Sociobiology 67(2): 292-300 (June, 2020) DOI: 10.13102/sociobiology.v67i2.4992

Sociobiology

An international journal on social insects

RESEARCH ARTICLE - TERMITES

Insecticidal activity of essential oils of species from the genus *Lippia* against *Nasutitermes corniger* (Motschulsky) (Isoptera: Termitidae)

DR Santos[1], LM Oliveira[2], AM Lucchese[3], AF Espeleta[3], JD Cruz[2], MS Lordêlo[3]

1 - Graduate Program in Plant Genetic Resources, Feira de Santana State University (UEFS), Bahia, Brazil

2 - Departament of Biological Sciences, Feira de Santana State University (UEFS), Bahia, Brazil

3 - Departament of Exact Sciences, Feira de Santana State University (UEFS), Bahia, Brazil

Article History

Edited by

Evandro Nascimento Silva, UEFS, Brazil

Received	30 January 2020
Initial acceptance	09 February 2020
Final acceptance	10 February 2020
Publication date	30 June 2020

Keywords

Verbenaceae, *Lippia lasiocalycina, Lippia insignis, Lippia thymoides,* Termites, Biopesticide.

Corresponding author

Daiane Rodrigues dos Santos

Graduate Program in Plant Genetic Resources

Feira de Santana State University (UEFS)

Av. Transnordestina s/nº, Novo Horizonte

Feira de Santana, Bahia, Brasil.

E-Mail: daibio@hotmail.com.br

Abstract

Lippia is one of the main genera in the family Verbenaceae, with 200 species described. Despite its richness in bioactive molecules, with several scientifically proven applications, there is little information on the insecticidal potential of its species. This study aimed to assess the insecticidal potential of essential oils from the species *Lippia thymoides* (Martius & Schauer); *Lippia lasiocalycina* (Schauer) and *Lippia insignis* (Moldenk) against *Nasutitermes corniger* (Motschulsky) (Isoptera-Termitidae). Insecticidal activity was evaluated by exposure to a contaminated surface, whereby plastic pots were lined with filter paper and imbibed in 1.5 ml of solution containing essential oils (10 µl/ml), with 10 *N. corniger* specimens per pot. The mortality count was performed at 24 and 48 h. The LC_{50} was determined by diluting the essential oils to concentrations of 0, 0.625, 1.25, 2.5, 5.0 and 10 µl/ml, which were chemically analyzed by GC-FID and GC-MS. The data indicated high toxicity for the essential oils for the *Lippia* species tested. The lowest LC_{50} (0.46 µl/ml) was recorded for *L. lasiocalycina*. The most common constituents were β-myrcene and (E)-ocimenone in essential oil of *L. lasiocalycina*, β-myrcene and limonene for *L. insignis,* and (E)-caryophyllene and caryophyllene oxide for *L. thymoides*. The results demonstrate the viability of developing biopesticides for *N. corniger* control.

Introduction

Isoptera (Termitidae) is an infraorder that includes social insects, with around 2,900 species described and widespread occurrence, predominantly in Neotropical regions (Constantino, 2002). They differ from other social insects in that they are divided into morphologically distinct castes with different work roles and biological functions; both sexes are diploid and hemimetabolous (Gallo et al., 2002).

The genus *Nasutitermes* is the largest in the world in terms of number of species and occurs in Bolivia, Venezuela, Paraguay, Uruguay and Brazil. In Brazil, its species can be found in regions of tropical forest, the Cerrado and Caatinga. Given its adaptability to urban settings, *N .corniger* is considered the primary pest in the genus *Nasutitermes* and causes significant economic losses, particularly in the construction and furniture industries (Constantino, 1999; Constantino, 2002).

