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DATA NOTE

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Selective and persistent toxicity of seven insecticides to five egg parasitoids of *Spodoptera frugiperda*

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Abstract

Background *Spodoptera frugiperda,* a major migratory and invasive pest, inflicts significant yield loss on rice and maize in China. As part of an integrated pest management system, biological control agents can be used against *S. frugiperda*, especially egg parasitoids. However, limited evidence exists regarding the combined and persistent effects of various pest control products on those parasitoids.

Results This study examined the selective and persistent toxicity of seven approved pesticides [chlorantraniliprole, lufenuron, emamectin benzoate, spinetoram (synthetic pesticides), *Mamestra brassicae* Nuclear Polyhedrosis Virus (MabrNPV), *Bacillus thuringiensis* (Bt) (biopesticides) and a chlorantraniliprole-lufenuron mixture (3:1) treatment] to five species of egg parasitoids, namely *Trichogramma dendrolimi*, *Trichogramma chilonis*, *Trichogramma mwanzai*, *Trichogrammatoidea lutea*, and *Telenomus remus*. The residual toxicity tests revealed that spinetoram showed high toxicity to *T. mwanzai*, *T. dendrolimi*, and *T. chilonis* in adults stage with mortality of over 92.6%, but caused low mortality in *T. lutea* (64.9%) and *T. remus* (49.2%) when used at the recommended rate. However, after treated by chlorantraniliprole, lunefuron, chl. + luf. (3:1), MabrNPV, and Bt, the mortality rates of all tested parasitoid adults were below 25% (lunefuron lower than 10%). A 3-day emamectin benzoate treatment caused 90% mortality in *T. mwanzai*, *T. lutea*, *T. dendrolimi*, and *T. chilonis* adults. The 3:1 mixture of chlorantraniliprole and lufenuron did not affect the lifespan of *T. mwanzai* and *T. lutea*. Chlorantraniliprole exhibited exceptional safety for all developmental stages (adult, egg, and pupa) of the five egg parasitoid species. A risk analysis indicated that chlorantraniliprole, MabrNPV, Bt, and the 3:1 mixture had the least toxicity to the five tested parasitoid adults, followed by lufenuron. Conversely, spinetoram and emamectin benzoate displayed moderate toxicity to adults of all tested parasitoid species.

Conclusion Our findings indicate that chlorantraniliprole is safe for the five egg parasitoids species studied even after prolonged use and can be used in conjunction with lufenuron. However, spinetoram and emamectin benzoate had the potential to be harmful to these parasitoids.

Keywords Biological control, Synthetic pesticides, Persistent toxicity, Integrated pest management, Risk assessment

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Introduction

The fall armyworm, *Spodoptera frugiperda* (J.E. Smith), is a notorious pest infesting a wide range of agricultural crops and has a strong preference for plants such as Poaceae, Fabaceae, Solanaceae, Asteraceae, Rosaceae, Maize and Chenopodiaceous (Kenis et al. 2023). These hazards have been documented losses up to 58% in certain years (Johnson 1987; Liu et al. 2022a; Wang et al. 2022a; Kenis et al. 2023; Cruz et al. 1999; Overton et al. 2021). Synthetic pesticides have been widely used as the primary defense tactic against this pest (van den Berg et al. 2021; Kenis et al. 2023). However, the application of these pesticides often poses harm to beneficial organisms (Desneux et al. 2007; Palma-Onetto et al. 2021; Xiao et al. 2016) and human health (Damalas and Eleftherohorinos 2011). Furthermore, the long-term misuse leads to the development of pest resistances (Yao et al. 2017; Paula et al. 2021; Kim et al. 2021; Li et al. 2022; Seal and Kakkar 2013; Yu 1991). Therefore, for green and sustainable pest management, it is recommended to utilize environmentally-friendly solutions, such as the use of biological control agents (BCAs). This can help reduce pesticide usage, and the combination of multiple pest control methods is also a manifestation of integrated pest management (IPM) strategies (Zang et al. 2021; Verheggen et al. 2022; Li et al. 2023).

