



# Avocado kernels, an industrial residue: a source of compounds with insecticidal activity against silverleaf whitefly

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## Abstract

Fruit processing waste, such as kernels (endocarp + seed) of avocado [*Persea americana* Mill. (Lauraceae)], could be used as raw material in the preparation of botanical insecticides. In light of this potential, this study assessed the insecticidal action of extracts and fractions from kernels of two avocado cultivars (Breda and Margarida) on *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) biotype B, an important pest species in tropical conditions. Ethanolic and aqueous extracts prepared from kernels of *P. americana*, regardless of the plant cultivar used, caused promising insecticidal activity to whitefly nymphs. Based on yield in crude extracts [10.32 and 9.85% (w/w), respectively, for cultivars Breda and Margarida], on the bioassay results with crude extracts and on the chemical profiles, the ethanolic extract of kernels of *P. americana* cv. Breda was chosen for the continuation of the study. Thus, the ethanolic extract of kernels of cv. Breda (LC<sub>50</sub> = 197.84 ppm and LC<sub>90</sub> = 567.19 ppm) was selected and subjected to fractionation by the liquid-liquid partition technique. The hexane and dichloromethane fractions of this extract caused significant mortality of nymphs. The analysis using the ultraviolet (UV) and hydrogen nuclear magnetic resonance (<sup>1</sup>H NMR) showed the presence of long-chain aliphatic compounds (alkanols or acetogenins of Lauraceae), alkylfurans (or avocadofurans), and unsaturated fatty acids in these fractions, which are possibly related to bioactivity observed in *B. tabaci*, besides saccharides. The results show that kernels of *P. americana* are promising sources of compounds with insecticidal action for the control of *B. tabaci* biotype B, a great opportunity to transform environmental problems into eco-friendly solutions to agriculture.

**Keywords** *Bemisia tabaci* biotype B · Allelochemicals · Avocadofurans · Bioinsecticide · Industrial residue

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## Introduction

Agro-industrial activities routinely generate a large amount of liquid and solid waste, usually rich in organic material that could cause serious environmental and health problems, in addition to economic costs required for correct disposal and treatment (Infante et al. 2013). One alternative to mitigate these drawbacks is to use this waste to produce by-products, which can also promote business diversification and add value to products, crucial aspects of sustainability for agro-industrial activities.

Brazil is the world's 6th largest producer of avocado *Persea americana* (Lauraceae) (Fao 2017), and its commercial cultivation is allocated mainly to supply the food, cosmetic, and medicine industries (Francisco and Baptistella 2005). However, regardless of the industrial segment, only the pulp (mesocarp) is used for commercial purposes, while kernels (endocarp + seed) are discarded. Besides the great abundance and availability at low cost, a limited number of studies have investigated ways of using avocado kernels.

Several classes of natural compounds have been isolated from *P. americana*, such as alkanols (sometimes called aliphatic acetogenins or acetogenins of Lauraceae), alkylfurans (avocadofurans), fatty acids, triglycerides, phytosterols, triterpenes, flavonoids, dimers of flavonoids, saccharides, proanthocyanidins, glycosylated abscisic acids, and derivatives from benzoic and cinnamic acids and from phenols (Rodríguez-Saona et al. 1999; 1998, b; Ding et al. 2007; Leite et al. 2009; Dabas et al. 2013; Rodríguez-Sánchez et al. 2013; Ragasa et al. 2014). To date, biological studies carried out with extracts and compounds obtained from kernels of *P. americana* have detected promising antimicrobial activities (Néman et al. 1970; Leite et al. 2009; Chia and Dykes 2010; Cardoso et al. 2017), antiprotozoal (Abe et al. 2005; Jiménez-Arellanes et al. 2013) and insecticidal effects against pest species of medical (Leite et al. 2009; Adesina et al. 2016), and agricultural (Stein and Klingauf 1990; Rodríguez-Saona et al. 1998; Santa-Cecília et al. 2010) importance, which provided the hypothesis on their use in the production of a bioinsecticide for the control of whitefly *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) biotype B. *B. tabaci* is an important pest species of polyphagous feeding habit that has caused major economic losses in Tropical America (Gilbertson et al. 2015; Barbosa et al. 2016; Fariña et al. 2019) due to the inefficiency of chemical control, the main method used for its control, especially because of the large number of populations resistant to active ingredients registered (Nauen and Denholm 2005; Naveen et al. 2017; Dângelo et al. 2018; Dângelo et al. 2018).

