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Impact of Selected Pesticides on the Population of Leaf Miners on Chinese Celery, *Apium graveolens* var. *secalinum*

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Abstract: In Malaysia, leaf miners, primarily from the Diptera order and Agromyzidae family, are significant agricultural pests that cause extensive damage to a wide range of crops. This study evaluates the efficacy of two insecticides, deltamethrin and cyromazine, in managing leaf miner populations on Chinese celery, *Apium graveolens* var. *secalinum*, in the Cameron Highlands. Utilizing a completely randomized design (CRD) approach, the research measured the impact of these pesticides on the pests' larval and pupal development stages. The field trials were conducted on an experimental plot in Habu, Ringlet, where treatments included three concentrations of cyromazine and a standard concentration of deltamethrin, with water serving as a control. Data collection methods combined direct and indirect observational techniques, including the use of Yellow Sticky Traps (YST) for adult activity and detailed leaf examinations for larval and pupal stages. Statistical analysis was performed using the Henderson-Tilton formula to assess the reduction in pest populations. Results indicated that deltamethrin significantly reduced leaf miner activity, demonstrating a 35.2% decrease in leaf mining and a 47.5% reduction in pupal emergence, confirming its immediate effectiveness. Conversely, cyromazine showed a gradual impact, significantly disrupting larval to pupal transition, with the highest dose achieving a 17.9% reduction in pupal emergence. This study highlights the differential impacts of these pesticides and contributes valuable insights into effective pest management strategies in tropical agriculture.

Keywords: leaf miner, deltamethrin, cyromazine, crop protection, pest management

1. Introduction

Leaf miners, primarily from the Diptera order and Agromyzidae family, pose a significant agricultural and ecological challenge in Malaysia, inflicting notable damage on a range of crops, ornamentals, and forest vegetation. These pests are distinguishable by their larval feeding habits, which result in distinctive serpentine tunnels in leaf tissues, and their distinctive damage signatures serve as indicators of infestation. The economic repercussions of leaf miner infestations are profound, manifesting as diminished crop yields, compromised produce quality, and escalated pest management costs (Hamza et al., 2023; Sadiq, Al-Nadawi, & Mahmood, 2020). Predominantly, three species, *Liriomyza sativae*, *L. huidobrensis*, and *Chromatomyia horticola*, have been identified, with the former being the most prevalent in local crops, including tomatoes, where it accounts for 56% of pest attacks (Ahya & Liyana, 2018). Unchecked, leaf miners can result in yield losses of up to 30% (Ahya & Liyana, 2018). Malaysia's climate, conducive to their year-round proliferation, exacerbates their impact across various host plants, encompassing vegetables like tomatoes, eggplants, and cucumbers; fruit trees including citrus, mango, and guava; and ornamentals such as chrysanthemums and roses. Furthermore, leaf miners' presence in forest ecosystems disrupts native plant life and ecological balance (Ridland et al., 2020). The life cycle of leaf miners progresses through egg, larva, pupa, and adult stages, with adult females depositing eggs on the leaf undersides, particularly near veins. Larvae emerge to burrow into and feed on internal tissues, thereby weakening the plant, increasing disease susceptibility, and diminishing aesthetic and vigour.

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In Malaysia, addressing leaf miner infestations necessitates a comprehensive strategy, integrating crop management techniques, biological control through natural enemies, and, when necessary, judicious insecticide application to balance pest control with environmental and health considerations. Specifically, methods such as crop rotation and the removal of infested foliage are coupled with the utilisation of parasitic wasps and predators to mitigate leaf miner populations (Mugala et al., 2022; Supartha et al., 2022). Among insecticides, deltamethrin, a synthetic agent within the pyrethroid class known for its broad-spectrum efficacy against a variety of insects, including mosquitoes and crop pests, is preferred or its relative mammalian safety and neurotoxic action on insects, causing paralysis and death (Yadav et al., 2023). Conversely, cyromazine, an insect growth regulator, targets pest development by obstructing chitin synthesis, thereby impeding their growth, and is effective against flies and certain worms (Khan, 2023). Notably, both are contact insecticides, necessitating direct exposure to the targeted pests for effectiveness. This study evaluates the efficiency of three distinct doses of Frontier™ (active ingredient: cyromazine) and a single recommended dose of Decis™ (active ingredient: deltamethrin) in controlling leaf miner populations on *Apium graveolens* var. *secalinum* in Cameron Highlands. The outcomes aim to elucidate the practicality and environmental safety of these pesticides in local agricultural practices, contributing to the development of sustainable pest management strategies.

