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Composition and fumigant protectant potential of Tunisian *Citrus aurantium* L. essential oils against *Rhyzopertha dominica* F. (Coleoptera: Bostrichidae) †

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† Presented at the 1st International Electronic Conference on Entomology (IECE 2021), 1–15 July 2021; Available online: https://iece.sciforum.net/.

Citation: Djebbi, T.; Soltani, A.; Chargui, H.; Yangui, I.; Messaoud, C.; Jemâa J.M.B. ; Composition and fumigant protectant potential of Tunisian *Citrus aurantium* L. essential oils against *Rhyzopertha dominica* F. (Coleoptera: Bostrichidae), in Proceedings of the 1st International Electronic Conference on Entomology, 1–15 July 2021, MDPI: Basel, Switzerland, doi:10.3390/IECE-10643

Published: 09 July 2021

Abstract: The valorization of Citrus peels from agro-industries is an important issue since Tunisia is known by the production of such crops. This work aims to assess the insecticidal potential of essential oils extracted from the bitter orange *C. aurantium* as protectant against adults of the less grain borer *R. dominica*. Results revealed that Limonene was the major and characteristic compound. The fumigant bioassay showed that *C. aurantium* from Nabeul was more toxic with LC_{50} and LC_{95} values were 307.9 and 652.9 µL/L air, respectively within 48h of exposure.

Keywords: Citrus aurantium; fumigation; grains; Rhyzopertha dominica

1. Introduction

In many regions of the world, the lasser grain borer, *R. dominica*, is one of the major pests feeds in quality of grains as larva's and adults (lose weight and the market value) and they are become generally unacceptable for human consumption [1,2,3]. Besides, the use of biological insecticides increases production costs as well as leads to environmental and consumer health risks [4,5,6]. In fact, Essential oil extracted from genus *Citrus*, have been identified to have insecticidal properties several coleopteran species [7,8,9] whith a variation in the composition of secondary metabolites [10,11]. This research assesed the chemical composition and the fumigant toxicity of the essential oils of peels of Tunisian *Citrus auranthium* against the adults of *Rhyzopertha dominica*.

2. Material and methods

2.1. Insect breeding

Rearing of the adults of R. dominica was affected in the Laboratory of Biotechnology Applied to Agriculture in the breeding chambre at 30 °C, 60 RH and photoperiod cycle of L:D 12:12h.

2.2. Sampling

At mature stage of the trees, Fruits of *C. aurantium* were collected from Bir Merwah and acheichia (Nabeul and Boussalem, north Tunisia), respectively. In the laboratory, fruits were rinsed with distilled water and we are recovered the peels.

2.3. Extraction of the essential oil

the extraction of the peels essential oil of C. aurantium were affected by hydrodistillation with the using of Clevenger type apparatus for two hours at 100 °C.

2.4. chemical composition

The quantitative and qualitative analysis of the volatile compounds in the essential oils was determined by combination of gas chromatography (GC) and mass spectrometry (MS) according to a standard analytical procedure BY using a DB-5MS column (30 m, 0.25 mm and 0.25 μm film thickness). The initial oven temperature was held at 60°C for 4min. it was then increased by 3 °C/min to 250 °C; the injection port and ionizing source were kept at 250 and 280 °C, respectively[12].

2.5. Insecticidal activities (Fumigant toxicity)

For the determination of the fumigant toxicity of bitter orange essential, 3,5 cm in diametre of whatman N°1 filter paper were impregnated with oil at doses calculated to release fumigant bioessay (41.67, 83.33 and 166.67 μL/L air), into, these filter paper impregnated was attached to the screw cap of the 24 mL plexiglas bottle that contains ten adults and these caps were screwed on tightly. Than, the number of dead and alive insects in each bottle was counted after 48h of exposure for the determination of LC_{50} and LC_{95}..

2.6. Statistical analysis

analysis of variance of the data was affected by using Statistical Package for Social Sciences (SPSS) version 11.0 and LC values were estimated by using the Probit analysis.

