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Adulticidal Activity of Coffee Essential Oils against Two Fruit Fly Species (Tephritidae) under Laboratory Conditions

Adalton Raga[1*], Sara Braga e Silva[1], Ester Marques de Sousa[1] and Nilson Borlina Maia[2]

[1]Instituto Biológico, Alameda dos Vidoeiros 1097, 13101-680, Campinas, SP, Brazil.
[2]Instituto Agronômico de Campinas, Avenida Theodureto de Almeida Camargo 1500, 13075-630, Campinas, SP, Brazil.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

The aim of this study was to evaluate the adulticidal effects of green coffee oil (GCO) and roasted coffee oil (RCO) on two species of fruit flies (Tephritidae) include *Ceratitis capitata* (Wiedemann) and *Anastrepha fraterculus* (Wiedemann). Adult flies of both species were exposed to GCO and RCO at 2%, 3%, and 4% dilutions in water under Potter spray tower. In general, exposure to GCO and RCO at the three dilutions resulted in higher numbers of dead adults of both species compared to the control populations. The mortalities increased substantially between 60–90 minutes after initial exposure. The 2% GCO dilution reached a maximum of 77.0% corrected mortality of *A. fraterculus* at 360 minutes*.* In general, the toxic effect of coffee oils occurred more rapidly in *C. capitata* than in *A. fraterculus.* GCO (2% and 3%) and RCO (2%) showed promising results for fruit fly management.

Keywords: Coffea arabica; insecta; Anastrepha fraterculus; Ceratitis capitata.

**Corresponding author: E-mail: adalton.raga@sp.gov.br;*

1. INTRODUCTION

Fruit flies (Diptera: Tephritidae) are among the most important insect pests on earth. The female fruit flies oviposit inside fruit and their larvae feed upon the pulp, which causes pre- and post-harvest losses and may cause quarantine restrictions from many importing countries [1,2]. *Ceratitis capitata* (Wiedemann) and *Anastrepha fraterculus* (Wiedemann) were considered the most economically important fruit fly species in Brazil due to their large distribution, dominance, and number of commercial hosts [3].

In Brazilian orchards, damage caused by fruit flies is usually managed through the application of synthetic insecticides in which the majority of active ingredients are derived from organophosphates, pyrethroids and neonicotinoids [4]. Risks associated with the use of synthetic insecticides have led to the growth of an environmental movement seeking sustainable alternatives in pest control [5].

Traditional aromatic plants and their derivatives are an integral source of pesticides [6], especially as insect repellents [7]. In general, essential oils are considered to be low-risk products and their constituents exert insecticidal effects or reduce and disrupt insect growth at several life stages; efficacy varies according to both the chemical constituents of the plant extract and the target insect [5].

Essential oils are characterized by the presence of two or more major compounds with few trace compounds [6] and mainly consist of monoterpenes, sesquiterpenes and phenols [7,8]. The main effects of essential oils for fruit fly management are ovipositing-deterrent [9,10,11], attraction [12,13], repellent [14], anti-feeding [15], and insecticidal effects [16,17,18,19,20,21, 22,23].

Effective pesticidal plants would be complementary to naturally bred resistance, but more research is required to evaluate a wider range of plants and to optimise the use of the material [24] and its sub-products. The majority of plant secondary compounds likely evolved as defensive agents against insects, other herbivores, and plant pathogens [25].

Approximately 300 volatile compounds have been identified in green coffee beans and 200 are found in roasted coffee beans and are responsible for the sensory attributes of coffee [26,27]. The potential application of pyrolysis bio-oil from excess organic products, such as bio-oil from dried coffee grounds, as a pesticide is a new field of study [28]. The coffee oil (unsaponifiable matter) consists of various compounds in free form such as diterpenes, alcohols, cafestol, and caveol [29], and those substances are responsible for the important biological effects of coffee oil as a fungicide [30] and an insecticide. Caffeine showed high efficacy as an insecticide against coffee berry borer *Hypothenemus hampei* [31] and demonstrated a larvicidal effect on *Aedes aegypti* [32].

