# Toxicity Assessment of Four Commonly Used Insecticides to the Corn-Infesting Rice Weevil (Coleoptera: Curculionidae)[1]

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**Abstract** Rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae), is one of the most significant pests of corn (*Zea mays* L.) and other stored grains. Historically, synthetic insecticides have been widely used to control pest populations due to their effectiveness, convenience of storage and application, and persistent activity. This study aimed to assess the toxicity profiles of four commonly used insecticides, including pirimiphos-methyl, deltamethrin, deltamethrin plus (S)-methoprene, and malathion, to rice weevil adults. The weevils were exposed to a range of concentrations of each insecticide to generate concentration–mortality response curves and median lethal concentrations ($LC_{50}$) of each insecticide. Pirimiphos-methyl exhibited the highest toxicity to the adult weevils, with an $LC_{50}$ of 0.74 ppm (95% confidence limit [CL], 0.57–0.92). Based on nonoverlapping 95% CLs, we determined that the $LC_{50}$ of pirimiphos-methyl was significantly lower than the $LC_{50}$ of malathion (7.08 ppm [95% CL, 5.44–9.01]), deltamethrin (9.11 ppm [95% CL, 6.89–12.6]), and deltamethrin plus (S)-methoprene (13.94 ppm [95% CL, 9.71–18.60]). Thus, the relative toxicity of these four insecticides to adult rice weevils was pirimiphos-methyl $>$ malathion $>$ deltamethrin $>$ deltamethrin plus (S)-methoprene. We subsequently compared the $LC_{50}$ value of each insecticide with that of the label-recommended application rate and found that only malathion aligned with the recommended label rate. These findings could prove useful in refining application rates for the effective control of rice weevils infesting stored grains.

**Key Words** stored-grain pest, insecticide, deltamethrin, insect growth regulator, malathion

Food grains and pulses are the primary sources of nutrition for our global population (Poutanen et al. 2022; Stagnari et al. 2017), especially in tropical regions, making them vital in combating food insecurity (Bouchard et al. 2022) and ensuring their continued standing as major portions of daily diets in countries worldwide (Singh and Singh 1992). Wheat (*Triticum aestivum* L.), corn (*Zea mays* L.), and rice (*Oryza sativa* L.) are major staple cereals (Erenstein et al. 2022). Corn is a cereal crop that has been cultivated for thousands of years (Danforth 2009) and has become a vital food source for billions of people and serves as a primary ingredient in animal feed (Serna-Saldivar and Perez-Carrillo 2019). Moreover, it is a critical raw material in various industrial processes, such as the production of ethanol and corn syrup

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(Zahniser et al. 2019). Despite its essential role in food and industrial production, corn production and storage face several challenges, including pest and disease damage, which have resulted in significant losses over the years (Lamaoui et al. 2018).

The rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae), is a highly destructive pest that infests grain products, including corn (Davis 2011). Because of its preference for newly harvested grain, it is classified as a primary pest (Srivastava and Subramanian 2016). Bhargude et al. (2021) suggested that *S. oryzae* is native to India and subsequently spread globally through trade. This pest poses a significant threat to agricultural production in the southern United States and developing countries because it attacks a wide range of crops such as wheat, rice, maize, sorghum (*Sorghum bicolor* (L.) Moench), and other grains. Both the adult weevils and larvae are responsible for causing severe and extensive damage to grains (Zhang et al. 2021).

