

SUMMARY

The objective of this study was to evaluate the efficacy of some plant oils (Mustard oil, Basil oil, Chamomile oil, Anise oil, Orange oil and Rosemary oil) against adult of *T. castaneum* and *S. oryzae* at $30\pm1^{\circ}\text{C}$ and $65 \pm 5 \%$ R.H. Meanwhile, the efficacy of some plant extracts (*Jatropha curcas*, *Peganum harmala*, *Callistemon citrinus*, *Melaleuca leucadendron*, *Rosmarinus officinalis* and *Punica granatum*) were evaluated against *T. castaneum* and *S. oryzae* at $30\pm1^{\circ}\text{C}$ and $65 \pm 5 \%$ R.H. Experiments were performed at the laboratory of the plant protection department of the Faculty of Agriculture, Moshtohor, Benha University.

1. Efficacy of some essential oils against the adult of *T. castaneum* and *S. oryzae*:

In this experiment, adult of *T. castaneum* and *S. oryzae* were exposed to different essential oils (Mustard oil, Basil oil, Chamomile oil, Anise oil, Orange oil and Rosemary oil) at different concentrations for 1, 2, 3, 5, 7, 14 and 21 days at 30°C temperatures. The results showed that mortality increased with increasing oil concentration and time of exposure. As following:

1.1. *T. castaneum*:

The results showed that mortality percentage was increased by increasing the plant oil concentration and period of exposure. After 21 days from the initial treatment, mortality was between 30-100% for Mustard oil, 13.33-100% for Basil oil, 30-100% for Chamomile oil, 16.67-100% Anise oil, 30-100% for Orange oil and 33.33-100% for Rosemary oil. The LC_{50} values were 3.05, 2.98, 2.05, 3.12, 1.88 and 2.53% for Mustard oil, Basil

oil, Chamomile oil, Anise oil, Orange oil and Rosemary oil, respectively after 7 days. Reduction in progeny was from 53.67-100, 49.80-100, 61.39-100, 35.91-100, 53.67-100 and 66.03-100% for Mustard oil, Basil oil, Chamomile oil, Anise oil, Orange oil and Rosemary oil, respectively.

1.2. *S. oryzae*:

The results showed that mortality percentage was increased by increasing the plant oil concentration and period of exposure. After 21 days from the initial treatment, mortality was between 28.89-100% for Mustard oil, 31.11-100% for Basil oil, 26.67-100% for Chamomile oil, 44.44-100% Anise oil, 23.33-100% for Orange oil and 17.78-100% for Rosemary oil. The LC₅₀ values were 0.53, 2.50, 0.70, 1.90, 0.72 and 2.84% for Mustard oil, Basil oil, Chamomile oil, Anise oil, Orange oil and Rosemary oil, respectively after 7 days. Reduction in progeny was from 64.60-100, 50.56-100, 46.62-100, 77.53-100, 66.85-100 and 43.82-93.25% for Mustard oil, Basil oil, Chamomile oil, Anise oil, Orange oil and Rosemary oil, respectively.

2. Efficacy of some plant extracts against the adult of *T. castaneum* and *S. oryzae*:

In this experiment, adult of *T. castaneum* and *S. oryzae* were exposed to different plant extracts (*J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*) by Petroleum ether, Aceton, Ethyl alcohol and Water serially at different concentrations for 1, 2, 3, 5, 7, 14 and 21 days at 30°C temperatures. The results showed that mortality increased with increasing oil concentration and time of exposure. As following:

2.1. Petroleum ether extracts from different plants:

2.1.1. *T. castaneum*:

The results showed that mortality percentage was increased by increasing the plant extract concentration and period of exposure. After 21 days from the initial treatment, mortality was between 38.89-100% for *J. curcas*, 28.89-100% for *P. harmala*, 42.22-100% for *C. citrinus*, 43.33-100% *M. leucadendron*, 68.89-100% for *R. officinalis* and 30-100% for *P. granatum*. The LC₅₀ values were 4.05, 6.55, 6.33, 3.83, 4.20 and 5.09% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively after 7 days. Reduction in progeny was from 45.56-100, 45.95-100, 75.67-100, 66.03-96.14, 66.03-98.08 and 66.03-98.84% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively.

