

# Studies on persistence and toxicity of two organophosphorus insecticides and plant extracts to stored product insect

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1- Toxicity of two organophosphorus insecticides and certain plant extracts against three insect species of stored products: The toxic effect of two organophosphorus insecticides i.e., malathion and pirimiphos-methyl as well as the acetone and petroleum ether extracts of four plants namely (Cubeb fruits *Piper cubeba*, Thyme flowering buds *Thymus vulgaris*, Damsissa leaves *Ambrosia maritima* and Marjoram leaves *Majorana hortensis*) was studied in the laboratory at  $26 \pm 2^\circ\text{C}$  and  $60 \pm 5$  RH against the adults of three insect species of stored products i.e., the rice weevil *Sitophilus oryzae* (L.), the lesser grain borer *Rhizopertha dominica* (F.) and the red flour beetle *Tribolium castaneum* (Herbst.) The obtained results showed the follows:

1.1- Malathion and pirimiphos-methyl (Actellic): The toxicity of malathion and pirimiphos-methyl to the adults of the three insect species was concentration and exposure - period — dependent. The toxic effect increased with increasing the concentration of the insecticide and the period of exposure.

SUMMARY -122- The susceptibility of the insects varied from insect species to another. *S. oryzae* adults were the most sensitive species followed by *T. castaneum* and *R. dominica* to the two insecticides. However, it was evident that the recommended concentration (8 ppm) of malathion for controlling stored product insects was not sufficient to achieve complete kill for the adults of *R. dominica* and *T. castaneum* even after longer exposure periods (3-7 days). Pirimiphos—methyl was more toxic to the various insect species than malathion. Results of the calculated lethal concentrations indicated that the LC<sub>50</sub> of malathion at 3 days exposure period was 0.93, 19.50 and 2.98 ppm for the adults of *S. oryzae*, *R. dominica* and *T. castaneum*, respectively. These values were 0.52, 11.22 and 2.20 ppm at 7 days exposure period for the three insect species, respectively. The LC<sub>50</sub> of pirimiphos—methyl were 0.09, 6.80 and 2.05 ppm after 3 days exposure period and 0.05, 4.98 and 1.36 ppm at 7 days exposure period for the various insect species, respectively. Meanwhile, the LC<sub>99</sub> of malathion at 3 days exposure period was 8.06, 556.30 and 50.22 ppm. These values were 2.86, 106.72 and 16.79 ppm at 7 days exposure period for *S. oryzae*, *R. dominica* and *T. castaneum*, respectively. The LC<sub>99</sub> values of pirimiphos — methyl were 0.42, 150.83 and 14.65 ppm at 3 days and 0.36, 24.98 and 5.76 ppm at 7 days for the three insect species, respectively. This result showed clearly that *R. dominica* adults were highly resistant to malathion and pirimiphos—methyl. Also, *T. castaneum* adults were highly tolerant to malathion and showed low tolerance ratio to pirimiphos—methyl. While, *S. oryzae* adults were sensitive to the two tested insecticides.

1.2- Plant extracts : The results revealed that the toxic effect of the various plant extracts was also concentration and exposure period—dependent. Susceptibility of the adults of the three insect species varied also from insect species to another. The adults of *S. oryzae* was the most sensitive species under study. Meanwhile, the toxic effect of the tested plant extracts varied from plant to another. The acetone extracts of the plants showed generally higher toxic effect than the petroleum ether extracts except the petroleum ether extract of Thyme flowering buds. For example, after 5 days exposure period the highest tested concentration (2% w/w) of the acetone extract of Cubab fruits caused 100, 100 and 36.7 % for the adults of *S. oryzae*, *R. dominica* and *T*