Frugivorous Tephritidae females lay their eggs inside fruit. The larvae feed on the pulp and eventually emerge from the fruit and bury themselves in the soil, where the pupa phase passes and the adult flies emerge. Consequently, adults are easier targets for controlling the fruit fly population.

Recently, green or roasted coffee oils have been commercially exploited and the biological activities of their components have been demonstrated [33]. The purpose of the current study was to evaluate the adulticidal effect of both types of coffee oils against *C. capitata* and *A. fraterculus* in a laboratory setting.

2. MATERIALS AND METHODS

2.1 Extraction of Essential Oils

Unlike essential oils which are normally extracted by steam distillation, the coffee oil used in this work was industrially extracted from coffee beans (*Coffea arabica* L.) using a cold-expeller press to extract the green (crude) coffee oil (GCO) and roasted coffee oil (RCO) without the use of solvents. The coffee oils tested in the present study were provided by Linax Indústria e Comércio de Óleos Essenciais Ltda. (Votuporanga, SP, Brazil). The product obtained by this press machine is a vegetable oil composed of triglycerides, fat acids, and a high content of unsaponifiable matter, reaching up to 12% [34].

2.2 Insect Colonies

Adults of *C. capitata* (also referred to as Mediterranean fruit-fly, or medfly) and *A. fraterculus* were obtained from colonies established in 1993 at the Instituto Biológico in Campinas, State of São Paulo, Brazil. Rearing methodology for both species were earlier described [35,36].

2.3 Adulticidal Evaluation

Five females and five males from 10-day-old *C. capitata* and 15-day-old *A. fraterculus* (one replication) populations were exposed to different aqueous dilutions of coffee essential oils under Potter tower using the bioassay methodology previously described [37]. Each coffee oil was diluted in distilled water at 2.0%, 3.0% and 4.0% (v/v). Control plots were sprayed in the same way using water only. The pH of the solutions was determined using an Alphalab pH meter (model PA 200).

The cumulative mortalities were evaluated at 15, 30, 45, 60, 90, 120, 150, 180, 240 and 360 minutes as well as 24 h after initial exposure.

2.4 Statistical Analysis

Ten replicates were used for each treatment. ANOVA (The SAS System for Windows, version 9.2) was performed using ranked data [38]. Three-factor ANOVA was used to compare the mortality of fruit flies: type of oil, time after exposure, species of fruit flies and sex.

3. RESULTS AND DISCUSSION

During the spraying procedure, the pH values of GCO 2.0%, 3.0%, 4.0% and RCO 2.0%, 3.0%, 4% and water (untreated control) were 8.50, 8.40, 8.31, 7.89, 7.45, 7.04 and 8.23, respectively. The number of dead adults differed significantly among the types of coffee oil and their concentrations ($F = 6,252$; $df = 27.34$; $P < 0.001$), time after exposure ($F = 3,756$; $df = 361,36$; $P < 0.001$) and between fruit fly species ($F = 1,252$; $df =14.07$; $P < 0.001$). There were no significant differences between sex ($F = 1,252$; $df = 1.12$; $P = 0.292$), but species and sex had a significant interactive effect on mortality ($F = 1,252$; $df = 4.80$; $P = 0.030$).

In general, adults of both species exposed to GCO and RCO in their respective dilutions resulted in higher numbers of dead insects compared with the control populations (Table 1). The number of dead insects from both coffee oils increased as the exposure interval increased (Fig. 1). Only GCO 2% showed a significantly higher number of dead adults compared with the control plots in four times of exposure and in both fruit fly species (Table 1). The lethal toxic effect for *C. capitata* and *A. fraterculus* increased substantially between 60–90 minutes of exposure (Fig. 1).

The fruit fly species as well as the type of coffee oil and respective concentrations showed interactions with significant influence on the specific lethality rate. A non-linear relationship between coffee oil concentration and dead insects (Table 1 and Fig.1) was observed; the survivorship increased with an increase in coffee oil concentration. The hormesis phenomenon appeared to occur with both coffee oils [39]. Occasionally, the insecticide concentration in the tissue initially increased and then decreased with increasing exposure concentration, due to the mechanisms used to eliminate the pesticide.