2. Material and Methods

2.1 Study Location and Experimental Design

The study was conducted on an experimental plot located in Habu, Ringlet, Cameron Highlands, Pahang (4.4489558, 101.3770207), maintained by a local farmer who undertook all horticultural activities. The research team was involved in pesticide application and data collection, utilising a battery-powered knapsack sprayer to ensure uniform application. The person administering the treatments remained consistent throughout the study to maintain application uniformity. All treatments were organized in a completely randomized design (CRD). The treatments included three concentrations of cyromazine and a single concentration of deltamethrin), with normal tap water serving as the control. The treatments were as follows: T1, Negative Control (Tap Water); T2, Positive Control (deltamethrin) at a concentration of 5.5 mL per 10 liters; T3, Cyromazine at 0.5x the recommended rate (1g per 10 liters); T4, Cyromazine at 1.0x the recommended rate (2g per 10 liters); and T5, Cyromazine at 1.5x the recommended rate (4g per 10 liters). Applications were made twice during the cropping season, specifically at 14 days after transplanting (DAT) and 22 DAT, with each treatment replicated three times.

2.2 Insect Collections

The study adopted a combination of direct and indirect observational methods to evaluate the impact of pesticide treatments on the populations of leaf miners, with a particular focus on their larval and pupal stages. Yellow Sticky Traps (YST) were strategically placed within the experimental plots 24 hours prior to the application of pesticides. Positioned approximately 30cm above the top of the leaf canopy, these traps were collected after 24 hours exposure for species identification and quantification in the laboratory. In addition to traps, the research methodology included non-destructive leaf observations. We examined ten leaves from ten randomly selected plants in each treatment group to detect leaf miner infestations. The examination's focus was on identifying larvae through obvious damage to the leaves. Finally, a destructive leaf sampling approach was employed, whereby leaves from ten plants per plot manifesting infestation symptoms were collected for in-depth analysis in the laboratory. These samples, transported in 4 oz. plastic containers, facilitated the examination of the pupal stage of leaf miners, paying close attention to emergence rates and parasitoid occurrences. All sampling were done simultaneously on the 14 and 22 DAT. Through these meticulously designed methods, the study aimed to provide a comprehensive assessment of pesticide efficacy against leaf miner populations across different developmental stages.

2.3 Statistical Analyses

To assess the efficacy of pesticide treatments on leaf miner populations, both larval and pupal stages were quantified before and after treatment applications. The reduction in leaf miner populations attributable to each treatment was determined using an adaptation method based on the Henderson-Tilton formula, a widely recognized method in entomological research for calculating the percentage of pest control provided by a treatment relative to untreated controls (Paoli et al., 2023). The formula is given by:

$$Percentage\ Control = 100 * \left(1 - \left(\frac{T_a * C_b}{T_b * C_a}\right)\right) \quad (1)$$

where:
T_b = Number of leaf miner symptoms observed per sampling unit before treatment,

T_a = Number of leaf miner symptoms observed per sampling unit after treatment,
C_b = Number of leaf miner symptoms in the untreated (check) plot before treatment,
C_a = Number of leaf miner symptoms in the untreated (check) plot after treatment.

This formula allows for the calculation of treatment efficacy by adjusting for natural changes in leaf miner populations in the control plots, thus providing a more accurate measure of treatment impact. To analyse the collected data, the Minitab19 software was employed, specifically to conduct statistical methods such as comparative analyses, including the t-test.