Organo-synthetic insecticides such as bifenthrin, cypermethrin, chlorfenapyr, fipronil, imidacloprid and permethrin are the most widely used worldwide to control *Nasutitermes*, but their harmful effects have prompted a growing search for new alternatives to combat these pests (Lima et al , 2013; Specht et al., 2014). In this respect, natural plant-based products have proved to be highly effective at controlling termites, as well as being economically viable and having less impact on the environment and human health (Verma et al., 2009; Cruz et al., 2012). The most prominent of these are essential oils, whose proven biological functions in the plant kingdom include attracting pollinating agents,

Open access journal: http://periodicos.uefs.br/ojs/index.php/sociobiology

ISSN: 0361-6525

repelling insects and protecting against certain plant pathogens (Rozwalka et al., 2008; Silva et al., 2015; Miranda et al., 2016), in addition to being widely used in the chemical, pharmaceutical, food and cosmetics industries (Pascual et al., 2001).

Lippia is one of the most important genera of the family Verbenaceae, with 200 species described and widely distributed in the Neotropics, Brazil being the largest endemic region (81 species described) (Flora do Brasil, 2017). Some *Lippia* species have proven biological activity, such as the antimicrobial and antioxidant properties of *Lippia origanoides* (Pinto et al, 2013; Teles et al., 2014) or the spasmolytic, anti-diarrheal and antimicrobial activity of *Lippia thymoides* (Silva et al, 2015; Menezes et al, 2018).

The insecticidal efficiency of this genus has been reported for the species *Lippia gracilis* and *Lippia sidoides* against *Sitophilus zeamais* (Castro Coitinho et al., 2006), *Aedes aegypti* and *Culex quinquefasciatus* (Costa et al, 2005), respectively. Although these studies reinforce the insecticidal properties of *Lippia* species against other classes of arthropods, research on the insecticidal potential of other representatives of this genus remains scarce, particularly against termites (Evans & Iqbal, 2015; Santos et al., 2017). Thus, the present study aimed to investigate the chemical composition and insecticidal potential of essential oils of the species *Lippia lasiocalycina, Lippia insignis* and *Lippia thymoides* to control *Nasutitermes corniger* Motsch (Isoptera: Termitidae).

Materials and Methods

Plant material

The plant species used in the experiments were obtained from the Medicinal and Aromatic Plant Collection of the Forest Garden (Horto Florestal) Experimental Unit belonging to Feira de Santana State University (UEFS). The following were identified through exsiccates of the species deposited in the institutional herbarium (HUEFS): *Lippia thymoides* Mart. and Schauer - 115371; *Lippia lasiocalycina* Cham. – 197676 and *L. insignis* Moldenke – 197674. Leaves were collected during flowering, between 07:00 and 08:00 a.m., and dried out in the dark at room temperature for 10 days.

Essential oil (EO) extraction

The EOs were extracted by hydrodistillation, as described by Teles et al. (2014), with some modifications. The dried leaves (200 g) were ground in a blender and extracted for 3h in a modified Clevenger-type apparatus, using enough distilled water to completely cover the plant material. The oils extracted were dried out with anhydrous sodium sulfate.

The EO content was calculated on a dry weight basis using the equation described by Santos et al. (2004):

$$To = \frac{Vo}{Bm - \left(\frac{Bm\ xU}{100}\right)} \text{x } 100$$

where: To is oil content in %, Vo the volume of oil in mL, Bm the plant biomass (g), U the moisture content or water present in the biomass, and 100 the percentage conversion factor.

Determining chemical composition

In order to determine the chemical composition of the EOs, 20 mg of oil was previously diluted in 1 mL of dichloromethane. Quantification was performed by gas chromatography with a flame ionization detector (GC-FID) andthe components were identified by gas chromatography-mass spectrometry (GC-MS).

A Varian® CP–3380 chromatograph equipped with an FID and Chrompack CP-SIL 5 capillary column (30 m x 0.5 mm) was used for CG-FID analyses, with a film thickness of 0.25 μm, injector and detector temperatures of 220 °C and 240 °C, respectively, and helium as carrier gas (1 $mL.min^{-1}$). The oven temperature program was60 to 240 °C (3 $°C.min^{-1}$), with a 240 °C isothermal run for 20 min.The GC-MS analyses were carried out in a Shimadzu® GC-2010 chromatograph coupled to a Shimadzu® GC/MS-QP 2010 mass spectrometer, equipped with a DB-5ms capillary column (30 m x 0.25 mm, film thickness of 0.25 μm), using an injector temperature of 220 °C, helium as carrier gas (1 $mL.min^{-1}$), 240 °C interface temperature and ionization source, 70 V electron ionization, 0.7 kV voltage, and a similar temperature program to that described above.