It is crucial to consider their potential harm to biological control when selecting synthetic pesticides in IPM programs. Notably, the specificity towards the targeted pest(s) and low risks to beneficial organisms, including parasitic wasps (Hall and Duncan 1984) is key for successful IPM. The wasps often exhibit higher sensitivity to pesticides, which can impact their effectiveness in pest control. Currently, 121 species of parasitoids targeting *S. frugiperda* have been studied extensively (Molina-Ochoa et al. 2003). Notably, *Trichogramma* spp. (Hymenoptera: Trichogrammatidae) (Pastori et al. 2007; Li et al. 2023; Kenis et al. 2023), *Trichogrammatoidea* spp. (Hymenoptera: Trichogrammatidae) (Sun et al. 2021), and *Telenomus* spp. (Hymenoptera: Scelionidae) (Agboyi et al. 2020; Colmenarez et al. 2022; Laminou et al. 2020) have been identified as among the most effective egg parasitoids for controlling *S. frugiperda*. Their importance stems from the fact that these parasitoids are highly effective in controlling insect pests, particularly during the early growth stages (Dai et al. 2014; Paredes-Sánchez et al. 2021; Sani et al. 2020; Zang et al. 2021). Multiple studies have shown the sensitivity of egg parasitoids to pesticides (Desneux et al. 2007). For instance, *T. dendrolimus* Matsumura was found to be susceptible to ethofenprox and cartap (Takada et al. 2001). Conversely, hormesis, where low pesticide doses benefit the species, has been observed in some cases (Wang et al. 2022b). Similarly, following 5 days of residual insecticide exposure, this hormetic effect can influence the parasitism rate of *Encarsia formosa* Gahan on whitefly eggs (Wang et al. 2019). Contrary to this, *Telenomus remus* Nixon adults showed significant sensitivity to pesticides, including to indoxacarb, tebufenozide, chlorfenapyr, methomyl, alpha-cypermethrin, and chlorpyrifos (Liu et al. 2016). Moreover, *E. formosa* was found affected synthetic pyrethroids (such as *β*-cypermethrin, deltamethrin, fenpropathrin) and most neonicotinoids (including imidacloprid, thiamethoxam, nitenpyram, acetamiprid), while cyantraniliprole, sulfoxaflor, and chlorantraniliprole showed adverse effects (Prabhaker et al. 2011; Sugiyama et al. 2011). Recently, several new insecticides, including insect growth regulators (IGRs), *Bacillus thuringiensis* (Bt), *Beauveria bassiana* and *Mamestra brassicae* nucleo polyhedron virus (MabrNPV), have emerged as popular modern insecticides in agriculture, horticulture, and public health for pest control due to their perceived safety towards beneficial arthropods (Siebert et al. 2008; Silva et al. 2015; Takada et al. 2001). These biopesticides exhibit high target specificity, long persistence, environmental friendliness, and a reduced risk of resistance development in *S. frugiperda* (Prasanna et al. 2018). However, the risk assessment of these biopesticides on parasitoids is rarely documented, with only a few reports conducted so far. It is therefore important to evaluate potential hazards of these biopesticides to parasitoids.

In our study, we aimed to assess the sensitivity of five parasitoid species of *S. frugiperda* (*Trichogramma mwanzai, Trichogrammatoidea lutea*, *T. remus*, *T. dendrolimi*, and *T. chilonis*) to potential replacement pesticides including chlorantraniliprole, lufenuron, chlorantraniliprole and lufenuron in a 3:1 ratio (C:L = 3:1), emamectin benzoate, spinetoram, Bt and MabrNPV. By conducting adult parasitoid toxicity bioassays, we evaluated the influence of these products on parasitoid mortality, lifespan, emergence and parasitism rates at various growth stages. These experiments were performed in a controlled laboratory setting and research findings will contribute to the development of precise insecticide dosage guidelines and provide scientifically informed recommendations for the safe and effective application of chemical and biological agents against *S. frugiperda* in both China and Africa.

Materials and methods

Host

For the purpose of this study, we collected the *S. frugiperda* egg masses from maize fields located in Qianxinan prefecture, Guizhou (25° 5′ 38″ N, 104° 54′ 44″ E) in June 2019. To ensure a consistent population for our research, we maintained the *S. frugiperda* population at

the Institute of Biological Control laboratory, affiliated with Jilin Agricultural University in Changchun, China. After hatching, larvae were reared in 10.5 cm diameter plastic Petri dishes until they reached the third-instar developmental stage. Subsequently, the larvae were transferred to individual compartments of 6-well plates and reared until they reached the pupal stage (Sun et al. 2020). To ensure proper egg collection, we held the emerged adults in a net cage (35.0 cm × 35.0 cm × 35.0 cm, length × width × height) under controlled laboratory conditions (Hou et al. 2022). The temperature was maintained at 26 °C, relative humidity oscillated between 65–75%, and a photoperiod regimen of 14-10 (L:D) was maintained.