3. Results

3.1 Yellow Sticky Traps (YST)

Prior to the pesticide application at 18 days after transplanting (DAT), Yellow Sticky Traps (YST) captured 181 adult leaf miners. Statistical analysis revealed no significant differences among the treatment groups (F (4, 10) = 0.99, p = 0.457). Post-application (22 DAT), this figure slightly decreased to 153, again without significant differences (F (4, 10) = 0.17, p = 0.950). Subsequent analysis comparing treatments to control post-spray revealed no statistically significant differences in adult leaf miner captures (Table 1) across all comparisons (control vs. deltamethrin, control vs. 0.5x cyromazine, etc.), with p-values ranging from 0.288 to 0.629. These findings suggest that the number of adult leaf miners caught on YST after treatment does not necessarily show how well the treatment worked. This could be because the plots were so close to each other (less than 1 meter), which allowed for contamination between treatments – even with the presence of a 5 x 2-meter PE plastic sheet as a physical barrier during spraying. Therefore, the captured adult leaf miner count serves merely as an approximate indicator of the farm's adult leaf miner population rather than a direct measure of the pesticide treatments' effectiveness.

Table 1: Comparative Analysis of Leaf Miner Captures via Yellow Sticky Traps Post-Pesticide Application

Pair Test	StDev	95% CI for difference	t-value	df	p-value
Control-deltamethrin	1.29	(-3.59, 2.26)	-0.63	4	0.561
Control-0.5x cyromazine	2.00	(-6.53,2.53)	-1.22	4	0.288
Control-1.0x cyromazine	2.77	(-7.61,4.94)	-0.59	4	0.587
Control-2.0x cyromazine	4.69	(-12.63,8.63)	-0.52	4	0.629

* P>0.05 indicates that there were no significant differences between the treatments in the number of adult leaf miners captured on the YST between treatments and control post-spraying.

3.2 Non-destructive leaf observation

Prior to the application of pesticides, the survey on *A. graveolens* var. *secalinum* revealed that leaf miners' larvae had infested a total of 389 leaves across a sample of 150 plants. The statistical analysis conducted at a 95% confidence interval indicated a uniform level of infestation across all experimental groups [F(4,145)=1.99, p=0.099], suggesting no significant difference in the initial distribution of leaf mining symptoms among the treatments. Notably, the highest mean number of symptomatic leaves was observed in the deltamethrin group [M=3.5, SD=2.57], implying a higher baseline presence of leaf-mining larvae within this group. However, this mean represents only the general presence of the pest in the field and not the efficacy of the treatment.

Following the pesticide treatments, the total number of leaves manifesting mining symptoms attributed to leaf miners' larvae decreased to 337 across the same cohort of 150 plants. Statistical analysis post-treatment revealed no significant variance in the efficacy of the treatments [F (4, 145) = 0.80, p = 0.525], suggesting a generally uniform reduction in leaf mining symptoms across all treatments, with the exception of the untreated control. Notwithstanding this overall reduction, the quantitative data alone does not confirm the treatments' true effectiveness. A noteworthy exception was the deltamethrin treatment, which demonstrated a statistically significant decrease in the average number of mines, from a mean of 3.50 (SD=2.57) pre-treatment to 2.27 (SD=1.75) post-treatment, confirmed by a t-test [t (58) = 2.18, p = 0.033], highlighting its singular efficacy among the treatments studied.

Figure 1 presents a comparative quantification summary of leaf mining symptoms on *A. graveolens* var. *secalinum* across five treatment groups, both pre- and post-pesticide application, distinguished by blue and orange bars, respectively. The control group exhibits a marginal increase in symptoms post-treatment, whereas the deltamethrin group demonstrates a marked reduction, as evidenced by a significantly lower mean post-spray (M=2.27) compared to pre-spray (M=3.5), denoting the only statistically significant difference across the treatments. The cyromazine-treated groups (0.5x, 1.0x, and 2.0x the recommended rate) show a modest decline in symptoms after spraying, yet not to a degree suggesting statistical significance. The error bars indicate variability within each group, which appears consistent across treatments and implies uniform reliability of the data collected. This graphical representation underscores deltamethrin's effectiveness relative to cyromazine and the control, within the study's parameters.