The components were identified by calculating Kovats retention indices and comparing these and the mass spectra to reference standards and literature data (Adams, 2007). The relative percentages of the constituents were calculated by peak area normalization.

Insecticidal activity analysis

The insects were transported to the laboratory and placed in transparent 500 ml plastic containers, sealed with voile held in place by an elastic band. To prevent contamination, the containers were kept in plastic trays with water. A sample of the termites was identified and deposited in the UEFS Zoology Museum (MZUEFS).

Insecticidal activity was evaluated by exposure to a contaminated surface, using the EOs of *Lippia* species at 1% concentration, diluted in water, with Tween 20 as emulsifier. Ten *N. corniger* individuals were placed in 140 ml plastic containers (7.5 x 6.5 cm) lined with filter paper imbibed in 1.5 ml of solution containing the essential oils. To better accommodate the insects, wood shavings from the tree where they were collected and fragments of their own nest were placed in the containers. The containers were sealed with voile held in place by an elastic band, kept in a room at ambient temperature throughout the experiment, and covered with black fabric to protect them from the light. Insect mortality was assessed 24 and 48 hours after treatment application. Insects that moved any part of their body, even when stimulated, were considered alive (Santos et al., 2007).

A completely randomized design, consisting of 6 treatments (concentrations) for each *Lippia* species and 4 repetitions, was used, with each repetition represented by one container housing 10 insects.

Determining the Median Lethal Concentration (LC_{50})

To determine the lowest lethal concentration capable of killing 50% of the individuals (LC_{50}), the EOs were diluted in water to obtain six different concentrations (0; 0.625; 1.25; 2.5; 5; 10 µl/ml), using Tween 20 as dispersing agent. Four repetitions were used per treatment, each consisting of one container holding 10 insects. The Tween solution was used as control, in the same proportion.

Insecticidal activity analysis

Insecticidal activity was assessed using adult *Nasutitermes corniger* Motschulsky termites collected from nests in Brazil wood trees (*Caesalpiniae echinata* Lam). The insects were transported to the laboratory and placed in transparent 500 ml plastic containers, sealed with voile held in place by an elastic band. To prevent contamination, the containers were kept in plastic trays with water. A sample of the termites was identified and deposited in the UEFS Zoology Museum (MZUEFS).

Insecticidal activity was evaluated by exposure to a contaminated surface, using the EOs of *Lippia* species at 1% concentration, diluted in water, with Tween 20 as emulsifier. Ten *N. corniger* individuals were placed in 140 ml plastic containers (7.5 x 6.5 cm) lined with filter paper imbibed in 1.5 ml of solution containing the essential oils. To better accommodate the insects, wood shavings from the tree where they were collected and fragments of their own nest were placed in the containers. The containers were sealed with voile held in place by an elastic band, kept in a room at ambient temperature throughout the experiment, and covered with black fabric to protect them from the light. Insect mortality was assessed 24 and 48 hours after treatment application. Insects that moved any part of their body, even when stimulated, were considered alive (Santos et al., 2007). A completely randomized design, consisting of 6 treatments (concentrations) for each *Lippia* species and 4 repetitions, was used, with each repetition represented by one container housing 10 insects.

Statistical analysis

Given the lack of variability in the results of some treatments, at both 24 and 48h, the initial insecticidal activity data were submitted to nonparametric analysis via the Kruskal Wallis test for multiple comparisons of the treatments (doses administered). Significance was set at 5% probability.

Tests in which different doses or concentrations of a drug are administered to individuals are known as dose-response analyses (Demétrio, 2001), in which the status of these individuals changes within a certain period after administration (a dead insect indicates success and survival, failure). The aim of these experiments is generally to model the chances of success and determine effective or lethal concentrations (LCs). Nonlinear logistical regression with two parameters (β_1 and β_2) is used to estimate this probability. In order to estimate the lowest lethal concentration capable of killing 50% of the individuals (LC_{50}), the logistical model is linearized using the logit function. Thus, LC_{50} is determined by dividing the two parameters in the model:

$$LC_{50} = -\beta_1 / \beta_2$$

All the results were obtained using R software (R Core Team, 2017).