2.1.2. *S. oryzae*:

The results showed that mortality percentage was increased by increasing the plant extract concentration and period of exposure. After 21 days from the initial treatment, mortality was between 28.89-100% for *J. curcas*, 31.11-100% for *P. harmala*, 67.78-100% for *C. citrinus*, 88.89-100% *M. leucadendron*, 63.33-100% for *R. officinalis* and 38.89-100% for *P. granatum*. The LC₅₀ values were 0.67, 0.86, 0.60, 0.53, 0.88 and 0.09% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively after 7 days. Reduction in progeny was from 85.96-100, 77.53-100, 71.91-100, 83.15-100, 91.50-100 and 80.77-100% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively.

2.2. Acetone extracts from different plants:

2.2.1. *T. castaneum*:

The results showed that mortality percentage was increased by increasing the plant extract concentration and period of exposure. After 21 days from the initial treatment, mortality was between 36.67-100% for *J. curcas*, 32.22-100% for *P. harmala*, 40-100% for *C. citrinus*, 33.33-100% *M. leucadendron*, 46.67-100% for *R. officinalis* and 51.11-100% for *P. granatum*. The LC₅₀ values were 4.10, 6.30, 7.35, 5.10, 2.44 and 6.61% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively after 7 days. Reduction in progeny was from 49.80-100, 34.36-100, 51.73-100, 53.67-100, 51.73-100 and 53.67-100% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively.

2.2.2. *S. oryzae*:

The results showed that mortality percentage was increased by increasing the plant extract concentration and period of exposure. After 21 days from the initial treatment, mortality was between 73.33-100% for *J. curcas*, 48.89-100% for *P. harmala*, 44.44-100% for *C. citrinus*, 73.33-100% *M. leucadendron*, 31.11-100% for *R. officinalis* and 51.11-100% for *P. granatum*. The LC₅₀ values were 0.58, 0.90, 0.97, 0.63, 1.69 and 0.64% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively after 7 days. Reduction in progeny was from 88.76-100, 71.91-100, 64.60-100, 83.15-100, 32.58-94.44 and 71.91-100% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively.

2.3. Ethyl alcohol extracts from different plants:

2.3.1. *T. castaneum*:

The results showed that mortality percentage was increased by increasing the plant extract concentration and period of exposure. After 21 days from the initial treatment, mortality was between 34.44-100% for *J. curcas*, 18.89-100% for *P. harmala*, 24.44-100% for *C. citrinus*, 18.89-100% *M. leucadendron*, 46.67-100% for *R. officinalis* and 21.11-100% for *P. granatum*. The LC₅₀ values were 3.53, 10.49, 5.37, 10.16, 7.06 and 10.85% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively after 7 days. Reduction in progeny was from 44.02-100, 15.06-75.67, 11.19-96.14, 11.19-75.67, 35.91-100 and 15.06-80.77% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively.

2.3.2. *S. oryzae*:

The results showed that mortality percentage was increased by increasing the plant extract concentration and period of exposure. After 21 days from the initial treatment, mortality was between 51.11-100% for *J. curcas*, 36.67-100% for *P. harmala*, 38.89-100% for *C. citrinus*, 31.11-100% *M. leucadendron*, 26.67-100% for *R. officinalis* and 47.78-100% for *P. granatum*. The LC₅₀ values were 1.58, 3.51, 3.64, 4.31, 5.17 and 4.86% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively after 7 days. Reduction in progeny was from 71.91-100, 46.62-100, 26.97-100, 26.97-100, 15.73-91.57 and 50.56-98.31% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively.