Several studies have investigated plant derivatives with potential use in the management of *B. tabaci* biotype B in tropical climate (Emilie et al. 2015; Deletre et al. 2016; Fogné et al. 2017). However, plant extracts often require cutting plant structures, which can be hindered by the scarcity of raw material. Thus, the use of waste of fruit processing is of great interest, since it provides useful raw materials that are economically viable for bioinsecticide production (Ribeiro et al. 2016; Ansante et al. 2017; Bernardi et al. 2017; Souza et al. 2019). In light of this potential, this study assessed the insecticidal action of extracts and fractions from kernels (endocarp + seed) of *P. americana*, from two cultivars (Breda and Margarida), on nymphs of *B. tabaci* biotype B. In addition, chemical analyses were carried out to identify the classes of compounds responsible for the bioactivity observed.

## Material and methods

### Plant material and extracts preparation

For extracts preparation, kernels of *P. americana* (cultivars Breda and Margarida) were obtained from fruits collected in July 2010, on the Cafetotal Farm, municipality of São

Sebastião do Paraíso, Minas Gerais, Brazil (Lat. 20° 54' 32" S, Long. 46° 58' 36" W, 1150 m above sea level). The kernels were dried in the sun and collected for milling when the moisture content was estimated between 10 and 15%. The kernels were then milled with a knife-hammer grinding system, and the powder was stored in hermetically closed glasses until use.

For preparation of aqueous extracts, the cold maceration method at a ratio of 1:5 [vegetable powder/deionized water (w/v)] was used. The solutions were shaken for 10 min and placed to rest for 24 h, and later, the material was filtered in filter paper. The remaining pie was again subjected to extraction, using the same ratios (vegetable powder/solvent). This procedure was repeated twice, totaling three filtering events. The filtered material was frozen immediately and then freeze-dried for 5 days to remove the liquid and obtain the crude water extract.

To obtain the ethanolic extracts, we also used the cold maceration technique at a ratio of 1:5 [vegetable powder/organic solvent (w/v)], using the same procedure described previously. However, we performed four filtering events at intervals of 72 h. At every change of solvent, the macerate was filtered through paper filter, and the solvent of the remaining sample was eliminated in a rotary evaporator at 50 °C and –600 mmHg pressure. After complete evaporation of the solvent in a chamber with airflow, the extraction yield was determined.

### Bioassays

All bioassays were conducted in the laboratory under controlled conditions (temperature  $25 \pm 2$  °C, relative humidity  $60 \pm 10\%$  and photophase of 14 h).

To assess the insecticidal activity of extracts, newly extended apical leaflets of tomato (cv. Santa Clara) were individualized and wrapped by a cylindrical cage (15 cm long and 6 cm diameter) made of thin fabric (*voile*), where 30 adults of whitefly *B. tabaci* biotype B from maintenance rearing were released and held for 24 h for oviposition. Afterwards, the adults were removed and plants with the leaflets selected were kept in the greenhouse until the complete embryonic development of the insect. After hatching and fixing the first instar nymphs, the extracts were applied on both faces of the leaflets that contained the nymphs through a microatomizer coupled with a vacuum pump, using a pressure spray  $0.5 \text{ kgf cm}^{-2}$ , and 2 mL of the solution were applied (dripping point) to the leaflets (repetition). After complete drying, the leaflets were removed from the plants and placed in glass tubes ( $8 \times 2 \text{ cm}$ ) containing deionized water and kept in a laboratory under the conditions mentioned above. Seven days after application, mortality of nymphs on the abaxial face of leaflets was evaluated with the aid of a stereoscopic microscope.

For the bioassay with the freeze-dried aqueous extracts, the concentration used was 20,000 ppm ( $\text{mg kg}^{-1}$ ), with six

repetitions (leaflets) per treatment. For the ethanolic extracts, the concentration used was 10,000 ppm ( $\text{mg kg}^{-1}$ ), with 10 repetitions (leaflets) per treatment. For both cases, concentrations were defined on preliminary tests. The control treatment comprised the same solvents used in resuspension of aqueous and ethanolic extracts (deionized water and acetone/deionized water [1:1 (p/p), respectively]). In view of the unavailability of effective alternative insecticides registered in the Brazilian market and impracticability of use of synthetic formulations in laboratorial tests, a positive control was not used in this modality of trial. In this step, our main objective was to assess the potential of avocado derivatives and characterize the chemical class of bioactive compounds. In the next step of our research program, the promising derivatives will be tested in semi-field and field conditions in order to compare the potency in relation to synthetic insecticides (positive controls).