Results

EO content and chemical composition

The highest essential oil content was obtained in *L. thymoides* (2.9%), followed by *L. lasiocalycina* (1.8%) and *L. insignis* (1.7%). Analyses of the EO chromatograms of *L. thymoides*, *L. lasiocalycina* and *L. insignis* identified 83.80, 96.30 and 95.40 (%) of the chemical compounds, respectively. The major compounds observed in *L. thymoides* Eos were sesquiterpenes, namely (E)-caryophyllene (29.55%), caryophyllene oxide (8.17%), germacrene D (6.59%) and cis-calamenene (5.59%). Monoterpenes predominated in *L. lasiocalycina* and *L. insignis*, whereas the primary constituents in *L. lasiocalycina* were β-myrcene (31.17%), (E)-ocimenone (24.10%), p-cymene (7.17 %) and (Z)-ocimenone (6.51%). The major compounds in the EOs of *L. insignis* were (E)-ocimenone (26.11 %), limonene (14.73%), *β*-myrcene (12. 48 %) and*p*-cymene (7.24%) (Table 1).

Insecticidal activity

The in vitro results indicated that the EOs of the three species were highly efficient at controlling *N. corniger*, promoting up to 60% mortality after 48 hours.Significant differences in insect mortality were observed in all the treatments when compared to distilled water (35.0%) and distilled water and Tween (40.0%). However, insecticidal activity was still inferior to that of the commercial agrochemical, which exhibited 100% mortality. *L. thymoides* and *L. lasiocalycina* stood out for causing 100% termite mortality in the shortest time period (24 h) (Table 2 - Fig 1).

Determining the median lethal concentration

The median lethal concentration (LC_{50}) of the essential oils (1%) was estimated by individually adjusting the models. *L. lasiocalycina* EO caused 50% *N. corniger* mortality at the lowest concentration (0.46 µl/ml), followed by *L insignis* (0.88 µl/ml) and *L. thymoides* (3.64 µl/ml) (Table 3 - Fig 2).

Table 1. Chemical composition of essential oils extracted from the leaves of *Lippia insignis* (Moldenk), *Lippia thymoides* (Martius & Schauer) and *Lippia lasiocalycina* (Schauer).

Compound	KI_{lit}	KI_{calc}	LL (%) ± SD	LI (%) ± SD	LT (%) ± SD
α-thujene	930	927-28	T	0.24±0.02	T
α-pinene	939	939	T	T	1.62±0.04
Camphene	954	951	-	-	0.19±0.00
Sabinene	975	973-75	1.23±0.07	0.16±0.00	1.83±0.55
β-pinene	977	977-79	-	t	0.88±0.04
β-myrcene	990	989-91	31.17±1.16	12.43±0.02	0.38±0.06
α-phellandrene	1002	1004	-	-	T
α-terpinene	1017	1016-17	-	1.17±0.03	T
p-cymene	1026	1024-26	7.17±0.62	7.24±0.84	0.78±0.51
Limonene	1029	1029-31	0.31±0.00	14.73±0.60	2.75±0.16
Eucalyptol	1031	1034	-	-	5.17±0.47
Z-β-ocimene	1037	1038	T	-	-
E-β-ocimene	1050	1049	1.67±0.06	-	-
E-β-ocimene	1050	1048	-	1.78±0.04	T
γ-terpinene	1059	1059-61	2.29±0.02	6.99±0.14	0.48±0.14
cis-Sabinene hydrate	1070	1068	-	t	-
Terpinolene	1088	1089	-	0.19±0.01	-
Linalool	1096	1096-98	1.29±0.09	2.80±0.79	-
Chrisanthenone	1127	1125	T	-	-
trans-pinocarveol	1139	1140	-	-	0.18±0.00
trans-verbenol	1144	1142	-	-	T
Ipsdienol	1145	1144-46	0.43±0.05	0.85±0.12	0.20±0.00
Myrcenone	1149	1151-53	4.05±0.35	6.37±1.14	-
Borneol	1169	1163-66	1.34±0.05	-	0.43±0.02
terpinen-4-ol	1177	1177-79	-	t	0.56±0.16
α-terpineol	1188	1188-90	T	0.55±0.06	-
α-terpineol	1188	1189	-	-	T
Myrtenol	1195	1195	-	-	T
(Z)-ocimenone	1229	1231	6.51±0.13	5.29±0.49	-
Thymol methyl ether	1235	1237	-	T	-
(E)-ocimenone	1238	1240-41	24.10±0.80	26.11±1.05	
Geraniol	1252	1255	0.34±0.02	-	-
Geranial	1267	1271	0.66±0.08	-	-
Thymol	1290	1290-92	T	t	-
Carvacrol	1298	1297-98	-	0.21±0.01	T
δ-Elemene	1338	1339	-	-	0.25±0.10
α-Cubebene	1348	1352	-	-	0.79±0.09
Piperitenone oxide	1368	1368	-		-
α-copaene	1377	1379	-	-	3.49±0.60
β-bourbonene	1388	1387	-	-	0.42±0.11
β-cubebene	1388	1391	-	-	0.31±0.07
β-elemene	1388	1392	-	-	0.39±0.05
β-elemene	1390	1393	0.50±0.05	-	-
α-gurjunene	1409	1411	0.39±0.02	-	-
E-caryophyllene	1419	1420-26	4.00±0.35	1.90±0.43	29.55±0.47
γ-elemene	1436	1435	-	-	0.22±0.01
α-guaiene	1439	1442	2.51±0.27	-	-