Parasitoids

Trichogramma mwanzai, *Trichogrammatoidea lutea*, and *Telenomus remus* are three indigenous African parasitoids that have been extensively studied and proven to be effective in the management of *S. frugiperda* (Sun et al. 2021; Chen et al. 2023). Due to their established effectiveness in the region, these species were sourced from the China-aid Zambia Agricultural Technology Demonstration Centre (15° 21′ 30″ S, 28° 27′ 27″ E), located in Lusaka, Zambia, during the summer of 2019. To establish a small-scale culture, parasitized *S. frugiperda* egg masses were carefully placed within a glass tube (10 cm × 1.5 cm, length × diameter) under controlled laboratory conditions. The culture was maintained at a temperature of 26 °C, with a relative humidity of 65–75%, and a 14-10 (L:D) photoperiod, facilitating optimal growth and development. The population of *T. mwanzai* and *T. lutea* were cultivated using rice moth [*Corcyra cephalonica* (Stainton)] eggs, whereas *T. remus* was cultivated using *S. frugiperda* eggs. These populations were intentionally maintained under the aforementioned laboratory conditions to simulate natural conditions and facilitate growth and development. To ensure genetic diversity and maintain the vitality of *T. mwanzai* and *T. lutea* populations, wild *S. frugiperda* eggs were intentionally introduced every five generations. This practice helps to preserve the adaptive traits and enhance the effectiveness of these parasitoids in pest management.

In this study, two indigenous Chinese *Trichogramma* species *T. dendrolimi* and *T. chilonis*, were used to determine their potential for controlling *S. frugiperda*, considering their previous excellent control efficacy on the eggs of many Lepidoptera insects. During 2011, *T. dendrolimi* and *T. chilonis* populations were obtained from parasitized eggs of *Chilo suppressalis* (Walker) (Lepidoptera: Crambidae) in Changchun (Jilin Province), located in northeastern China (43.89° N, 125.32° E). Furthermore, the parasitoid populations were successfully cultivated on *C. cephalonica* eggs and reared under controlled laboratory conditions, following the same protocols as described above.

Pesticides

During toxicity testing, a selection of seven commercial pest control product formulations were carefully selected. Information on the composition and properties of each pest control product provides is provided in Additional file 1: Table S1. These pesticides were obtained through direct purchase or donations from various manufacturers in China. The pest control products were tested at the recommended field dosages, and their residual concentrations were measured.

Lethal effect of pesticides on parasitoid adults

The objective of this bioassay, following protocols described by Sugiyama et al. (2011), was to evaluate the residual contact toxicity of pesticides on combined male and female parasitoids. This study aimed to assess the toxic effects of pesticides on these parasitoids, particularly the residual effects after their exposure to the pesticides. According to preliminary test results, all the pesticides were treated with five different concentrations as follows: the concentration of chlorantraniliprole was 10, 20, 30, 40, 50 mL/L; lufenuron concentrations were 5×10^{-1}, 1, 2, 4, and 8 mL/L. Emamectin benzoate tested at concentrations of 2×10^{-1}, 4×10^{-1}, 6×10^{-1}, 8×10^{-1}, and 10 mL/L. Spinetoram concentrations were 5×10^{-2}, 2×10^{-1}, 3.5×10^{-1}, 5×10^{-1}, and 6.5×10^{-1} mL/L; and the compound C:L = 3:1 was tested at concentrations of 8×10^{-1}, 1, 1.2, 1.4, 1.6 mL/L. To ensure a comprehensive evaluation across various production systems, four synthetic pesticides (chlorantraniliprole, lufenuron, emamectin benzoate, spinetoram, and the compound C:L = 3:1) and two biopesticides (*Mamestra brassicae* Nuclear Polyhedrosis Virus and *Bacillus thuringiensis*) were carefully selected for this study. This selection allows for the assessment of the efficacy and potential differences between synthetic and bio-based pest control methods. The pesticides and biopesticides were prepared by mixing with distilled water at their respective recommended dosage (Additional file 1: Table S1). The test groups were treated with these prepared solutions, while the control group received distilled water without any pest control products. To ensure unbiased and reliable results, a randomized design with 20 replicates was employed for this study. This design allows for the random allocation of treatments, reducing the potential for bias and allowing for a robust statistical analysis. To ensure a uniform distribution of the pesticides, each pesticide was evenly applied into a glass tube (2 cm in diameter, 10 cm in length). The tubes were then gently