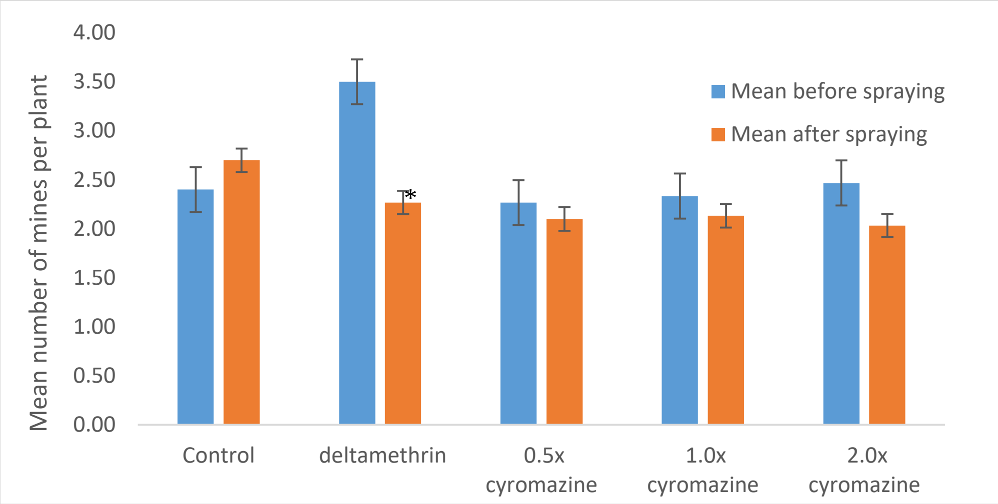

Fig. 1: Impact of Pesticide Treatments on Leaf Mining Symptoms: Pre- and Post-Application Analysis.

Post-treatment observations indicated a variable reduction in leaf mining activity across the treatment groups. In particular, deltamethrin was the most efficient, lowering the incidence of mines by 35.2%, as seen in a decrease in mine presence from 0.35 to 0.23 instances per plant (Table 2). This contrasted with the 2.0x cyromazine treatment, which yielded a 17.6% reduction, lowering the incidence from 0.25 to 0.20. Conversely, the control group saw a 12.5% increase in mining incidents, suggesting the potential for a leaf miner population surge if not adequately managed. These results underscore the varying degrees of success in controlling leaf miner infestations and highlight the risk of further outbreaks should the plot remain unchecked.

Table 2: Differential Efficacy of Pesticides Treatments Against Leaf Miner Larvae Based on Mining Incidence (n=300)

Treatment	Mean incidence of mines before treatment	Mean incidence of mines after treatment	% Increase/decrease
control	0.24	0.27	+12.5
deltamethrin	0.35	0.23	-35.2
0.5x cyromazine	0.23	0.21	-7.4
1.0x cyromazine	0.23	0.21	-8.6
2.0x cyromazine	0.25	0.20	-17.6

A deeper analysis utilising the Henderson-Tilton formula further elucidated the relative efficacy of the pesticide treatments (Table 3). Deltamethrin emerged as the most efficacious treatment, with a 42.4% control rate, followed by the 2.0x cyromazine treatment, which achieved a 26.7% control rate. The 1.0x and 0.5x cyromazine treatments registered control rates of 18.7% and 17.6%, respectively. This methodological approach provides a robust framework for assessing the impacts of pesticide applications on leaf miner larvae, considering the population dynamics before and after the intervention.