Table 1. Chemical composition of essential oils extracted from the leaves of *Lippia insignis* (Moldenk), *Lippia thymoides* (Martius & Schauer) and *Lippia lasiocalycina* (Schauer). (Continuation)

Compound	KI_{lit}	KI_{calc}	LL (%) ± SD	LI (%) ± SD	LT (%) ± SD
trans-muurola-3,5-diene	1453	1453	-	-	0.55±0.15
α-humulene	1454	1455-57	1.43±0.14	0.52±0.12	2.65±0.12
Alloaromadendrene	1460	1464	-	-	0.79±0.11
germacrene D	1485	1481-85	0.41±0.04	1.97±0.60	6.59±2.98
trans-muurola-4(14),5-diene	1493	1493	-	-	0.99±0.16
Bicyclogermacrene	1500	1496-98	0.41±0.05	2.55±0.90	-
α-muurolene	1500	1499	-	-	0.63±0.70
β-bisabolene	1505	1504-09	T	-	0.54±0.09
Cuparene	1505	1507	-	-	2.18±0.57
α-bulnesene	1509	1508	1.22±0.20	-	-
Cubebol	1515	1516	-	-	0.43±0.10
cis-calamenene	1529	1526	-	-	5.59±0.99
trans-cadina-1,4-diene	1534	1535	-	-	0.42±0.04
germacrene B	1561	1561	-	-	2.53±0.72
Spathulenol	1578	1577-80	1.70±0.19	1.32±0.17	-
Caryophyllene oxide	1583	1583-88	1.77±0.40	0.29±0.04	8.17±3.40
α-muurolol	1646	1648	-	-	0.70±0.05
Total number of compounds identified			96.30±0.20	95.49±1.55	83.80±1.02
Content			1.8%	1.7%	2.9%

*KIlit = Kovats retention index from the literature; KIcalc = Kovats retention index calculated;
(-) compound absent from the sample.LT: *Lippia thymoides*, LL: *Lippia lasiocalycina*, LI: *Lippia insignis*.