2.4. Water extracts from different plants:

2.4.1. *T. castaneum*:

The results showed that mortality percentage was increased by increasing the plant extract concentration and period of exposure. After 21 days from the initial treatment, mortality was between 32.22-100% for *J. curcas*, 35.56-100% for *P. harmala*, 23.33-100% for *C. citrinus*, 26.67-100% *M. leucadendron*, 38.89-100% for *R. officinalis* and 23.33-100% for *P. granatum*. The LC₅₀ values were 7.02, 5.39, 6.57, 6.55, 6.64 and 9.80% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively after 7 days. Reduction in progeny was from 44.02-100, 49.80-100, 22.77-94.21, 51.73-100, 22.77-96.14 and 34.36-92.28% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively.

2.4.2. *S. oryzae*:

The results showed that mortality percentage was increased by increasing the plant extract concentration and period of exposure. After 21 days from the initial treatment, mortality was between 28.89-100% for *J. curcas*, 30-100% for *P. harmala*, 21.11-100% for *C. citrinus*, 31.11-100% *M. leucadendron*, 26.67-100% for *R. officinalis* and 23.33-100% for *P. granatum*. The LC₅₀ values were 2.24, 3.61, 3.53, 2.63, 4.62 and 6.48% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively after 7 days. Reduction in progeny was from 32.58-100, 71.91-100, 32.58-100, 50.56-100, 10.11-100 and 21.33-98.87% for *J. curcas*, *P. harmala*, *C. citrinus*, *M. leucadendron*, *R. officinalis* and *P. granatum*, respectively.

3. Efficacy of Orange oil, Mustard oil, acetone extracts

from *R. officinalis* and petroleum ether extracts from *P. granatum* on germination of wheat grains:

Results indicated relative that relatively no significant differences were found in germination of wheat seeds treated with petroleum ether extracts from *P. granatum* and acetone extracts from *R. officinalis* but significant differences were found in germination of wheat seeds treated with Orange oil and Mustard oil.

4. Preliminary phytochemical screening:

The phytochemical investigation of *R. officinalis* illustrated that they was rich in carbohydrate, tannins and sterols. On the other hand, *P. granatum* was rich in carbohydrate, tannins, flavonoides and Sterols.

5. Differential hemocyte count:

The results of the effect of LC₅₀ of Orange oil on blood cells of larvae of *T. castaneum* showed mildeffect that appeared on cells membrane and empty cells was appeared.

The results of the effect of LC₅₀ of acetone extracts from *R. officinalis* on blood cells of larvae of *T. castaneum* showed strong influence on the blood cells appeared where empty cells, destructive cells and cells with clumped contents were appeared.

6. Histological studies:

The results of the effect of LC₅₀ of Orange oil on midgut of *T. castaneum* adult showed the epithelium cells are destroyed and separated completely from the broken basement membrane. The cellular debris from degenerating cells filled the gut lumen.

The results of the effect of LC₅₀ of acetone extracts from *R. officinalis* on midgut of *T. castaneum* adult showed the epithelial cell lost their close association with the basement membrane and with each other. The epithelial cells were destroyed and loss their columnar structure in some point and caused disorganization of peritrophic membrane. The basement membrane appear and still intact and the epithelium cells destruced in some point.

7. Physiological studies:

7.1. Effect of different treatments on the total proteins, lipids and carbohydrates of the treated adult of *T. castaneum*:

Total proteins, total carbohydrates and total lipids content were significantly decreased reduced to acetone extracts from *R. officinalis* and Orange oil treatment in compared to the control treatment.

7.2. Effect of different treatments on the total proteins, lipids and carbohydrates of the treated adult of *S. oryzae*:

Total proteins, total carbohydrates and total lipids content were significantly decreased reduced to petroleum ether extracts from *P. granatum* and Mustard oil treatment in compared to the control treatment.

7.3. Effect of different treatments on the enzymes activies in adult of *T. castaneum*:

No significant decrease in the activity of **(GOT)** for acetone extracts from *R. officinalis* and Orange oil in compared to control treatment. On the other hand, the obtained results of **(GPT)** indicated that no significant increase in the activity enzyme.