Table 3: Percentage Control of Pesticides Treatments Against Leaf Miner Larvae Based on the Henderson-Tilton Formula

Treatment	Recommended rate	Number of mines before treatment	Number of mines after treatment	% control
deltamethrin	5.5ml/10L	105	68	42.4
0.5x cyromazine	1g/10L	68	63	17.6
1.0x cyromazine	2g/10L	70	64	18.7
2.0x cyromazine	4g/10L	74	61	26.7
control	-	72	81	-

3.3 Destructive leaf sampling

After the spraying treatments were carried out, there was a decrease in the number of pupae emerging in all treatment groups, including the control group (Table 4). This indicates an inhibitory effect on the life cycle of leaf miners. A significant drop was seen after using deltamethrin, with a 47.5% reduction in pupal emergence, suggesting its high effectiveness in preventing leaf miners from maturing into adults. The 2.0x concentration of cyromazine also led to a 17.9% decrease in emergences. On the other hand, the 1.0x and 0.5x concentrations of cyromazine showed only slight impacts, with decreases of 3.2% and 2.9%, respectively, revealing that cyromazine effectiveness is dependent on its dosage. However, while these results hint at a trend of reduced pupal emergence post-treatment, they cannot definitively be linked to the rate due to how the observations were conducted. The data gathered mainly estimates the remaining population size after treatment, offering insights into curbing leaf miner populations and indirectly indicating the impact of treatments on their life cycle. These findings, while indicative, underscore the necessity for further detailed research to accurately determine the pupal emergence rates and understand the full implications of pesticide applications on leaf miner populations.

Table 4: Impact of Post-Pesticide Applications on Pupal Emergence Rates (n=300)

Treatment	Mean number of emergences before treatment	Mean number of emergences after treatment	% Increase/decrease
control	0.14	0.10	-29.3
deltamethrin	0.20	0.10	-47.5
0.5x cyromazine	0.12	0.11	-2.9
1.0x cyromazine	0.10	0.10	-3.2
2.0x cyromazine	0.13	0.11	-17.9

In the evaluation of pesticide efficacy against leaf miner pupae on *A. graveolens* var. *secalinum* using the Henderson-Tilton formula, deltamethrin emerged as the singular treatment, achieving a control percentage of 25.7%, as delineated in Table 5. This indicates a discernible reduction in pupal emergence post-application, setting deltamethrin apart from other treatments, which did not exhibit any control effect on pupal emergence. Such a distinct outcome underscores the specific effectiveness of deltamethrin in curbing the developmental stage of leaf miner pupae. In contrast, treatments involving various concentrations of cyromazine showed no statistically significant control effect, suggesting that, despite cyromazine known efficacy under certain conditions, it did not effectively prevent pupal emergence in this specific set of experimental conditions. The lack of control effect observed with cyromazine treatments points to the critical role of environmental and management factors in influencing pesticide efficacy. Although laboratory findings have indicated the potential effectiveness of cyromazine, the practical application outcomes observed in these experiments suggest that its efficacy may be compromised. These results highlight the necessity of considering a broad spectrum of environmental and application-related variables in evaluating pesticide performance, particularly in field conditions, where such factors can significantly impact the outcomes of pest control measures.

Table 5: Comparative Efficacy of Pesticide Treatments on Leaf Miner Pupa Emergence Post-Application Using the Henderson-Tilton Formula

Treatment	Recommended rate	Number of emergences before treatment	Number of emergences after treatment	% control
deltamethrin	5.5ml/10L	59	31	25.7
0.5x cyromazine	1g/10L	35	34	No control
1.0x cyromazine	2g/10L	31	30	No control
2.0x cyromazine	4g/10L	39	32	No control
control	-	41	29	-