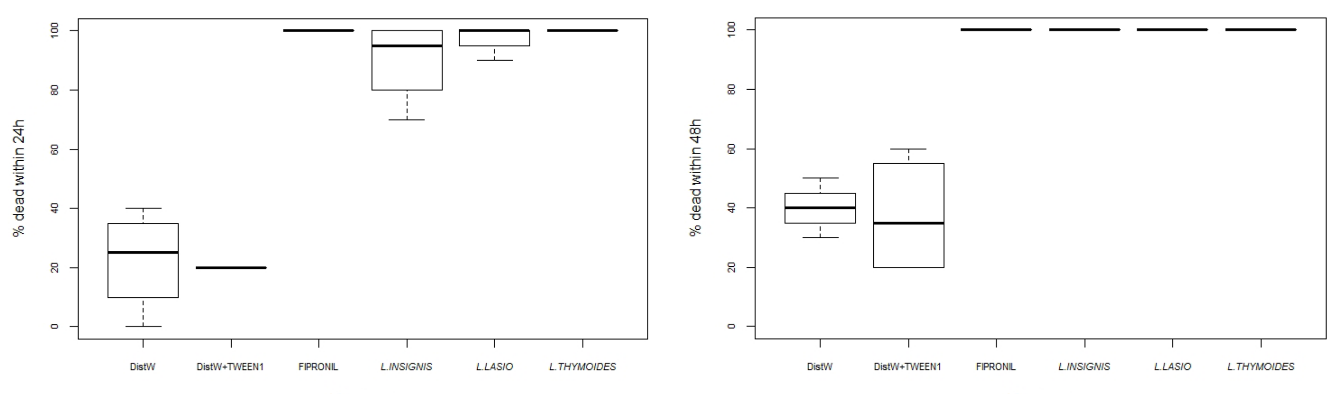

Fig 1. Mortality of *Nasutitermes corniger* (Motschulsky) 24 and 48 hours after exposure to a surface contaminated with essential oils of *Lippia. Insignis* (Moldenk)*, Lippia. Lasiocalycina* (Schauer) and *Lippia thymoides* (Martius & Schauer). DisW: distilled water; DisW+ TWEEN: distilled water and Tween (1%); FIPRONIL: commercial insecticide; *L lasio*: *Lippia lasiocalycina*.

Discussion

The results obtained demonstrate the insecticidal properties of EOs from the species *L. lasiocalycina, L. insignis* and *L. thymoides* in the biocontrol of *N. corniger*. Despite their inferior insecticidal activity when compared to the commercial agrochemical, essential oils are less harmful to the environment and do not promote the development of resistance in insects (Campos & Andrade, 2002).

This study is pioneering in that it assesses insecticidal activity against termites of *Lippia* species*,* which are still underexplored. The significant insecticidal results of the EOs studied can be justified by the presence of terpenes such as thymol, carvacrol, geranial, linalool, p-cymene, carvone, neral, limonene, (E)-caryophyllene, caryophyllene oxide, mircene. and γ-terpinene, whose insecticidal activity has been widely studied and confirmed (Gomes et al., 2011 ; Soares & Tavares-Dias., 2013; Kamanula, J. F., 2017; Blank, A. F., 2019).

Table 2. Mortality of *Nasutitermes corniger* (Motschulsky) 24 and 48 h after treatment with essential oils of *Lippia thymoides* (Martius & Schauer), *Lippia lasiocalycina* (Schauer) and *Lippia insignis* (Moldenk) at a concentration of 1%.

ESSENTIAL OILS		
TREATMENTS	**MORTALITY**	
	24 hrs	**48 hrs**
Lippia thymoides	100.0a	100.0a
Lippia lasiocalycina	100.0a	100.0a
Lippia insignis	95.0a	100.0a
Distilled water and Tween 20 (1%)	20.0b	40.0b
Distilled water	25.0b	35.0b
Fipronil (c+)	100.0a	100.0a

*Data were submitted to nonparametric analysis via the Kruskal Wallis test and subsequently compared by rank. Medians followed by the same lowercase letter in the column do not differ significantly.

Table 3. Equations used to determine the medial lethal concentration (CL50) of EOs of *Lippia thymoides* (Martius & Schauer); *Lippia lasiocalycina* (Schauer) and *Lippia insignis ((*Moldenk) against *Nasutitermes corniger* (Motschulsky).

