Journal of Kerbala for Agricultural Sciences Issue (1), Volume (9), (2022)

Study the effectiveness of marijuana and rosemary alchoholic extracts in control of red rusty flour beetle *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae)

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Received:
Jan. 27, 2022

Accepted:
Feb. 28, 2022

Published:
Mar. 10, 2022

Abstract

Laboratory experiments were conducted to determine the effectiveness of plant extracts of vitex and rosemary in red rust flour beetle *Tribolium castaneum* (Herbst) control. Results showed a significant effect of extracts oncentrations in the percentage of larval stage mortality . concentration of 5%, caused the highest mortality rates in the larval stage reached to 71.11, 76.66, 83.33 % insects / plate after 24, 48 and 72 hours, respectively. The mortality rates were equal for both concentrations 2.5 and 5%, which amounted to 90 % after 72 hours. Vitex plant extarct was significantly superior to the rosemary plant *Rosmarinus officinalis* in the mortality rates for all concentrations and periods post application in this study.

Keywords: Marijuana, Rosemary, *Tribolium castaneum*

Introduction

Insects are a serious problem in grain stores all over the world, and it statistically is causing a loss in the quantity and quality of grains up to 75%, spreads all over the world and is very destructive [1]. This insect cannot infect healthy grains, but it attacks broken grains and grain products such as flour, bran, and the materials made from them. As for the larvae, they feed on the internal content of the grains that is previously affected by primary insects [2]. In the case of severe infestation of flour, its color turns to gray due to contamination with integuments, insects, excrements and dead individuals. The taste changes and acquires a distinctive unpleasant odor as a result of insect's smell glands secretions [3]. Because of the damage caused by this insect on food stock, it is very necessary to control this pest with high caution to human health. Methods that can be safe and effective is the use of plant derivatives such as palm tree *Vitex agnus* - castus L. It is a deciduous tree that can grow to about 6.7 meters, grows along with rivers in southern Europe, Mediterranean countries, central Asia and the Middle East [4]. The plant contains several active components, the most important of which are flavonoids, glycosides, terpenes, phenols, carbohydrates, saponins, and volatile oil approximately 1.5%. The plant also contains a small percentage of essential oils in about 0.5%, the most important of which is limonene oil [5]. Likewise, the rosemary plant Rosmarinus officinalis is one of the aromatic medicinal plants, evergreen herbaceous with small flowers, indigo or blue color. Its aroma has been preferred since antiquity Natural herbs such as rosemary and other types of plants contain antioxidants, rosemary

is widely used as a food additive, having protective traits for human body This due to its antioxidant activity as they contain a large amount of phenolic compounds, flavonoids and natural acids [6]. The use of chemicals against insects has become ineffective due to the development of resistance in different strains [7]. Many researchers indicated that plant-derived materials do not cause insect resistance, with a broad-spectrum activity, and are safe for biological enemies. This made such plant compounds to be suitable for IPM as biological control agents [8]. The study aimed, there for, to evaluate the effect of different concentrations of alcoholic extract of Vitex agnus - castus L. and rosemary or Rosmarinus officinalis on the biological performance of the rusty red flour beetle.

Materials and methods

Insect collection, raring and identification

Adults of the *T. castaneum* insect were collected from infected flour stored in flour stores in Karbala. The insect was diagnosed by the professors of the Department of Protection / College of Agriculture / University of Karbala, using the classification keys of the family Tenebrionidae [1]. For the purpose of perpetuating the colony of the insect, 250 gm of bran was placed inside a sterilized glass bottle of 8 cm dia. and 15 cm height, and then 50 pairs of insect adults aged between 48-24 hours were released into each Bottles were covered with a muslin cloth, sealed with elastic band to prevent insects from escaping, incubated at 2+28°C and a relative humidity of 5+70%. The feed material (bran) was renewed every two months to obtain young insects for subsequent tests.

Processing, identification and storing plant samples

The Vitex and rosemary plant parts were obtained from the local markets in Karbala governorate on 2020. The plant parts were ground by a large electric mill till became a fine powder, placed in sterile and labeled cloth bags and kept in a cool place until use.

Table (1): The plants used in the study

	The scientific name	Family	English name	local name	used part
1	*Vitex agnus – castus L.*	Lamiaceae	Chaste tree	Mary's palm	leaves and steams
2	*Rosmarinus officinalis*	Lamiaceae	Rosemary	Rosemary	leaves and flowers

Preparation of alcoholic extracts

Absolute ethyl alcohol was selected in the extraction process according to the modified [9] method. 10 g of plant powder was taken per 200 ml of alcohol and placed in an electric shaker for 2 hours. Then the sample was dried in an electric oven at a

temperature of 50-45°C. The process was repeated several times to obtain a sufficient amount of the raw material that were kept in the refrigerator until use.
For the purpose of estimating the efficacy of alcoholic extracts of vitex and rosemary plants on the red rust flour beetle, 5 gm of dry residue was taken from each extract separately and dissolved in 5 ml of ethyl alcohol and supplemented the volume to 100 ml of distilled water to obtain a stock solution of 5%. Concentrations (2.5 and 1.25%) were prepared for the treatments, while the control treatment was using 5 ml of solvent and 95 ml of distilled water.