### Chromatographic profiles and spectroscopy analysis of crude extracts

Chromatographic qualitative profiles of crude ethanolic extracts of *P. americana* cvs. Breda and Margarida were obtained by high performance liquid chromatography (HPLC) using a liquid chromatograph of Agilent Technologies 1200 model, equipped with a quaternary pump (G1311A), a degasser (G1322A), an automatic injector (G1329A), and a UV detector (G1314B). The equipment was coupled with an interface (G1369A), and the chromatograms were recorded using EZChrom Elite software. The chromatograms were obtained using an eclipse C18 column ( $150 \text{ mm} \times 4.6 \text{ mm} \times 5 \mu\text{m}$ ), using a flow of  $0.8 \text{ mL min}^{-1}$ , with detections at wavelengths 224, 254, and 312 nm. Elution was held as follows: MeOH:H<sub>2</sub>O (1:9, v/v) isocratic for 5 min, MeOH:H<sub>2</sub>O (1:1, v/v) isocratic for 15 min, and MeOH:H<sub>2</sub>O (95:5, v/v) isocratic for 20 min. The samples were tested at a concentration of  $100 \mu\text{g mL}^{-1}$ , and the injection volume was 20  $\mu\text{L}$ .

Profiles of hydrogen nuclear magnetic resonance (<sup>1</sup>H NMR) of ethanolic crude extracts of *P. americana* cvs. Breda and Margarida were obtained through the Spectrometer Bruker Avance™ III NanoBay 9.4 T (400 MHz) using the deuterated solvent CDCl<sub>3</sub>, and chemical shifts were reported in units  $\delta$  using TMS as internal standard ( $\delta_{\text{H}}$  0.0 ppm).

### Concentration-response curve of most promising extract

Based on the extraction yield, on the results of bioassays and on the chemical profile of kernel extracts of both avocado cultivars studied, obtained through the HPLC analysis, we selected the most promising extract to estimate lethal concentrations. To this end, preliminary tests were carried out with this extract to check basic concentrations that caused approximately 95% of mortality of nymphs of *B. tabaci* biotype B. We also checked

concentrations that caused mortality similar to that obtained in the control, according to the method described by Finney (1971). After establishing the concentrations (range: 0–501.18 ppm), the same procedure of bioassay and experimental conditions described previously were applied.

### Bioguided fractionation of the promising extract

To identify the classes of compounds responsible for bioactivity observed in the most promising crude extract, we used fractionation by the liquid-liquid partition technique. For this, the selected extract was resuspended twice in methanol:water (1:3, v/v) and submitted to the partition using a separation funnel and organic solvents of increasing polarity (hexane, dichloromethane, ethyl acetate, and hydroalcoholic). Partitions (fractions) obtained were concentrated in rotary evaporator under the same conditions of temperature and pressure already mentioned.

The organic fractions obtained were tested to evaluate the effect of each partition on the survival of nymphs of *B. tabaci* biotype B, adopting the same experimental procedure described earlier. The concentration used was the LC<sub>50</sub> estimated in the previous bioassay.

### Data analysis

All bioassays were conducted in a completely randomized design. The data obtained were analyzed for normality, homoscedasticity, and the presence of outliers. Whenever necessary, the data were transformed according to the method of maximum power of Box-Cox (Box and Cox 1964). Afterwards, the data were subjected to analysis of variance (ANOVA) by the *F* test, and when there was a significant difference, the means were compared by the Tukey test ( $p < 0.05$ ). All analyses were conducted using the statistical package SAS version 9.2 (SAS Institute Inc. 2008).

Estimates of lethal concentrations were performed by means of the Probit analysis (Finney 1971), using the program Polo Plus 1.0 (LeOra software 2003).

## Results

### Lethal toxicities of crude derivatives from kernels of *Persea americana*

The extracts obtained from kernels of *P. americana* showed promising insecticidal activity for 1st instar nymphs of *B. tabaci* biotype B, both in aqueous ( $F = 8.28$ ,  $p = 0.0064$ ; at 20,000 ppm) (Table 1) and ethanolic solvents ( $F = 2162.50$ ,  $p < 0.0001$ ; at 10,000 ppm) (Table 2). Thus, because the bioactivity of extracts was similar and the concentration of aqueous extract was the double than that of the ethanolic extract,

we can infer that the ethanolic solvent has a greater capacity to extract active compounds in avocado kernels than the aqueous extract. Comparing both avocado cultivars (Breda and Margarida), there was no significant difference in mortality on *B. tabaci* biotype B nymphs exposed by 7 days to ethanolic crude extracts prepared from both avocado cultivars (Tables 1 and 2).