Species	CL_{50} (µl/ml)	Logit	r2
L. insignis	0.88	$Y = -1.926 + 2.179\ c_i$	0.74
L. thymoides	3.63	$Y = -1.610 + 0.443\ c_i$	0.719
L. lasiocalycina	0.46	$Y = -0.841 + 1.800\ c_i$	0.787

The insecticidal properties of essential oils of *L. lasiocalycina, L. insignis* and *L. thymoides* corroborate the findings of Lima et al. (2013), who studied the action of OEs of *L. alba, L. gracilis* and *L. sidoides* against *N. corniger*, with *L sidoides* proving to be particularly effective. The authors attributed this activity to their high thymol content and synergistic effects of *p*-cymene and thymol methyl ether. Tests using different chemotypes of *L. gracillis* against *Diaphania hyalinata* (Lepidoptera: Pyralidae) demonstrated that chemical races consisting primarily of the phenolic terpenes thymol and carvacrol were more toxic (Melo et al., 2018). Essential oil of *L. origanoides* Kunth, rich in carvacrol and p-cymene, was also efficient at controlling *Aedes aegypt* Linn, *Tetranychus urticae* Koch. and *Cerathapis lataneae* Boisd (Mar et al., 2018).

Terpenes, the predominant compounds in *Lippia* essential oils, encompass a series of substances whose defensive properties in plants are well-known. These metabolites protect the plants that produce them through neurotoxic action by inhibiting acetylcholinesterase, the main neurotransmitter responsible for acetylcholine reuptake in the synaptic cleft (Tak & Isman, 2017). Another effect of EOs is their inhibition of the neuromodulator octopamine, leading to hyperpolarization of the calcium channels modulated by GABA (gamma-aminobutyric acid) (Priestley et al., 2003). Octopamine acts as a neurohormone and neurotransmitter, responsible for regulating the heart rate, behavior and metabolism of insects (Castro Coitinho et al., 2011; Enan et al., 200101). The toxicity of *Lippia* EOs causes delayed growth and appetite suppression in insects, as well as hampering maturation and reducing their reproductive capacity, leading to death (Viegas Júnior, 2003).

Although the phenolic terpene content in the species studied here was low, the presence of other compounds with proven insecticidal activity, such as limonene, p-cymene and ß-myrcene, may have contributed to this finding. Limonene is one of the most common components of pesticides such as insecticides and insect repellent, and its presence has been