Exogenous substances can provoke alterations at the membrane level and affect physiological homeostasis [40]. Several monoterpenes contained in EOs are neurotoxic to insects [5]. Increases in the concentrations of essential oils can produce different toxic responses in the same insect species.

At up to 180 minutes, the number of dead *C. capitata* adults under RCO 4% exposure was similar to the control plots. At 360 minutes, all coffee oils and respective concentrations significantly increased the number of dead adults, with GCO 2%, GCO 3% and RCO 2% exhibiting similar numbers of dead insects in each fly species. GCO 2% reached a maximum of 77.0% corrected mortality of *A. fraterculus* at 360 min. At this interval of exposure, each coffee oil at 2% exhibited a similar number of dead adults between fruit fly species (Table 1), where *C. capitata* males were more susceptible than females (Fig. 1).

Medflies exposed to 3% dilutions of both coffee oils were more susceptible than *A. fraterculus.* In general, the toxic effect of coffee oil exposure occurred more rapidly in *C. capitata* than in *A. fraterculus* (Fig. 1). Medflies exposed to 3% dilution of both coffee oils presented higher mortality than *A. fraterculus*. RCO 4% reached intermediate levels of toxic effect in both fly species.

Regardless of the oil tested, no differences in susceptibility were detected between fly species by sex separately, except for *C. capitata* males at 60 and 90 minutes, which exhibited higher susceptibility than *A. fraterculus* males. In general, the toxic effect of both coffee oils was highest at the beginning of the exposure period. Medfly females were more resistant than males exposed to GCO 2% (Fig. 1). *C. capitata* females were more resistant than males exposed to limonene, linalool and α-pinene [41].

Table 1. Cumulative number of dead adults per plot (n = 10) of two species of fruit flies (Tephritidae) exposed to different concentrations of two coffee oils in the laboratory

Treatment	Time after initial exposure									
	60 min		**90 min**		**180 min**		**360 min**		**24 h**	
	Ceratitis capitata	***Anastrepha fraterculus***	***Ceratitis capitata***	***Anastrepha fraterculus***	***Ceratitis capitata***	***Anastrepha fraterculus***	***Ceratitis capitata***	***Anastrepha fraterculus***	***Ceratitis capitata***	***Anastrepha fraterculus***
GCO 2%	5.90 ± 0.62aA	2.90 ± 0.35aB	5.90 ± 0.62aA	7.10 ± 0.85aA	6.90 ± 0.67aA	7.40 ± 0.92aA	7.70 ± 0.65aA	7.90 ± 0.78aA	7.80 ± 0.55aA	8.30 ± 0.72aA
GCO 3%	4.70 ± 0.50aA	1.20 ± 0.29bB	4.80 ± 0.49aA	1.50 ± 0.37bcB	5.80 ± 0.70aA	3.50 ± 0.67bB	7.70 ± 0.65abcA	4.10 ± 0.66abcB	8.00 ± 0.65aA	5.30 ± 0.72abB
GCO 4%	2.00 ± 0.58cbA	0.80 ± 0.20bcA	2.00 ± 0.58bcdA	1.00 ± 0.39bcA	3.30 ± 0.50abA	2.10 ± 0.67bcA	4.10 ± 0.81bcA	3.20 ± 0.49cA	6.20 ± 0.53abA	4.40 ± 0.54bB
RCO 2%	3.00 ± 0.76abcA	1.90 ± 0.35abA	3.00 ± 0.76abcA	2.00 ± 0.37bA	6.20 ± 0.94aA	4.10 ± 0.77abA	6.90 ± 1.00abA	6.30 ± 1.00abA	8.10 ± 0.64aA	6.50 ± 0.99abA
RCO 3%	3.90 ± 0.96abA	1.20 ± 0.47bcB	4.00 ± 0.94abA	1.30 ± 0.54bcB	5.00 ± 0.83aA	2.30 ± 0.63bcB	6.00 ± 0.88abcA	3.00 ± 0.71bcB	7.20 ± 0.55aA	4.00 ± 0.61bB
RCO 4%	0.70 ± 0.30cdA	1.40 ± 0.34bA	0.70 ± 0.30cdB	2.00 ± 0.47bA	2.10 ± 0.57bcB	3.40 ± 0.75bA	3.40 ± 0.90cA	5.30 ± 0.79bcA	4.70 ± 0.80bA	6.00 ± 0.73abA
Control	0.30 ± 0.15dA	0.00 ± 0.00cA	0.30 ± 0.15dA	0.00 ± 0.00cA	0.50 ± 0.17cA	0.10 ± 0.10cA	0.50 ± 0.22dA	0.20 ± 0.13dA	1.40 ± 0.37cA	1.10 ± 0.31cA