To control *S. oryzae* infestations, producers commonly use a range of management practices from cultural methods to chemical interventions for managing the pest in stored grains (Shankar and Abrol 2012). Some of the common practices include thorough cleaning and drying of grain to eliminate debris and prevent mold growth (Bhattacharyya et al. 2022). In addition, ensuring adequate ventilation in storage facilities to regulate temperature and moisture levels plays a crucial role in preventing mold growth and insect infestations. The use of airtight containers or bags is also recommended to mitigate moisture-related damage and deter insect activity. Using heat treatment, fumigation, and insecticide application also has proven to be effective in *S. oryzae* management (Bhattacharyya et al. 2022; Hagstrum et al. 2012). The use of insecticides in managing stored pests is a reasonable approach for preserving food products in storage, minimizing economic losses, and reducing hazards (Chulze 2010). Pests such as *S. oryzae* can infest food products during storage, resulting in damage and contamination that can adversely affect the quality and quantity and pose health risks (Kumar and Kalita 2017). However, the regulated application of insecticides can effectively control pest populations, thereby ensuring the safety and preservation of stored food products (Damalas and Eleftherohorinos 2011).

Since the 1950s, synthetic pesticides and fumigants have been used to protect stored grain due to their effectiveness, ease of storage and application, and long-term persistence (Attia et al. 2020). The commonly used fumigants are phosphine and methyl bromide (Hasan et al. 2020). However, methyl bromide poses significant risks to human health, causing a range of health issues and contributing to ozone layer depletion (Kim et al. 2021), thereby necessitating stringent restrictions on its use (Bramavath et al. 2017; Rajendran 2001). Presently insecticides commonly used include organophosphates, pyrethroids, and carbamates (Kljajic et al. 2006). In addition, insect growth regulators (IGRs) are considered a class of pesticides (Siddall 1976) widely used for controlling insect pests such as mosquitoes, cockroaches, and various Lepidoptera (Shinoda 2021). They function by targeting specific proteins associated with molting, thereby inducing a range of abnormal growth responses in insects, from postecdysis deformities to failed larval or pupae development (Sánchez-Ramos et al. 2024; Subramanian and Shankarganesh 2016).

Resistance to these insecticides is common in stored grain pests, including *S. oryzae*, underscoring the importance of regular monitoring (Studebaker et al.

2022). The main objective of this study was to assess the toxicity of four commonly used insecticide formulations, including pirimiphos-methyl, deltamethrin, deltamethrin plus (S)-methoprene (an IGR), and malathion. although the latter has been banned in the European Union but continues to be the most commonly used organophosphate insecticide in the United States (Kiely et al. 2004) to manage *S. oryzae*. Our ultimate objective in this study was to evaluate the toxicity of the current application rates of these materials to provide recommendations for enhancing the management strategies for controlling *S. oryzae* infestations in corn and other stored grains.

## Materials and Methods

**Insects.** Specimens of *S. oryzae* were collected from different corn-growing regions in Arkansas from 2018 to 2020 and transported to the toxicology laboratory at the Department of Entomology and Plant Pathology, University of Arkansas (Fayetteville, AR). The test insects were reared in 4.4-L plastic containers (Rubbermaid, Atlanta, GA), wherein whole kernels of corn were provided as the food source. The containers were modified with mesh lids to ensure proper airflow while preventing escapes. During the study, the laboratory was maintained at a constant temperature of 23°C and a relative humidity of 60–65%. This approach closely mimicked their natural diet and conditions to ensure the growth and reproduction of the insect population throughout the study.

**Bioassay.** The candidate insecticides were those commonly used for stored grain pest control (Table 1). They were formulated as either liquids or dust. Liquid insecticides were mixed with distilled water to obtain the desired concentrations and then applied to the rice weevils by using a specially designed Lab Spray Tower with a spray nozzle (Burkard Scientific, Uxbridge, United Kingdom; Belsky et al. 2021); distilled water was used as the control. The dust formulated insecticide was mixed with white corn flour to obtain the desired concentrations (Gazab Products, United Trading Inc., Des Plaines, IL); the control was corn flour alone. Treatment concentrations were chosen first based on the recommended application rate and then adjusted gradually based on the results of a series of preliminary bioassays. The primary goal of these preliminary bioassays was to determine the range of concentration that caused 5–95% mortality for each insecticide. To generate a response curve representing each insecticide's toxicity profile, at least five concentrations of each insecticide with three replications of 15 individuals/replication were used (Robertson et al. 2007).