Probit analysis of obtained data showed that toxicity of ethanolic extract prepared from kernels of *P. americana* cv. Breda was concentration dependent. For this extract, the estimated LC<sub>50</sub> and LC<sub>90</sub> were 197.84 and 567.19 ppm, respectively (Table 3, SM-Table1). For aqueous extracts, it was not possible to estimate the lethal concentration values because within the range of concentrations used (0–10,000 ppm), insufficient mortality was observed.

### Chromatographic profiles and spectroscopy analysis of ethanolic extracts from kernels of avocado cultivars

HPLC (Fig. 1a and b) and <sup>1</sup>H NMR (Fig. 2a and b) analysis of ethanolic extracts prepared from cultivars Breda and Margarida of *P. americana* showed similar profiles, both at the peaks of most compounds and in the abundance of substances in the extracts prepared from both cultivars. In the exploratory elution gradient, measurements were made using wavelengths at 224 nm for the detection of aliphatic acetogenins (or acetogenins of Lauraceae), alkylfurans (avocadofurans), fatty acids, triglycerides, phytosterols, compounds, and saccharides; at 254 nm for the detection of proanthocyanidins and glycosylated abscisic acids, benzoic acids, derivatives of cinnamic acids, and phenols; at 312 nm for detection of flavonoids, dimers of flavonoids and coumarin (Kaouadji et al. 1992; Santos et al. 2005), corresponding to the absorption of compounds previously isolated of extracts of avocado fruit (Rodríguez-Saona et al. 1998, b, 1999; Ding et al. 2007; Leite et al. 2009; Dabas et al. 2013; Rodríguez-Sánchez et al. 2013; Ragasa et al. 2014). Chromatograms A

**Table 1** Mortality (±SE) of nymphs of *Bemisia tabaci* biotype B exposed to aqueous extracts of freeze-dried kernels of two cultivars of *Persea americana* (20,000 ppm)

Treatments	N	Mortality (%) <sup>*1</sup>
<i>P. americana</i> cv. Breda	413	90.00 ± 2.06 a
<i>P. americana</i> cv. Margarida	411	89.09 ± 3.43 a
Control (deionized water)	413	0.86 ± 0.57 b

<sup>\*</sup> Means followed by the same letter did not differ significantly by the Tukey test (α = 0.05)

Temp.: 25 ± 2 °C; RH: 60 ± 10%; photophase of 14 h

<sup>1</sup> Means of original data. For the statistical analysis, the data were transformed by the method of maximum power of Box-Cox (Box and Cox 1964)

**Table 2** Mortality (±SE) of nymphs of *Bemisia tabaci* biotype B exposed to ethanolic extracts of freeze-dried kernels of two cultivars of *Persea americana* (20,000 ppm)

Treatments	N	Mortality (%) <sup>*1</sup>
<i>P. americana</i> cv. Breda	387	95.3 ± 1.86 a
<i>P. americana</i> cv. Margarida	383	98.3 ± 1.12 a
Control [acetone: eionized water (1:1)]	390	0.9 ± 0.46 b

<sup>\*</sup> Means followed by the same letter did not differ significantly by the Tukey test (α = 0.05)

Temp.: 25 ± 2 °C; RH: 60 ± 10%; photophase of 14 h

<sup>1</sup> Means of original data. For the statistical analysis, the data were transformed by the method Box-Cox Power

and B (Fig. 1) have great similarity to each other, both in terms of constituents (number of peaks) and quantity (intensity of peaks). Chromatograms at 312 nm feature only peaks of very low intensity, indicating that the concentration of flavonoids and coumarin is very low or absent in these crude extracts. The chromatograms using wavelength at 254 nm characterize the compounds with n-π transitions of ketones, carboxylic acids, and secondary transitions of aromatic compounds (proanthocyanidins and glycosylated abscisic acids, acid benzoic derivatives, cinnamic acids, and phenols).

Chromatograms using wavelength at 224 nm can detect majority compounds of extracts of cultivars of *P. americana* [aliphatic acetogenins (alkanols or acetogenins of Lauraceae), alkylfurans (avocadofurans), fatty acids, triglycerides, phytosterols, triterpenes, and saccharides (Rodríguez-Saona et al. 1998, b, 1999; Ding et al. 2007; Leite et al. 2009; Dabas et al. 2013; Rodríguez-Sánchez et al. 2013; Ragasa et al. 2014)]. The presence of broad and intense bands in the chromatograms obtained with the wavelength at 224 nm, with retention times between 3 and 4 min, indicates high concentrations of aliphatic acetogenins (or acetogenins of Lauraceae), alkylfurans (or avocadofurans), fatty acids, and saccharides. These observations are confirmed in the discussion using spectra of <sup>1</sup>H NMR.