rotated to ensure a thorough coating of the pesticides on the inner surfaces, as recommended by Snodgrass (1996). After the pesticide-coated tubes had dried, groups of twenty newly emerged parasitoid adults (10 males and 10 females, all within 6 h of emergence) were introduced into each tube. To maintain experimental conditions, the tubes were carefully sealed with nylon gauze, allowing for ventilation while preventing the escape of the parasitoid adults. Subsequently, the sealed tubes were incubated under the specified laboratory conditions of temperature, humidity, and lighting. After 1 h, the adult parasitoids were carefully transferred into separate pesticide-free glass tubes to measure their acute toxicity (Liu et al. 2016). To ensure their nutritional needs, a 20% honey solution was supplied as a food source. After 24 h, the number of surviving and dead adults was observed and determined by using a binocular microscope. Survival assessments were conducted daily until the mortality of all adults. Dead adults were classified based on specific criteria as described by Wang et al. (2019).

Indirect effects of pest control products on parasitoid juveniles

To assess the impact of the five synthetic pesticides and two biopesticides on the emergence and developmental stages of five parasitoids, studies were conducted 1, 3, and 5 days post-parasitism under laboratory conditions of 26 ± 1 °C, RH 70 ± 5% and a photoperiod of 14:10 (L:D) h. By following this exposure schedule, the study encompassed the 1-day-old egg stage, 3-day-old larvae stage, and 5-day-old pupae stage of the parasitoids (Cônsoli et al. 2001; Ruberson and Kring 1993). The preparation of immature stages for each parasitoid species followed the methodology of Sun et al. (2021). To assess the effect of the pesticides on the emergence and immature developmental stages of parasitoids, parasitized egg masses containing 100 to 120 eggs were briefly submerged in vials containing 100 mL of pesticide-infused solution at field-recommended rates (Additional file 1: Table S1) for a duration of 5 seconds, followed by air-drying in a ventilated enclosure (Prabhaker et al. 2007). As a control group, distilled water was included to provide a baseline comparison for the effects observed in the pesticide-treated groups. After the drying process, treated egg masses were individually transferred into transparent glass tubes, and five newly emerged (less than 8 h old), mated, female parasitoids were introduced. These tubes were maintained under the previously described environmental conditions of 26 ± 1 °C, RH 70 ± 5%, and a photoperiod of 14:10 (L:D) h. *S. frugiperda* egg masses were carefully examined daily to monitor their development and assess the presence of parasitism. Unparasitized *S. frugiperda* larvae were meticulously extracted using a soft brush under a binocular microscope. This procedure aimed to remove unparasitized larvae (to prevent larvae from feeding on parasitic eggs after hatching as it can significantly impact the success of parasitism) and using binocular microscope to ensure accurate identification. Systematic observations were conducted to assess the level of parasitism, emergence rate, and the sex ratio of the parasitoids. To ensure the reliability and robustness of the results, each treatment involving parasitoids and pest control products was replicated five times.

Statistical analysis

Probit analysis

Toxicity data obtained from the experiment were analyzed using SPSS version 20 software (SPSS Inc., Chicago, IL, USA). A probit analysis was performed to ascertain the median lethal concentration (LC_{50}) of the seven pest control products, which would result in the mortality of 50% of the sample population of *T. mwanzai*, *T. lutea*, *T. remus*, *T. dendrolimi*, and *T. chilonis* within 1 h. The safety of the synthetic pesticides and biopesticides for parasitoids was assessed quantitatively through risk quotients (Wang et al. 2013), a crucial metric for evaluating beneficiary risk in field conditions. Risk quotients were calculated by comparing the exposure concentration of the pesticide to the field-recommended dose, providing insights into the potential risk to beneficial organisms such as parasitoids (Stark et al. 1995). Risk quotient = field recommended dose (g a.i. ha^{-1})/Lethal Concentration 50% (LC_{50}) of beneficial insect (mg a.i. L^{-1}). Pesticides with risk quotient (RQ) values below 50, calculated based on the comparison of exposure concentration to the field-recommended dose, are considered safe (Desneux et al. 2004). Pesticides with RQ values ranging from 50 to 2500 are regarded as slightly to moderately toxic, while those surpassing 2500 are categorized as highly toxic (Preetha et al. 2010).