Effect of vitex and rosemary plant extract on mortality rates of T. castaneum larvae and adults

Ten larvae/replicate of *T. castaneum* were taken with three replicates for each concentration (1.25%, 2.5%, 5%) of prepared plant extract. The larvae were placed in a disposable petri dish, treated directly (topical spray), using a small sprayer with a capacity of 5 ml and sprayed from a height of approximately 25 cm. The dishes were incubated at 28± 5C° and a relative humidity of 70±5%. The mortality rates were recorded after 24, 48 and 72 hours of treatment, and the results were corrected according to Abbott's equation [10].

Experiment design and statistical analysis

The experiment parameters were distributed using the Completely Randomized Design (CRD). The data were analyzed using the SAS statistical analysis program. The least significant difference (L-S-D) test was used at the probability level (0.05) for differences between the treatments [11]. The death percentages were corrected according to the Abbott Formula [12] Corrected death percentages were calculated according to the following equation:

$$\text{Abbott corrected mortality} = \frac{\text{\% mortality in treatment} - \text{\% mortality in control}}{100 - \text{\% mortality in control}} \times 100$$

The corrected death percentages were converted to data log values that are not included in the statistical analysis.

Results and Discussion

Effect of vitex and rosemary plant extract on mortality rates of *T. castaneum* larvae 24h post treatment

The results (Table 2) showed that there was a significant effect of the plant extract type *Vitex agnus - castus* L. and Rosmarinus officinalis and the different concentrations 1.25, 2.5 and 5% and their interaction on mortality percentage of the flour beetle

Tribolium castaneum larvae. The results showed that Vitex extract led to the highest larvae mortality rate after all periods under study (24, 48 and 72 h post treatment), while the lowest mortality rate resulted from rosemary extract treatments. The results showed a significant effect of the concentrations of the extract used compared to the control treatment. The highest larvae mortality rate was at the concentration of 5% mg/ml, followed by the concentration 2.5% and 1.25% which resulted in the lowest mortality rate. The highest mortality rate after 24 hours was 86.66% in the treatment of vitex at the highest concentration (5%) compared to 43.33% mortality rates in the treatment of rosemary at the concentration 2.5%. Similarly, the highest mortality rate after 48 hours of application was in the treatment of vitex with the highest concentration, which was 90% compared to the lowest mortality rate of 45% in the treatment of rosemary at the same concentration. In general, the mortality percentage increased significantly after 72 hours of application in the case of treatment with rosemary and for all concentrations, while the mortality rate after 72 hours did not sign a significant effect in vitex extract from what resulted after 48 hours post treatment.

Table (2): Effect of plant extracts at different concentrations and period post application on larvae mortality rate (%) of the rusty flour beetle *T.castaneum*

Treatments		Mortality at hours post treatment HPT		
Plant extract	**Concentration (%)**	**24HPT**	**48HPT**	**72HPT**
Vitex agnus	**1.25%**	**43.33**	**53.33**	**63.33**
	2.5%	**83.33**	**86.66**	**90.00**
	5%	**86.66**	**90.00**	**90.00**
Rosmarinus officinalis	**1.25%**	**60.00**	**73.33**	**90.00**
	2.5%	**43.33**	**54.66**	**70.33**
	5%	**45.66**	**45.66**	**76.66**
L.S.D.$_{(P\leq 0.05)}$		**11.86**	**13.907**	**13.425**
Treatments		**Mortality at hours post treatment HPT**		
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Rosmarinus officinalis	**1.25%**	**60.00**	**73.33**	**90.00**
	2.5%	**43.33**	**54.66**	**70.33**
	5%	**45.66**	**45.66**	**76.66**
L.S.D.$_{(P\leq 0.05)}$		**11.86**	**13.907**	**13.425**

The study showed that *Vitex agnus - castus* L. leaf powder extract had an effect on the percentage of death and led to a decrease in the population of the first generation F1, as well as its repellent effect on the red rusty flour beetle *Tribolium castaneum* [13]. [14] reported that the use of ML/L34 of palm oil led to the death of 96.6% of red flour beetle adults within 24 hours of treatment. [15] found that the LC_{50} of rosemary oil extract in fumigation against *Callosobruchus maculatus* (F.) was 15.69ML/L.

The variation in the percentages of mortality may be due to the toxic effect through contact of the extract with the surface of the body and the penetration of the chemical compounds of the cuticle through penetration into the flexible areas in it or through the respiratory openings, causing paralysis and rapid death. Glycosides are active compounds that act as feeding or repellent inhibitors that lead to inhibition of the egg-laying process, hatchability, the process of molting of larvae and the death of adults [16]. These results were in line with what was mentioned by [17] who found that the use of plant extracts to control the southern cowpea beetle, including rue and garlic, led to an increase in the death rate of wholes with an increase in exposure time and a high concentration of the extracts used, and the death rate reached 60% for rue and 92% for garlic at the highest concentration (1800 parts per million) and this effect decreased in the lowest concentrations until it reached 16% for solute and 18% as a maximum for garlic at the lowest concentration of 5.112 ppm.

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