The <sup>1</sup>H NMR spectra of ethanolic extracts from cultivars Breda and Margarida (Fig. 2 a and b) were obtained and present great similarity to each other, both regarding constituents (signs of chemical shifts and position) and quantity (intensity of signs). These spectra show signs of more intense chemical shifts in δ<sub>H</sub> 0.9, 1.2, 2.0 [characteristic of long chain hydrocarbons in aliphatic acetogenins (alkanols or acetogenins of Lauraceae), alkylfurans (avocadofurans), and fatty acids, which are common in *P. americana* (Rodríguez-Saona et al. 1998; Alexandri et al. 2017)]. Intense signals between δ<sub>H</sub> 3.1–4.0 and doublets in δ<sub>H</sub> 4.1, 4.5, and 4.8 feature high concentration of carbohydrates in the extracts (Tesfay 2009; Rönnols et al. 2013). Low intensity

**Table 3** Estimation of the LC<sub>50</sub> and LC<sub>90</sub> (in ppm) and confidence interval of ethanolic extract of *Persea americana* cv. Breda for nymphs of *Bemisia tabaci* biotype B

$n^1$	Slope ( $\pm$ SE)	LC <sub>50</sub> (CI) <sup>2</sup>	LC <sub>90</sub> (CI) <sup>2</sup>	$\chi^2$ <sup>(3)</sup>	$d.f.$ <sup>4</sup>	$h^5$
2470	2.80 $\pm$ 0.16	197.84 (150.58–240.10)	567.19 (429.61–976.71)	4.53	2	2.26

<sup>1</sup>  $n$ : number of insects tested

<sup>2</sup> CI: confidence interval at 95% of error likelihood

<sup>(3)</sup>  $\chi^2$ : values of chi-square calculated

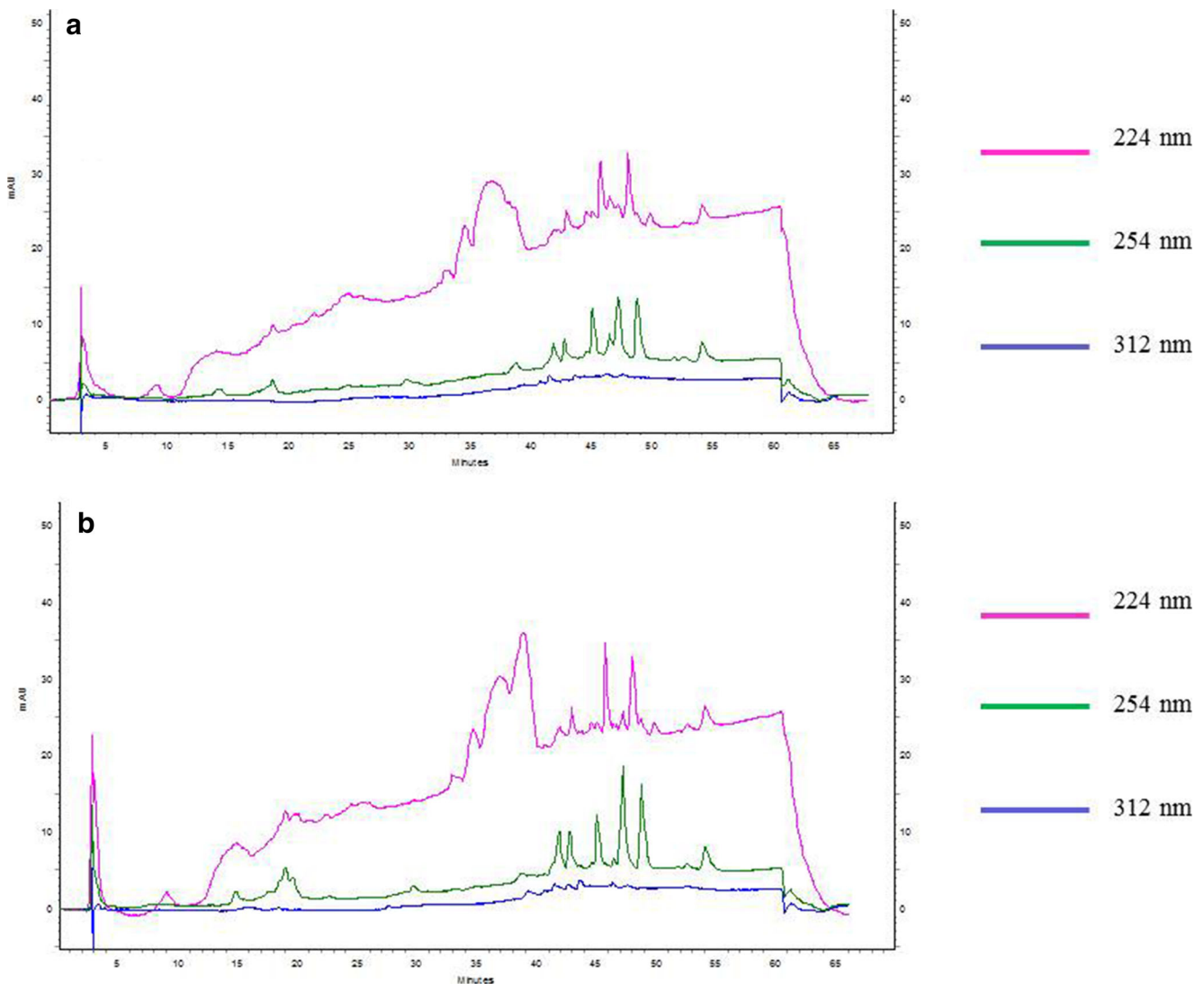
<sup>4</sup>  $d.f.$ : degree of freedom