For the bioassays, the specific insecticide treatment or control was sprayed onto Whatman filter paper Grade 1 WHA1001-045 (Sigma-Aldrich Inc., St. Louis, MO) and placed in a 120-ml polypropylene container (PLA-03346; Qorpak, Clinton, PA) to ensure uniform distribution. A group of 15 rice weevils was introduced into the container with a twist cap as a barrier to prevent escapes. This methodology aligns with the guidelines provided by Haliscak and Beeman (1983) and based on those by Busvine (1980). For the dust formulation, the desired treatments were mixed with corn flour and assigned to the prepared containers after which the insects were introduced in the containers and then shaken vigorously.

**Data collection and statistical analysis.** In each treatment, the mortality of *S. oryzae* adults was recorded every 24 h after exposure for 10 d to determine

Table 1. List of pesticides used in the bioassays including the commercial name, common name, mode of action group, and formulation for each insecticide tested.

| Product | Formulation | Chemical Group (IRAC Code) | Mode of Action | Physical Form |
|---|---|---|---|---|
| Actellic 5E (WinField United, St. Paul, MN) | Pirimiphos-methyl 57% | 1B - organophosphate | Acetylcholinesterase inhibitor | Liquid |
| Centynal EC (Central Life Sciences, Schaumburg, IL) | Deltamethrin 4.75% | 3A - pyrethroids, pyrethrins | Sodium channel modulator | Liquid |
| Diacon IGR Plus (Central Life Sciences) | Deltamethrin 4.75% + (S)-methoprene 11.4% | 3A - pyrethroids, pyrethrins + juvenile hormone | Sodium channel modulator + insect growth regulator | Liquid |
| Malathion Big-6 Dust (Balcom Chemicals Inc., Greeley, CO) | Malathion 6% | 1B - organophosphate | Acetylcholinesterase inhibitor | Dust |

whether there was delayed mortality (Biddinger et al. 1998; Phan et al. 2020). The data were analyzed using probit analysis in POLO Plus 2.0 (LeOra Software LLC 2005) as described by Robertson et al. (2007). The slope ± SE of the response curve, median lethal concentrations ($LC_{50}$s), 90% lethal concentrations, $LC_{90}$s, corresponding 95% confidence limits (CLs) of each were also calculated in the probit analysis with POLO Plus. The slope ± SE presented the rate at which mortality changed with exposure level. $LC_{50}$ (95% CL) and $LC_{90}$ (95% CL) provided the ranges that could be used to evaluate risk and compare with the application rates for further decision-making. The $LC_{50}$ ratios (95% CL) were used to compare the toxicity profiles of pesticide products and to determine whether one pesticide was significantly more or less toxic than the others.

## Results and Discussion

We found the toxicity profiles of the selected insecticides pirimiphos-methyl $>$ malathion $\geq$ deltamethrin $\geq$ deltamethrin plus (S)-methoprene by comparing the $LC_{50}$ values and their 95% CLs. Pirimiphos-methyl was most toxic, being ~15 times more toxic than the other three insecticides, followed by malathion and deltamethrin. Deltamethrin plus (S)-methoprene was the least toxic insecticide among all tested (Table 2; Fig. 1). Mortality reached the maximum at 48 h after exposure. Although the test insects were maintained and mortality was observed and recorded for 10 d after exposure, we observed no delayed mortality.

In a separate step, we compared the label-recommended application of these insecticides with their respective $LC_{50}$ values and found that the recommended rate for pirimiphos-methyl (6–10 ppm) not only exceeded its $LC_{50}$ at 0.74 ppm (95% CL, 0.57–0.92) but also surpassed its $LC_{90}$ at 1.68 ppm (95% CL, 1.31–2.56) by ~4 times (Fig. 1). By contrast, for deltamethrin and deltamethrin plus (S)-methoprene, their recommended rates (0.5–1.0 ppm) were lower than their respective $LC_{50}$ values at 9.11 (95% CL, 6.89–12.6) and 13.94 (95% CL, 9.71–18.60) (Fig. 1). Only the recommended application rate of malathion aligned with the toxicity profile.