A Significant increase in activity of **Alpha esterases** in case of acetone extracts from *R. officinalis* and Orange oil treatments in compared to control treatment. No Significant decrease in activity of **Beta esterases** in case of acetone extracts from *R. officinalis* treatment in compared to control treatment. A Significant increase in activity of **Beta esterases** in case of Orange oil treatments in compared to control treatment.

A Significant increase in activity of **Acetylcholinesterase** in case of acetone extracts from *R. officinalis* in compared to control treatment, while no significant decreased under Orange oil treatment.

Significant decrease in the activity of **Invertase** enzyme under acetone extracts from *R. officinalis* and Orange oil in compared to control treatment.

A Significant increase in activity of **Trehalase** in case of acetone extracts from *R. officinalis*, while no significant decreased under Orange oil treatment in compared to control treatment.

A Significant increase in activity of **Amylase** in case of acetone extracts from *R. officinalis* and Orange oil in compared to control treatment.

7.4. Effect of different treatments on the enzymes activities in adult of *S. oryzae*:

Significant decrease in the activity of **(GOT)** for petroleum ether

extracts from *P. granatum*, while on the contrary significant increase of enzyme activity under Mustard oil treatment in compared to the control treatment. On the other hand, the obtained results of **(GPT)** indicated that significant decrease in the activity enzyme to petroleum ether extracts from *P. granatum*. While on the contrary significant increase of enzyme activity under Mustard oil treatment in compared to the control treatment.

A Significant increase in activity of **Alpha esterases** in case of petroleum ether extracts from *P. granatum* treatments. While on the contrary significant decrease of enzyme activity under Mustard oil treatment in compared to the control treatment.

Significant decrease in activity of **Beta esterases, Invertase, Trehalase** and **Amylase** in case of petroleum ether extracts from *P. granatum* and Mustard oil treatment in compared to the control treatment.

No significant increase in the activity of **Acetylcholinesterase** enzyme under petroleum ether extracts from *P. granatum*, while significant increased under Mustard oil treatment in compared to the control treatment.

8. Mammalian toxicological studies:

8.1. Hematological studies:

Overall, the results showed the treatments were non-significant on blood of albino rats. While Orange oil was significant in **Hb, PCV, MCV** and **MCH**. On the other hand, petroleum ether extracts from *P. granatum* was significant in **Hb and PCV**. Mustard oil was significant in **Neutrophils** and **Monocytes**.

8.2. Pathological study:

8.2.1. Liver:

Photomicrograph showing normal feature of hepatocytes and central veins and blood sinusoids of the liver of Orange oil, Mustard oil, acetone extracts from *R. officinalis* and petroleum ether extracts from *P. granatum* group.

8.2.2. Kidney:

Photomicrograph showing normal structure of kidney of acetone extracts from *R. officinalis* and petroleum ether extracts from *P. granatum* group. While showing congestion of the glomeruli of the kidney of Orange oil and Mustard oil group.

8.2.3. Testis:

Photomicrograph showing normal structure of seminiferous tubules and interstitial tissues of the testis of Orange oil, Mustard oil, and petroleum ether extracts from *P. granatum* group. While showing normal structure of seminiferous tubules but congestion of blood vessels in the interstitial tissues was seen in testis of acetone extracts from *R. officinalis* group.

8.2.4. Stomach:

Photomicrograph showing congestion of blood capillaries of stomach of Orange oil, Mustard oil, and petroleum ether extracts from *P. granatum* group. While showing nearly normal stomach structure in acetone extracts from *R. officinalis* group.

We concluded that this Orange oil, Mustard oil, acetone extracts from *R. officinalis* and petroleum ether extracts from *Punica. granatum* are

good effective and less harmful in the case of control *T. castaneum* and *S. oryzae* in the stored grains and recommended feeding of animals on these grains treated.