<sup>5</sup>  $h$ : heterogeneity factor

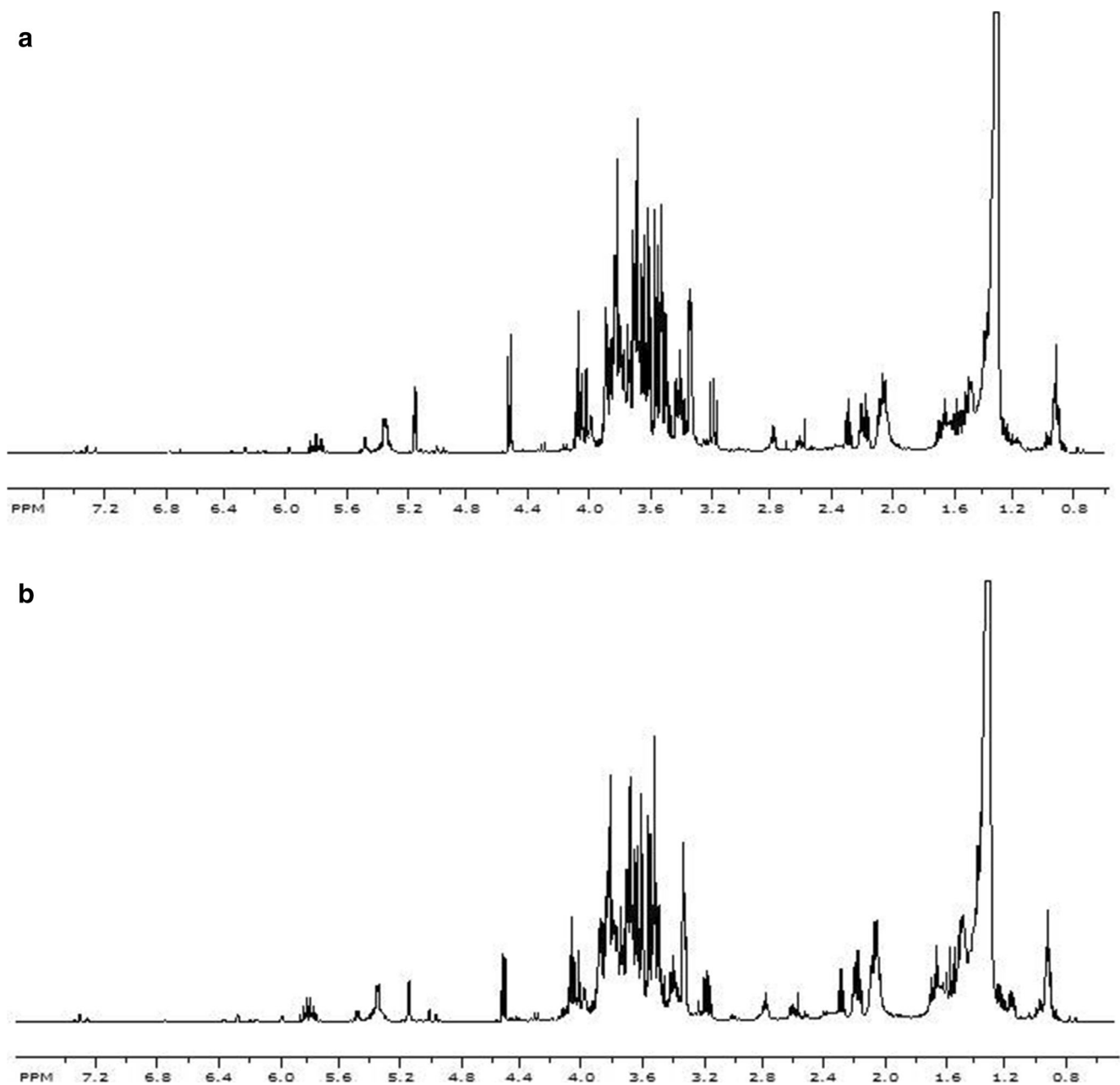
Temp.: 25  $\pm$  2 °C; RH: 60  $\pm$  10%; photophase of 14 h