The variations in the toxicity profiles among the selected insecticides can be attributed to differences in their modes of action and target specificity. For example, pirimiphos-methyl and malathion, classified as organophosphate insecticides, exert their insecticidal activity by inhibiting the action of acetylcholinesterase, an essential enzyme for proper nervous system function in insects (Brown 2000). Deltamethrin disrupts the normal functioning of the nervous system (Tapia et al. 2020) by affecting the voltage-gated sodium channels (Paudyal et al. 2016), leading to an influx of sodium ions and subsequent nerve excitation (Pitzer et al. 2021). This disrupts the transmission of nerve signals (Bhanu et al. 2011; Palmquist et al. 2012), resulting in paralysis and ultimately causing the death of the insects (Gupta et al. 2019; Magby and Richardson 2017). By contrast, (S)-methoprene, functioning as an IGR (Guo et al. 2023), operates by impeding chitin synthesis (Ghosal 2018), with chitin being a crucial component of the exoskeleton (Muthukrishnan et al. 2020). By inhibiting chitin synthesis, (S)-methoprene interferes with the proper growth and development of insects, ultimately leading to their control (Fulton et al. 2013; Merzendorfer and Zimoch 2003).

**Table 2. Toxicity response of *Sitophilus oryzae* to selected insecticides at 48 h after treatment.**

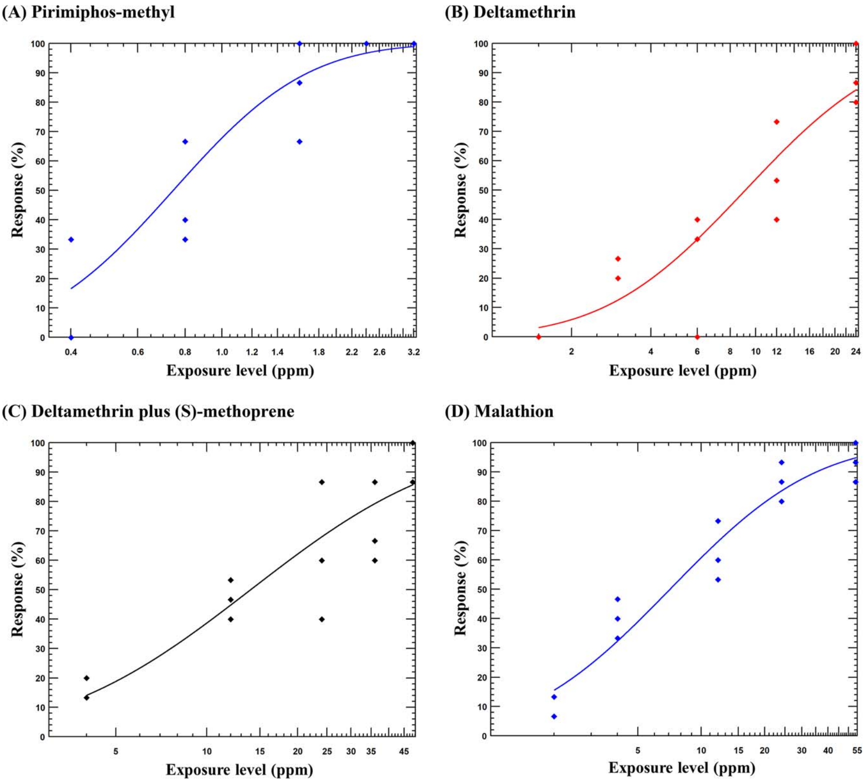
| Active Ingredient* | *N*** | Slope ± SE† | $LC_{50}$ (ppm) (95% CL) | $LC_{90}$ (ppm) (95% CL) | Recommended Application Rate (ppm)‡ |
|---|---|---|---|---|---|
| Pirimiphos-methyl | 225 | 3.608 ± 0.411 | 0.74 (0.57–0.92) | 1.68 (1.31–2.56) | 6–8 |
| Malathion | 225 | 1.842 ± 0.214 | 7.08 (5.44–9.01) | 35.13 (25.00–57.58) | 10 |
| Deltamethrin | 225 | 2.381 ± 0.283 | 9.11 (6.89–12.6) | 31.45 (20.34–70.50) | 0.5–1.0 |
| Deltamethrin + (S)-methoprene | 225 | 1.984 ± 0.262 | 13.94 (9.71–18.60) | 61.69 (41.05–131.0) | 0.5–1.0 |