castaneum, respectively. The corresponding mortality values using the petroleum ether extract of the same plant were very low (8.9, 7.8 and 5.4%) for the three insect species, respectively. In case of the acetone extract of Thyme flowering buds at 2% (w/w) concentration and 5 days exposure period, mortalities were low (27.8, 11.1 and 4.4%) for the three insect species, respectively. Petroleum ether extract of this plant gave higher values (100, 100 and 16.5%). For Damsissa leaves acetone extract, mortalities were (36.7, 26.7 and 6.7%) with the three insect species, respectively. While, the petroleum ether extract of this plant gave also lower mortality values (20.0, 10.3 and 3.3%). The obtained mortalities with the acetone extract of Marjoram leaves were 33.3, 15.6 and 4.4 % at 2 % (w/w) concentration and 5 days exposure period for the three insect species, respectively. Petroleum ether extract of this plant gave 30.0, 10.0 and 4.5% indicating that the two extracts of this plant had low toxic effect on the insects. It was evident that only the acetone extract of Cubab fruits and petroleum ether extract of Thyme flowering buds were the most effective extracts. While, the petroleum ether extract of Cubab fruits was the least effective extract under study. Meanwhile, the reduction rates in the Fi-progeny of the insect species resulted from various concentrations [0.125-2% (w/w)] of the most effective plant extracts under study were between 16.8-100%, 30.2-100% and 21.8-100% with the acetone extract of Cubeb fruits for *S. oryzae*, *R. dominica* and *T. castaneum*, respectively. For the petroleum ether extract of this plant, these values were 8.2-63.8%, 5.0 - 34.8% and 4.6 - 24.2% for the three insect species, respectively. The corresponding values caused by the acetone extract of Thyme flowering buds were 14.8-55.1%, 3.9-49.9% and 19.7-49.2.% and for the petroleum ether extract of this plant were 20.3-100%, 14.4-100% and 4.9-79.8% for *S. oryzae*, *R. dominica* and *T. castaneum*, respectively. It was evident, that reduction rates in the Fi-progeny of *T. castaneum* caused by various plant extracts were generally higher than that of mortality.

2- Residual toxicity of the two organophosphorus insecticides and the plant extracts : The residual toxicity of the two organophosphorus insecticides i.e. malathion and pirimiphos-methyl was determined using two methods namely the GLC and a bio-test using the adults of *S. oryzae*, the most sensitive insect species under study. This bio-method was used only for determining the residual toxicity of the various plant extracts. The obtained results showed that pirimiphos-methyl had longer persistence than malathion and gave a greater protection extended to one year to the adults of *S. oryzae*. On the other hand, the tested plant extracts gave shorter protection to the adults of *S. oryzae* indicating that they have lower persistence than the two organophosphorus insecticides. Furthermore, results of the GLC indicated that pirimiphos-methyl persisted relatively longer in the tested grains and seeds than malathion. On the other hand, the persistence of the two insecticides was markedly greater. for cowpea seeds compared with maize and wheat grains. Also, loss rates of the two insecticides by washing of wheat and maize samples were around 50% of deposits of the unwashed grains at the initial time, while these were only around 35 % for the two insecticides in case of cowpea seeds. It was also evident, that the persistence of various the plant extracts was obviously lower than that of the two organophosphorus insecticides.

3- Effect of two plant extracts at 1% (w/w) alone and under MA of CO<sub>2</sub> on the populations of *S. oryzae* and *R. dominica* at various storage periods: Experiments were performed inside metal drums which were situated inside a breeding room at  $26 \pm 2^{\circ}\text{C}$  and  $60 \pm 5 \text{ RH}$ . \_ 200g wheat grains were treated with 1% (w/w) of Cubeb fruits (*P. cubeba*) acetone and/or petroleum ether extract of Thyme flowering buds (*T. vulgaris*) alone and under MA of 25% CO<sub>2</sub> against *S. oryzae* and *R. dominica*. Grains were infested with 32 adult insects of each species. Three replicates were used in each treatment. The results indicated that, all treatments caused a complete inhibition of *S. oryzae* adult populations in the wheat grains during the storage periods of 12 months. The number of *R. dominica* adults in wheat grains treated with *P. cubeba* fruits acetone extract, *T. vulgaris* flowering buds petroleum ether extract, MA of 25% CO<sub>2</sub> alone and untreated grains was 185.5, 173.0, 0.3 and 1922.0 after 2 months, respectively. While the combined treatments of each extract under MA gave a total inhibition of the two insect species populations during the whole storage periods. Thus, the combined treatment of the plant extract under MA of CO<sub>2</sub> was high effective against the tested insects and could be considered as an alternative method for methyl bromide to control stored product insects.