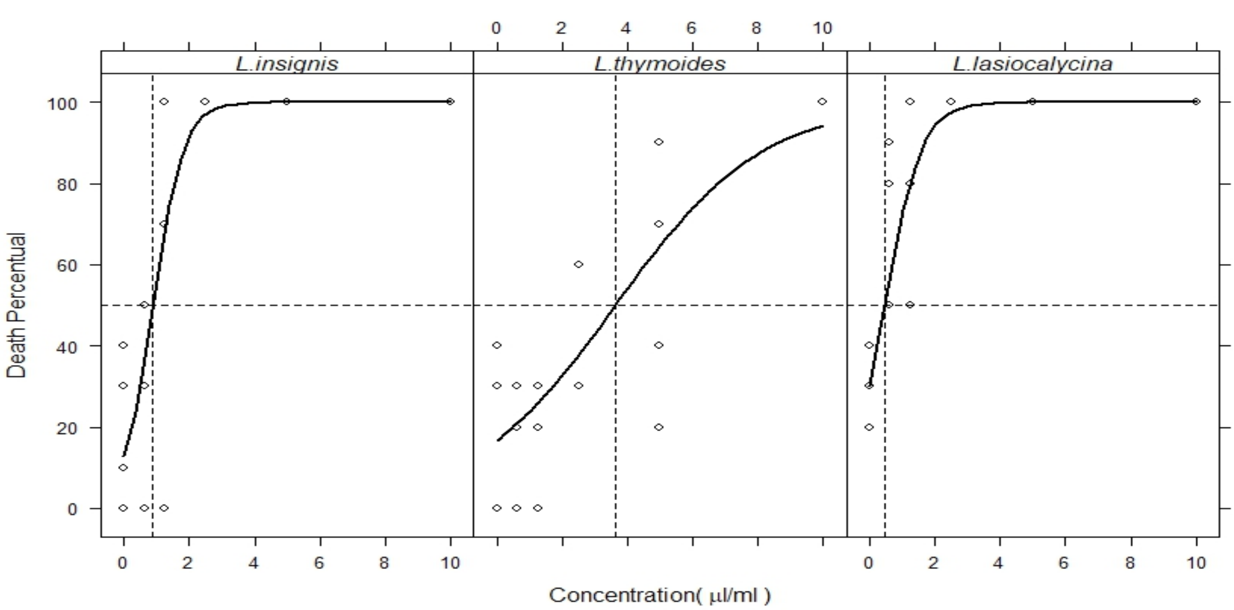

Fig 2. Median Lethal Concentrations (LC_{50}) of essential oils from *Lippia insignis* (Moldenk)*, Lippia thymoides* (Martius & Schauer) and *Lippia lasiocalycina* (Schauer) against *Nasutitermes corniger* (Motschulsky).

reported in *Lippia alba* EO, a species proven to be effective against *Spodoptera frugiperda* (J. E. Smith) (Niculau et al., 2013). P-cymene, found in *Lippia sidoides* and *Lippia lasiocalycina*, exhibits proven insecticidal action against *Aedes aegypti* (Diptera: Culicidae) and *Heterotermes sulcatus* Mathews (Isoptera: Rhinotermitidae) (Costa et al., 2005). However, in termites, pinenes have been identified as the most volatile components in defending against *N. Corniger*, enhancing the chemical response of these insects to pinene-rich products (Lima et al., 2013). β-myrcene was active against adult *Sitophilus zeamais* with a dose-dependent relationship to exposure time (Yildirim et al., 2013).

The median lethal concentrations (LC_{50}) recorded in our study demonstrated that OEs of *L. lasiocalycina* (0.46 µl/ ml) and *L. insignis* (0.88 µl/ ml) were more effective after 48 hours, indicating that very little oil is needed from these species to obtain a toxic response in *N. corniger,* using the contact method. Another study investigated the median toxicity of *Lippia sidoides* Cham and *Pogostemon cablin* Benth EOs against the termites Microcerotermes indistinctus and Amitermes A. cf. *amifer.* The authors found that Pogostemon cablin EO was the most effective (LC_{50}) at 0.32 µl/ ml and 0.29 µl/ ml, concentrations lower than those observed here (Bacci et al.,2015), while *L. sidoides* was less effective, with a median effect on the termites tested at 2.41 and 3.47 µl/ ml, respectively. The insecticidal activity of *L sidoides* against *Cryptotermes brevis* (Walker, 1853) (Kalotermitidae) has also been reported using the fumigation test, with CL_{50} estimated at 9.10 and 23.6 µL/ L of beans (Santos et al., 2017). It is important to note that individual characteristics such as cuticle thickness, associated with different application methods, can influence inter or intragroup tolerance and resistance (Haddi et al., 2015; Pinto-Zevallos & Zarbin, 2013).

Given that their social behavior is one of the main barriers to successful termite control, in most cases the topical use of synthetic insecticides leads to recurrence (Lima et al., 2013). As such follow-up studies should be conducted in the field to chemically control this insect pest, in conjunction with the partition and isolation of the chemical constituents of the oils tested, as well as preparing stable essential oil solutions from the *Lippia* species studied.

Conclusions

The essential oils of *L. lasiocalycina*, *L. insignis* and *L. thymoides* showed *in vitro* insecticidal potential to control *N. corniger* termites*. L. lasiocalycina* EO exhibited the lowest median lethal concentration, followed by *L. insignis* and *L. thymoides*. In conclusion, this study demonstrates the potential of developing bioinsecticides from the EOs studied to control termite populations, particularly considering the substantial toxicity to humans of the synthetic products currently used, their significant environmental impact and the high recurrence of pests after topical treatment with these products due to their social behavior.

Acknowledgments

The authors are grateful to the Research Support Foundation for Bahia State (FAPESB) and the National Council for Scientific and Technological Development (CNPq) for the funding provided to carry out this project. We would also like to thank Feira de Santana State University (UEFS) and its Graduate Program in Plant Genetic Resources (PPGRGV/UEFS.

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