* The products are listed based on toxicity profile, from high to low.

** *N* is the number of individuals tested for each product. Response regression lines are presented by slope ± SE, $LC_{50}$ (in ppm), and $LC_{90}$ (in ppm).

† Control mortality was 0% during the study period.

‡ Recommended application rates were obtained from the pesticide product labels (Balcom Chemicals Inc. 1975; Central Garden & Pet Company 2016a, 2016b; Winfield Solutions LLC 2015).



(A) Pirimiphos-methyl

(B) Deltamethrin

(C) Deltamethrin plus (S)-methoprene

(D) Malathion

**Fig. 1. Toxicity response of *Sitophilus oryzae* to selected pesticides: (A) pirimiphos-methyl, (B) deltamethrin, (C) deltamethrin plus (S)-methoprene, and (D) malathion at 48 h after treatment.**

The effectiveness of pirimiphos-methyl in controlling *S. oryzae* has been demonstrated in several studies. Huang and Subramanyam (2005) found that pirimiphos-methyl at the rate of 4–6 ppm was toxic to *S. oryzae*, whereas Rumbos et al. (2013) reported 100% mortality of adult *S. oryzae* after 7 d of exposure to doses of 1–4 ppm of pirimiphos-methyl. In a recent study conducted by Soltan et al. (2020), the acute toxicity of different insecticides, including pirimiphos-methyl and deltamethrin, was compared where researchers found that concentrations ranging from 0.35 to 0.85 ppm of both pirimiphos-methyl and deltamethrin were effective against *S. oryzae*.

In the study by Derbalah et al. (2021), the researchers evaluated different methods for controlling *S. oryzae* by considering parameters such as adult mortality, offspring production, mode of action, and grain quality. They found that a treatment rate of malathion at 0.06 ppm was effective in controlling *S. oryzae*. By contrast, previous studies by Iqbal et al. (2012) evaluated the toxicity of cypermethrin and malathion to *S. oryzae*. The results of this study indicated that malathion at the rate of 5 ml with an $LC_{50}$ of 7.54 ppm after 48 h of exposure was effective. Regarding (S)-methoprene, Wijayaratne et al. (2018) found limited toxicity of (S)-methoprene towards stored product adult pests. Its effectiveness in managing adult insects is comparatively lower than its efficacy against juvenile insects. Another possible reason for the difference in toxicity could be the development of resistance within the *S. oryzae* population toward deltamethrin plus (S)-methoprene, deltamethrin, and malathion due to the repeated use of these pesticides, thereby reducing the effectiveness of these chemicals; in contrast, there is currently no evidence of widespread resistance to pirimiphos-methyl in *S. oryzae* (Khan et al. 2022).

The difference between the recommended label rates and the $LC_{50}$ values may arise from several factors. First, the recommended label rates are typically established as technical rates, designed to address a range of insect pests beyond those specifically mentioned in the study. As a result, these rates might be either overapplied or underapplied when applied to the insect species under investigation. However, these rates may align well with other groups of insect pests listed on the approved labels of these pesticides. Moreover, these recommended rates are applied as preventative methods and therefore may have higher rates. Furthermore, in this study, we used formulated concentrations of the insecticides which could lead to variations in the effectiveness to the target insects and influence the observed outcomes.

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