4- Effect of two plant extracts at 1% (w/w) alone and under MA of 25% CO on the weight loss of stored wheat grains : The weight loss of wheat grains was determined for one year in

200 g wheat grains treated with 1% (w/w) of *Cubeba* fruits *P. cubeba* acetone extract and Thyme flowering buds *T. vulgaris* petroleum ether extract alone and under MA of 25% CO<sub>2</sub> as well as MA of 25% CO<sub>2</sub> alone compared with untreated grains. Grains were infested with thirty two adults (0-15 days old) of *S. oryzae* or *R. dominica*. Results showed that the produced progenies differed from one treatment to another and the loss caused consequently differed according to the resulted progenies. The weight loss of wheat grains caused by *S. oryzae* adults at 6 months were 11.3, 4.8, 10.1, 5.4, 5.6 and 29.4% for the acetone extract of *Cubeba* fruits, *Cubeba* fruits acetone extract under MA of CO<sub>2</sub>, Thyme flowering buds petroleum ether extract, Thyme extract under MA of CO<sub>2</sub>, MA of 25% CO<sub>2</sub> alone and untreated grains, respectively. The corresponding values caused by *R. dominica* were 31.3, 1.3, 70.4, 0.8, 4.5 and 74.8% at 6 months storage for aforementioned treatments, respectively. Reduction rates in grains weight loss resulted from various treatments were 64.3, 84.6, 68.9, 83.5 and 81.0% and 42.9, 98.0, 0.5, 98.9 and 93.9% at 12 months storage period for *S. oryzae* and *R. dominica* adults, respectively. Data also indicated that the treatment of wheat grains with each plant extracts at 1% (w/w) under MA of 25% CO<sub>2</sub> reduced markedly the grain losses caused by the two insect species infestation.

5- Effect of certain treatments on germination of wheat grains and chlorophyll content of the seedlings : Results of tests performed to investigate the effects of four plant extracts at 4, 2 and 1 % (w/w) and two insecticides i.e. malathion at 16, 8, 4 ppm as well as pirimiphos-methyl at 20, 10, 5 ppm on wheat grains germination and chlorophyll content of the seedlings showed significant decrease in germination rates of wheat grains treated with the acetone and petroleum ether extracts of *P. cubeba* fruits and *T. vulgaris* flowering buds acetone extract at all tested concentrations, as well as, at the two highest concentrations of the petroleum ether extract of *T. vulgaris* and *A. maritima* leaves. Meanwhile, similar result was achieved at the highest concentration [4% (w/w)] of *A. maritima* acetone extract. On the other hand, no significant differences in the germination of wheat grains were found for two extracts of *M. hortensis* leaves and two insecticides at all tested concentrations compared with untreated grains. At the same time, no obvious differences between the chlorophyll content of the treated seedlings and control were found.

6- Effect of sublethal concentrations of two plant extracts, malathion and pirimiphos-methyl on some biological aspects of *T. castaneum* : Results of various treatments to *T. castaneum* adults, which treated with the sublethal concentrations (causing 50-60% adults mortality) of *Cubeba* fruits acetone extract, acetone and petroleum ether extracts of Thyme flowering buds, malathion and pirimiphos-methyl indicated significant reduction in the number of eggs laid daily per female during an observation period of two weeks for all tested treatments compared with the control. Percentages of decrease in egg numbers were 64.6, 51.2, 32.5, 39.0 and 27.8% for *Cubeba* acetone extract, Thyme acetone extract, Thyme petroleum ether extract, malathion and pirimiphos-methyl, respectively. On the other hand, no significant differences were achieved between all treatments and control for hatchability rates of eggs, duration of pupal stage and sex ratio. The mortality rates of larval instars were 33.3, 16.7, 16.7, 0.0 and 0.0% for aforementioned treatments, respectively. Treatment of *T. castaneum* adults with the sublethal concentration of *Cubeba* fruits acetone extract prolonged significantly pre-oviposition period, incubation period of eggs and durations of second and fourth larval instars in comparison to control. But, no significant differences were observed between the treatment and the control for duration of the larval stage and total developmental period of immature stages. The sublethal concentration of Thyme flowering buds acetone extract indicated significant decrease in the pre-oviposition period, durations of second and third larval instars, larval stage period and total developmental period of immature stages of *T. castaneum* compared with control. While, no significant differences were obtained between the treatment and control for incubation period of eggs. Also, significant decline in incubation period of eggs and the durations of third and fourth larval instars was achieved using sublethal concentration of petroleum ether extract of Thyme plant. But, no significant differences was found for pre-oviposition period, larval stage period and total developmental period of immature stages. Results indicated also that treatment of adults with the sublethal concentration of pirimiphos-methyl decreased significantly larva and the total developmental period of immature stages. Significant differences were found between the control for pre-oviposition period and incubation period of *T. castaneum* malathion and stage period. While, no differences were found of eggs.