signals in  $\delta_H$  5.37 (m), 6.28 (m), and 7.29 (d distorted) are characteristic of alkylfurans with unsaturation and indicate their presence in extracts, as reported earlier by Rodriguez-Saona et al. (1998). The absence of signal

in  $\delta_H$  5.24, characteristic of hydrogen methionine of glyceryl (CHO, Alexandri et al. 2017), indicates the absence of triglycerides in detectable quantities in ethanolic extracts of both avocado cultivars.



**Fig. 1** Liquid chromatogram obtained by high performance liquid chromatography (HPLC) of ethanolic extracts of *Persea americana* cultivars Breda (A) and Margarida (B) at wavelengths 224, 254, and 312 nm



**Fig. 2**  $^1\text{H}$  NMR spectra of ethanolic extracts of *Persea americana* cultivars Breda (A) and Margarida (B) ( $\text{CD}_3\text{OD}$ , 400 MHz)

### Insecticidal effects of semi-purified fractions

Based on yield in crude extracts [10.32 and 9.85% (w/w), respectively, for cultivars Breda and Margarida], on the bioassay results with crude extracts and on the chemical profiles, we chose the ethanolic extract prepared from kernels of *P. americana* cv. Breda for the continuation of the study.

After fractionation of the ethanolic extract of kernels of cv. Breda, using the technique of liquid-liquid partition, significant mortality was observed [ $F = 0.0001$ ,  $p < 226.30$ ; at previous estimated  $\text{LC}_{50}$  (197.84 ppm)] of nymphs of *B. tabaci* biotype B exposed to hexane and dichloromethane fractions

of the respective extract; however, without occurring differences between both (Table 4).

### Discussion

Our results showed the potential use of agro-industrial wastes from avocado chain for the production of a bioinsecticide, adding value to the business, besides enabling the correct allocation of environmental contaminants. The use of extracts from kernels of *P. americana* in the management of whitefly populations could be reached both through the formulation of

**Table 4** Mortality ( $\pm$ SE) of nymphs of *Bemisia tabaci* biotype B exposed at fractions of ethanolic extract of kernels of *Persea americana* cv. Breda

Fractions	N	Mortality (%) <sup>*1</sup>
Hexane	440	80.85 $\pm$ 3.69 a
Dichloromethane	427	81.34 $\pm$ 2.82 a
Ethyl acetate	434	9.35 $\pm$ 1.30 b
Hydroalcoholic	440	9.94 $\pm$ 2.77 b
Control [acetone: deionized water (1:1)]	437	5.65 $\pm$ 2.04 b

\* Means followed by the same letter did not differ significantly by the Tukey test ( $\alpha = 0.05$ )

<sup>1</sup> Means of original data. For the statistical analysis, the data were transformed by the method Box-Cox Power

Temp.: 25  $\pm$  2 °C; RH: 60  $\pm$  10%; photophase of 14 h

a commercial bioinsecticide and, in this case, with an adequate quality control and continuous offer during the year, and through homemade preparations produced by farmers from the material available on their farms, reducing thus technological dependence, an important aspect especially for small farmers in underdeveloped and developing countries (Pavela 2016; Pavela and Benelli 2016). In this context, despite a lower extraction capacity of active compounds compared with ethanolic solvent, water should be preferentially used because of economic costs and preparation facilities. Alternatively, isolation and structural determination of active compounds in extracts of *P. americana* kernels could provide possible model prototypes for synthesis of insecticides that are more environmentally suitable.

Strategically, plants of the genus *Persea* biosynthesize secondary compounds that mediate their interaction with the environment and their defense against herbivores, accumulating the compounds in differentiated and specialized cells called idioblastic oil cells (Rodríguez-Saona and Trumble 2000). These cells are distributed in different structures, such as seeds, fruits, stems, peduncles, leaf blades, and root tissues (Platt and Thomson 1992; Rodríguez-Saona et al. 1998; Domergue et al. 2000). A greater concentration of these cells is observed in the fruit mesocarp, which can represent up to 2% of the pulp (Cummings and Schroeder 1942). Therefore, as the pulp is widely consumed *in natura* or processed and avocado oil is widely used in the production of cosmetics, with no reports of harmful effects to human health, these compounds are expected to show no toxicity to mammals, which should be investigated in future research.

