possible in the shortest time Furthermore larvae remain on the diet for a shorter time if the quantity and quality of nutrients is enough for pupation and if they reach the minimum weight needed for pupation. Nevertheless, the

standard conditions in which all experimental and control groups are conducted allowed us to infer that the effects observed were due to the effect of the holly basil essential oil, that is the same when comparing with the positive control. A previous study tested the larvicidal effects of aqueous crude extract from *Pouteria sapota* (Jacq.) (Sapotaceae) on the same blowfly species and found the same behavior: a time decreasing larval period (Carriço et al., 2014). This finding is important, once the larval period is one of the most vulnerable in flies' development cycle. During this period, the insects are more susceptible to predation.

The pupal period is when the most important hormonal changes are taking place in holometabolous insects, such as the blowflies. Cabral et al. (2007) suggested that compounds extracted from plants and tested to control insects could modify specific physiological processes such as the endocrine control of insect growth, the neuroendocrine system or the production of some hormones. Holy basil essential oil could be, somehow, affected those physiological processes. Kostyukovsky et al. (2002) stated that treatments with essential oils or their purified constituents resulted in visible symptoms suggesting a neurotoxic mechanism of action similar to that produced by carbamates and organophosphates. In another study, it was observed that the terpenes of essential oil have been shown to be competitive inhibitors of the acetylcholinesterase isolated from the electric eels, as well as the cholinesterase of the domestic fly and cockroach heads (Ryan and Byrne, 1998).

The newly hatched larvae adult period or the complete developmental period is the most efficient parameter to evaluate the substance efficacy as an insecticide, because it prevents distortions between the larval and pupal periods (d'Almeida et al., 2001). According to Table 2, higher concentrations affected this variable less than the lower ones, which is not usual. A possible explanation for this fact is to consider the nature of the substance applied. In this particular case, the application of a pure essential oil makes it more susceptible to evaporation under lab conditions than when dissolutions in DMSO are used. Thus, the larvae treated with lower concentrations could be in contact with the tested substance for longer periods of time, while the higher concentrations could lose the active substances through evaporation. Another possibility can be related with the DMSO use. Some authors have suggested the ability of DMSO to increase the membrane permeability. Notman et al. (2006) referred that DMSO was able to induce pores in the membrane, while Gurtovenko and Anwar (2007) referred that this substance induced thinning and expansion of a phospholipid bilayer increasing their fluidity. This hydrophobic core can explain why DMSO promotes the permeation of solutes, particularly in hydrophobic entities. Nevertheless, and in spite of any theoretical explanation, the essential oil tested at all concentrations proved to be effective against the insect development.

The reduction of the larval period was also observed by Carriço et al. (2014) using crude aqueous extract of *P. sapota* on the same blowfly species. Further, Carvalho et al. (2012) when testing the effects of cocaine on the developmental rate of *Chrysomya albiceps* (Wiedemann, 1819) and *C. putoria*, found the same effects. They considered that this performance was due to some effect on the endocrine system of the flies; this could also be the cause of this particular behavior in the present study.

The corrected mortality was superior that 50% in the lower concentrations reaching a top of 70% for the experimental group C. This C group as well as group B showed values that exceed the obtained for the positive control, demonstrating a good activity. Once again, the groups with higher concentrations of essential oil were less effective than groups with lower concentrations. The possibility of evaporation and/or the DMSO effect again helps to explain this particular behavior. Considering the non-linear tendency of the most concentrated experimental groups (E, F) those mortality values were skipped while computing the LC_{50}, rendering a 7.47 mg/mL value.

In the group A, from a total of 92 flies that emerges as adults, the 13.04% (12 flies) results in some kind of morphological alteration; while for the group B, seven flies from 67 (10.44%) appear also with some handicap. This behavior appears just in the groups treated at minor concentrations, indicating how toxic could be the essential oil to the postembryonic cycle of this fly since when; if is not

able to kill, produces some alteration affecting their normal development. The most common morphological alterations found were a contraction of the abdomen, ptilinum not retracted, and a twisted hemolymph affecting the wingspan (Fig. 3A-B).

All insects from control group were normal (Fig. 3C). The morphological alterations affected directly the ability to fly, and consequently to find food and to reproduce. So, these results demonstrated that basil essential oils may represent an effective alternative method to control blowfly.

CONCLUSIONS

The results obtained in this work indicate that the essential oil of *Ocimum sanctum* var. *cubensis* Gomes is a good option for the control of the blowfly *Chrysomya putoria,* in some cases with better performance than the positive control *Cymbopogon citrates* essential oil. The principal strength of the present work consists not only in the first report of the effect on the post-embryonic development of this blowfly, but also that it is possible to have a good activity with low concentrations of this essential oil on this species of fly with medical and sanitary importance, reaching 70% mortality with a 20.63 mg/mL concentration and a LC_{50} estimated as 7.47 mg/mL.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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Concepts or ideas	X	X	X	X	X	X
Design	X	X	X			X
Definition of intellectual content	X	X	X	X	X	X
Literature search	X	X	X			X
Experimental studies	X	X		X		
Data acquisition	X	X		X		
Data analysis	X	X	X	X	X	X
Statistical analysis	X	X	X		X	
Manuscript preparation	X	X	X	X	X	X
Manuscript editing	X	X	X	X	X	X
Manuscript review	X	X	X	X	X	X

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