Mean numbers (±SE) in the same column followed by the same lower case are not significantly different (Tukey´s test; P < 0.05). Mean numbers (±SE) within rows of respective transect followed by the same upper case are not significantly different (Tukey´s test; P < 0.05)
GCO = green coffee oil; RCO = roasted coffee oil

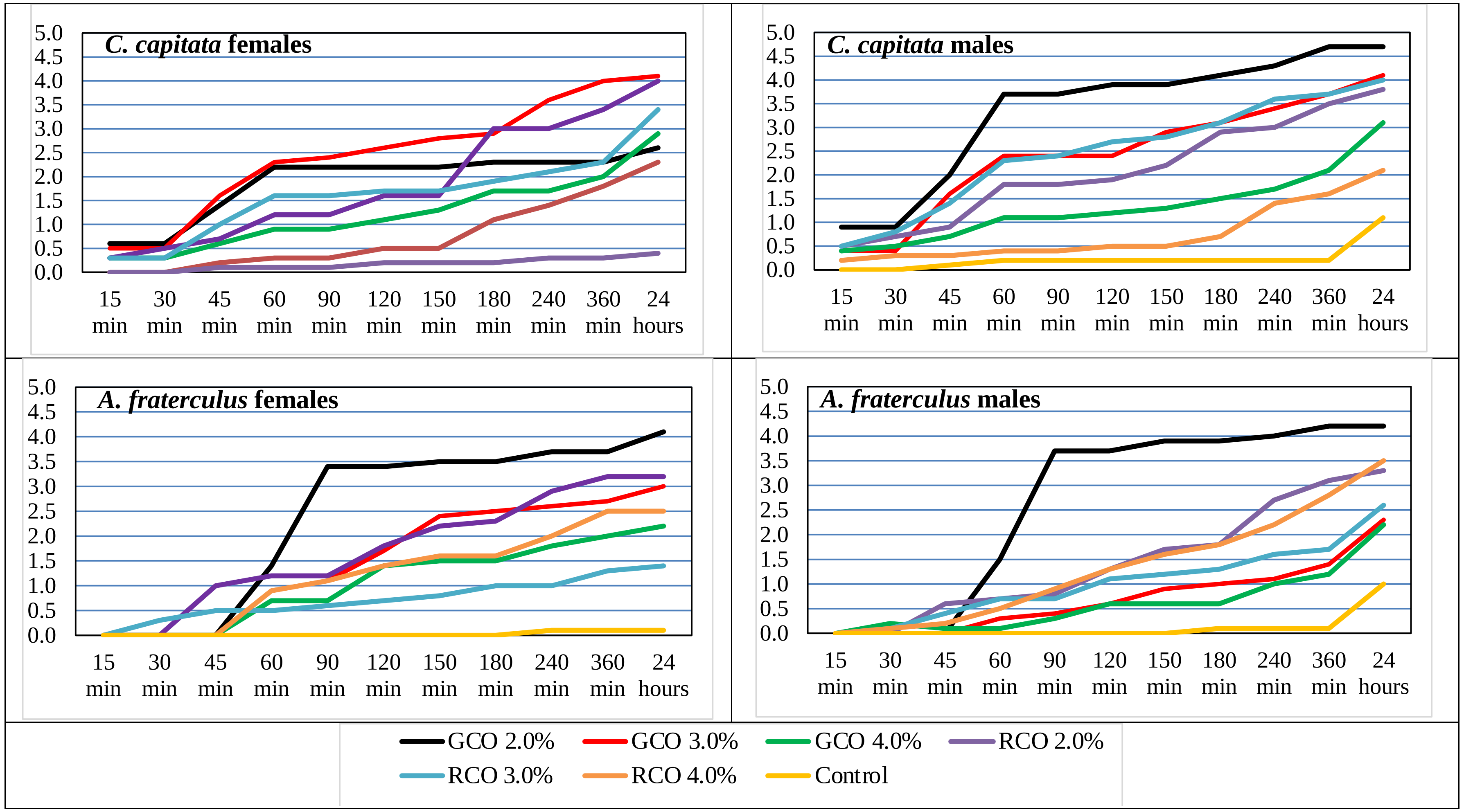

Fig. 1. Cumulative number of dead *C. capitata* and *A. fraterculus* adults after exposure to different concentrations of green coffee oil (GCO) and roasted coffee oil (RCO) in the laboratory

The unsaponifiable fraction of GCO consists of sterols, tocopherols, phosphatides, diterpenes, ceramides and other minor components. Palmitic, linoleic, oleic, stearic and arachidic acids are found in Brazilian roasted coffee oil [33,42], which has been shown to have insecticidal activity [43,44,45].

Essential oils from aromatic and medicinal plants are among the substances considered to be natural agents for insect management [46]. Essential oils have multiple modes of action due to the presence of several active components [47] and could be employed together with synthetic insecticides to create a synergistic effect [48]. In many cases, EOs are responsible for larval resistance in fruit pests.

Along with other phenolic compounds such as tannic acid, chlorogenic acid, and caffeic acid [32], caffeine is a chemical with natural pesticidal properties found in coffee grounds [28], which showed larvicidal action against *Aedes aegypti* (Linnaeus) (Diptera: Culicidae) [49]. Probably, caffeine and other compounds found in green and roasted coffee beans may have an adulticidal action, acting in combination or not.

Insecticide assays indicated that pyrolysis bio-oil from coffee grounds had to be diluted in water at a minimum of 10% to exhibit any acute toxicity in less than 48 hours against the Colorado potato beetle *Leptinotarsa decemlineata* Say [28].