3. Results and discussion

3.1. yields and chemical composition of the essential oils

Yields of *C. aurantium* essential oil were 0,555 % and 0,851 % in peels from Nabeul and Bousselem, respectively. Table 1 showed the percentages of the chemical compounds identified in the essential oil extracted from the peels of *C. aurantium* of two locations. in fact, GC/MS identified 99.99 % of the total essential oil. Monoterpenes were the main components in essential oil from peels of *C. aurantium* with 97.63 and 98.29 %, respectively from Bousselem and Nabeul. In fact, limonene represented the major composant of the peels essential oils od *C. aurantium* (93.69 and 92.36 %), β-myrcene (1.77 and 1.7 %), β-ocimene (0.72 and 0.77 %), α-pinene 0.58 and 0.49%), β-pinene (0.29 and 0.93 %), sabinene (0.16 and 0.22%), β-fenchol (0.12 and 0.32%) and nerol (0 and 0.15 %) were the main components in essential oil of Bousselem and Nabeul, respectively and linalool (0.3 and 1.45 %) was the only oxygenated monoterpenes in peel essential oil of Bousselem and Nabeul, respectively. Furthermore, sesquiterpene hydrocarbons were represented by germacrene D (0.09 %) in only the essential oil of Nabeul. the fraction of the non terpenic compounds was essentialy represented by aldehydes (0.06%) and esters (0.69%). Zarrad et al. [12] reported that *C. aurantium* peel oils from Chott Mariem (Sousse, Tunisia) was caracterised essentially by limonene with a percentage of 87.523 %. Additionally, Moraes et al. [13] reported that peels essential oils from Brazilan C. aurantium consist mainly for Limonene and myrcene with a percentage of 98% and 1.45%. These differences in the compositions of essential oils may be depended to different factor, for exemple, seasonal, maturational stage and pedoclimatic variation [14,15], also, the extraction method and the age of the trees can influence the quantity and the quality of the essential oils [16,17]. Thid study shows also the sensitivity of adults of *Rhyzopertha dominica* to the peels essential oils of *C. aurantium*.

Table 1. Chemical composition of peels essential oils from *C. aurantium* .

Compounds	Percentages (%)	
	***C. aurantium* (Bousselem)**	***C. aurantium* (Nabeul)**
Monoterpenes	97.63	98.39
Monoterpene hydrocarbons	97.33	96.94
Oxygenated monoterpenes	0.3	1.45
Sesquiterpene hydrocarbons	_	0.09
Non terpenic compounds	2.27	1.51
Aldehydes	0.06	_
Esters	_	0.69
Others	2.21	0.82
Total	99.9	99.9

3.2. Insecticidal activities (Fumigant toxicity)

Results showed that *Citrus aurantium* peel oils were toxic to the adults of *Rhyzopertha dominica* and the percentage of mortality was related to the doses of the essential oils. Analysis of results showed also that *C. aurantium* from Bousselem was less toxic to *R. dominica* than essential oil from Nabeul. The corresponding LC_{50} and LC_{95} of Nabeul and Bousselem were respectively 307.89 and 440.55 µL/L air and 652.91 and 1283 µL/L air.

Furthermore, our results does not agree with the results of Souza et al. [18], hwo are shown that the esssential oils of bitter orange has an effect in the killing of adults of *R. dominica* with an LC_{50} and LC_{100} values equal to 69.36 µL/L air and 130.68 µL/L air, respectively. the insecticidal activities of *C. aurantium* essential oils have previously been evaluated against several insect species [19]. Moreover, Pala and Pathipati [20] reported that essential oils of *C. aurantium* were highly effective with 76% mortality of *R. dominica* at 8.5 mg/cm². Indeed, The toxicity of these oils could be explained by its richness in monoterpenoids compounds that possess insecticidal activity of the limonene against several insect species [21] and the toxic effective of the limonene are mediated by penetrating the insect bodyy through the resoiratory systeme [7]. Tripathi et al. [22] reported the insecticidal activities of limonene against *R. dominica* and other insects, for exemple, *Sytophylus oryzae*, *Tribolium castaneum* [18].

Table 2. LC_{50} and LC_{95} value of *C. aurantium* peel essential oils from Bousselem and Nabeul against *R. dominica* adults.

	***C. aurantium* (Bousselem)**	***C. aurantium* (Nabeul)**
LC_{50} (µL/L air)	440.55	307.89
LC_{95} (µL/L air)	1283	652,91
Slope±SEM	0.002±0.002	0,005±0.002
χ^2	1.36	3.28
Degree of freedom	1	1

Units LC_{50} and LC_{95} = µL/Lair, applied for 48 h at 25 ◦C.

4. Conclusion

This study, proved the fumigant toxisity of two North Tunisian *C. aurantium* peel oils into adults of *R. dominica*. Indeed, these proceeding showed that *C. auranthium* essential oils can be a source for eco-friendly alternatives in whitefly integrated control program.

Author Contributions:"Conceptualization, Tasnim Djebbi and Jouda Mediouni Ben Jemâa ; methodology,Tasnim Djebbi; software, Tasnim Djebbi; validation, Tasnim Djebbi, Abir Soltani and Jouda Mediouni Ben Jemâa ; formal analysis, Tasnim Djebbi; investigation, Tasnim Djebbi; resources, Tasnim Djebbi; data curation, Tasnim Djebbi; writing-original draft preparation, Tasnim Djebbi; writing-review and editing, Tasnim Djebbi; visualization, Tasnim Djebbi; supervision, Jouda Mediouni Ben Jemâa; project administration, , Jouda Mediouni Ben Jemâa; funding acquisition, Jouda Mediouni Ben Jemâa. All authors have read and agreed to the published version of the manuscript.

Funding: please add: "This research received no external funding".

Informed Consent Statement: "Informed consent was obtained from all subjects involved in the study."

Conflicts of Interest: "The authors declare no conflict of interest".

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