4. Discussions

The comparative assessment of deltamethrin and cyromazine efficacy against leaf miner populations reveals a nuanced landscape of pesticide performance, with deltamethrin consistently demonstrating superior control capabilities. This differential efficacy underscores the importance of selecting an appropriate insecticidal strategy based on the pest's biological characteristics and the environmental context of the application. Deltamethrin, a member of the pyrethroid class, leverages its neurotoxic effects to incapacitate and eliminate a wide range of insect pests effectively. Its mechanism, characterized by disrupting the nervous system, results in rapid paralysis followed by death, a trait that underscores its broad utility in both the agricultural and public health sectors. The insecticide's contact activity, coupled with its notable residual efficacy, ensures prolonged protection against pests, thereby reducing the necessity for frequent reapplications. Such broad-spectrum utility, however, necessitates careful consideration of its impacts beyond the targeted pests. An illustrative example of deltamethrin's application and its broad-ranging effects comes from a study in Tunisia, which focused on the impact of deltamethrin treatments on chickpea leaf miner larvae and associated parasitoids (Soltani et al., 2020). While the application of deltamethrin successfully reduced the population of adult leaf miners, it also led to higher mortalities among natural enemies within the chickpea ecosystem. This outcome highlights a critical challenge associated with the use of broad-spectrum insecticides like deltamethrin: the inadvertent impact on non-targeted and beneficial insect populations.

On the other hand, cyromazine operates through a distinct mode of action as an insect growth regulator, targeting the synthesis of chitin, a critical component of the insect exoskeleton. By impeding normal development, cyromazine exhibits specific efficacy against certain pests, such as flies and worms, which rely heavily on chitin for their larval stages. However, our study's findings suggest that, while effective in specific contexts, cyromazine overall pest control performance may fall short when compared to the more potent deltamethrin. The requirement for direct contact with the target pest, combined with its absence of systemic action, limits cyromazine ability to control pests that reside within the plant tissue or beyond the immediate surface, potentially diminishing its effectiveness in comprehensive pest management strategies. Research done by Wang et al. (2020) reveals that cyromazine, despite showing no immediate lethal effects on larvae, significantly impacts the subsequent life stage, with treated larvae failing to progress to viable pupae. On the other hand, additional research revealed that cyromazine has the potential to endure in water and soil (Yao et al., 2024). As a result, its sustained existence within the ecosystem could potentially augment its effectiveness for a prolonged duration.

The efficacy of insecticides, including deltamethrin and cyromazine, is subject to the intricacies of application methods, target pest biology, environmental conditions, and the emerging challenge of resistance development. Thus, an integrated pest management approach, emphasizing the strategic use of both insecticides, rotation to mitigate resistance, and adherence to best application practices, is imperative for sustaining effective control over pest populations and safeguarding agricultural productivity. For instance, resistance to cyromazine has been observed in populations of pink stem borer, (PSB) across Pakistan, China, and India (Mansoor, 2023). Additionally, research on the *Spodoptera frugiperda* (FAW) has highlighted the role of the P450 gene, SfCYP321A8, which, when upregulated in larvae, contributes to deltamethrin resistance (Chen & Palli, 2022). These few examples illustrate the critical importance of incorporating biological knowledge into Integrated Pest Management (IPM) strategies to effectively mitigate resistance development and ensure the long-term efficacy of pest control measures. Such findings also underscore the complexity of resistance mechanisms and the need for careful management and rotation of insecticides to slow the evolution of resistance in pests such as the leaf miners.

5. Conclusions

In light of these observations, it becomes evident that deltamethrin's broader spectrum of action, enhanced potency, and extended residual effect position it as a more effective option for managing leaf miner infestations. In comparison to the immediate efficacy observed with deltamethrin, cyromazine delayed impact on pest populations through interference with development stages underscores a complementary approach to pest management. While deltamethrin offers rapid control of existing pest populations, cyromazine contributes to long-term population management by preventing the emergence of future generations. This dual approach, leveraging the immediate and residual impacts of different insecticides, is integral to developing comprehensive pest management strategies that are both effective and sustainable. The contrasting but potentially synergistic effects of deltamethrin and cyromazine highlight the complexity of pest control and the necessity for integrated strategies that address both immediate infestations and future leaf miner population resurgence in the Cameron Highlands.

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