Few studies have explored treatment with botanical extracts to control Diptera species of agricultural importance. Aerial part extractions of *Melaleuca alternifolia* (Maiden & Betche) Cheel (Myrtaceae) exhibited adulticidal effects against *C. capitata* [17]. Males of *Drosophila suzukii* (Matsumura) were more susceptible to essential oils of *Eucalyptus* spp. and *Melaleuca* spp. leaves and their components than females [50]. *Trans*-anethole and estragole, which are constituents from basil (*Ocimum basilicum* L.) essential oil, applied at 2.5% or higher caused > 77% mortality in *C. capitata*, *Bactrocera dorsalis* (Hendel) and *Bactrocera cucurbitae* (Coquillett) after 2 hours of exposure [51]. Peel extracts of lemons and grapefruits are toxic to larvae of *A. fraterculus* [52].

The use of essential oils to control fruit flies could improve the sustainable management of pests in horticultural crops. Further research is needed to evaluate the efficacy of coffee oil on fruit flies in the field and to determine the residual period and the bioactive compounds necessary to facilitate the formulation. Additionally, further study of the efficacy of coffee oil via toxic bait (ingestion) and the possibility of its mixture in the application of conventional insecticides is needed.

CONCLUSIONS

Green coffee oil (GCO) and roasted coffee oil (RCO) diluted in water at 2.0%, 3.0% and 4.0% exhibited insecticidal effects against *C. capitata* and *A. fraterculus* adults in a laboratory setting. A substantial increase in the adulticidal effect was observed between 60–90 minutes after initial exposure. Both fruit fly species showed differences in susceptibility to coffee oils and their respective concentrations over time. GCO (2% and 3%) and RCO (2%) were shown to be promising for management of the tested fly species.

4. DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

COMPETING INTERESTS

Authors have declared that no competing interests exist

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