To date, studies have isolated fatty acids, triglycerides, and aliphatic acetogenins (acetogenins of Lauraceae) from idioblastic cells, more specifically the persin, isopersin, and compounds known as avocadofurans (Rodríguez-Saona et al. 1998; Rodríguez-Saona and Trumble 2000). In this study, based on analysis of <sup>1</sup>H NMR spectra, we verified the

presence of compounds of aliphatic acetogenins (acetogenins of Lauraceae), alkylfurans (or avocadofurans), and fatty acids in the active fractions of ethanolic extracts of *P. americana*, cultivars Breda and Margarida, which are possibly associated to the bioactivity observed on nymphs of *B. tabaci* biotype B. Corroborating this study, Rodríguez-Saona et al. (1998) found toxic and phagodeterrent effects of avocadofurans and triolein triglycerides to larvae of *Spodoptera exigua* (Hübner) (Lepidoptera: Noctuidae). The activity (synergy) was increased through the incorporation of compounds belonging to both classes into the artificial diet of that pest species. Thus, avocadofurans and triolein with insecticidal properties were subsequently patented by the authors (Thomson et al. 2000). Similar to the present study, Néeman et al. (1970) verified antimicrobial activity of long-chain aliphatic compounds isolated from kernels of *P. americana* for 13 species of bacteria and yeasts.

Some results available in the literature demonstrate the potential of using derivatives of *P. americana* in the control of other pests of agricultural importance, especially chewing and sucking insects, corroborating our results. Stein and Klingauf (1990) found a promising insecticidal activity of ethanolic extract of leaves of *P. americana* in control of *Myzus persicae* (Sulzer) (Hemiptera: Aphididae) and *Plutella xylostella* (Linnaeus) (Lepidoptera: Plutellidae). Santa-Cecilia et al. (2010) found that the aqueous extract of fruit husk (epicarp) of *P. americana*, at concentration of 250 mg mL<sup>-1</sup>, causes mortality to more than 77% of cochineal nymphs of *Planococcus citri* (Risso) (Hemiptera: Pseudococcidae).

For the control of hematophagous vector mosquitoes, Leite et al. (2009) found toxicity of hexane (LC<sub>50</sub>: 8.87 mg mL<sup>-1</sup>) and methanolic (LC<sub>50</sub>: 16.7 mg mL<sup>-1</sup>) extracts of avocado kernels against larvae of *Aedes aegypti* (Linnaeus) (Diptera: Culicidae). After fractionation, the authors found the presence of  $\beta$ -sitosterol and 1,2,4-trihydroxynonadecane compounds in the hexane extract of kernels of *P. americana*, which may be related to their larvicidal activity. Despite the promising biological activity, further studies are necessary to determine the possible modes and mechanisms of action of isolated compounds of *P. americana* on insects. According to Rodríguez-Saona and Trumble (2000), acetogenin persin affects insects possibly due to its interference in the biosynthesis of lipids, acting as a mimic of monoglycerides of linoleic acid.

Thus, according to the results obtained in laboratory conditions, kernels of *P. americana* could be used as raw material for the production of bioinsecticide for the control of *B. tabaci* biotype B. Therefore, bioguided fractioning should be performed to isolate and characterize compounds responsible for bioactivity. Moreover, tests in semi-field and field conditions are needed to assess the effectiveness of derivatives and compounds isolated from kernels of *P. americana* for the control of this pest. Despite the promising results, further studies are needed to both evaluate the toxicological action of

these derivatives on non-target organisms, mainly for human health, and to assess their biodegradability under environmental conditions, which are important aspects to be known before the formulation of pesticides based on extracts or isolated compounds of kernels of *P. americana* and their safe recommendation. Furthermore, studies on the structure-activity relationship may also facilitate biotransformation assays aimed at the enhancement and/or expansion of the range of activities of these compounds and the minimization of any potential inconveniences.

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**Availability of data and materials** The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

**Authors' contributions** LPR, SSC, and JDV planned and designed research; SSC and LPR conducted experiments; MRF, MFGFS, KUB, and KBF conducted chemical analysis; LPR, SSC, JDV, and JBF conducted statistical analysis and wrote the manuscript. All authors read and approved the manuscript.

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## Compliance with ethical standards

**Conflict of interest** The authors declare that they have no conflict of interest.

**Ethical approval and consent to participate** Not applicable.

**Consent for publication** Not applicable.

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