Bioassay analysis

For each bioassay, data sets including percent mortality, number of parasitized eggs, percent emergence of individual developmental stages, and adult longevity, were analyzed using a linear model. The analysis considered parasitoid species (5 levels) and pesticides species (7 levels) as factors. Tukey's honest significant difference (HSD) test was employed at a significance level of $P < 0.05$ to determine differences between the levels of parasitoid species and pesticide species. To address variance heterogeneity, a log transformation was applied to the number of parasitized eggs and longevity, while percent mortality and percent emergence was arcsine square-root transformation prior to conducting a GLM analysis.

Subsequently, the transformed data were subjected to the Shapiro–Wilk test.

Results

Toxicity assessment of egg parasitoid adults and various developmental stages at field concentrations

The seven pest control products, at their corresponding field-recommended concentrations, had notably detrimental effects on the survival rates of the examined adult insects ($F_{7, 118}$= 123.06, P < 0.0001; *T. lutea*: $F_{7, 135}$= 23.00, P < 0.0001; *T. dendrolimi*: $F_{7, 119}$= 76.63, P < 0.0001; *T. chilonis*: $F_{7, 133}$= 133.81, P < 0.0001; *T. remus*: $F_{7, 123}$= 12.16, P < 0.0001) (Fig. 1). Spinetoram caused over 92.6% mortality in *T. mwanzai*, *T. dendrolimi*, and *T. chilonis* after 1 day, while a lower mortality rate was observed in *T. lutea* (64.9%) and *T. remus* (49.2%). Chlorantraniliprole, chl.+luf. (3:1), MabrNPV, and Bt showed mortality rates below 25% in all tested adult parasitoids ($F_{4, 96}$= 22.45, P < 0.0001). Additionally, mortality caused by lunefuron applied alone ranged from 1.8 to 9.8%. After 3 days, Emamectin benzoate resulted in over 90% mortality in adults of four parasitoid species,*T. remus* being the exception. In contrast, chlorantraniliprole, chl.+luf. (3:1), MabrNPV and Bt showed less than 50% mortality in all tested adult parasitoids, except *T. mwanzai* on MabrNPV (68.6%). After 10 days, the cumulative mortality rate (66.0%) of *T. lutea* treated with chl.+luf. (3:1) was significantly lower than the control group (79.7%) (P < 0.05). In comparison to other egg parasitoids, *T. remus* exhibited significantly lower cumulative mortality when exposed to all seven pest control products.

Dose–response analyses of five parasitoid adults exposed to pesticides

The seven pest control products were categorized into two groups based on their respective acute toxicity levels and risk quotient values (refer to Additional file 1: Table S1). The five synthetic pesticides displayed a range of acute toxicities towards adult parasitoids, with varying levels of toxicity. The biopesticides MabrNPV and Bt, when used at field-recommended concentrations, resulted in mortality rates of parasitoid adults at 21.5% and 13.6%, respectively, making it challenging to estimate the LC_{50} values for MabrNPV and Bt in adults (refer to Table 1). Among the tested pesticides, chlorantraniliprole exhibited the lowest toxicity, with LC_{50} values ranging from 246.77 to 1.93×10^6. It was followed by lufenuron, with LC_{50} values ranging from 19.7 to 1.6×10^4 mg a.i. L^{-1}, and the combination of chl.+luf. (3:1) (2.07–9.02 mg a.i. L^{-1}). Risk quotients were calculated based on toxicity data and subsequently categorized. MabrNPV, *B. thuringiensis*, chlorantraniliprole, lufenuron, and chl.+luf. (3:1) were all considered safe for all assessed parasitoid adults, with risk quotients below 5 (ranging from 0.00016 to 4.83). Lufenuron was determined to be safe for adults of *T. mwanzai*, *T. lutea*, *T. remus*, and *T. dendrolimi* (risk quotients ranging from 0.05 to 84.66). However, emamectin benzoate (risk quotient ranging from 375 to 1090.91) and spinetoram (risk quotient ranging from 95.2 to 1471.74) were deemed slightly to moderately toxic (according to Table 1).

Longevity of parasitoids subjected to seven pest control products at field-recommended concentrations

Upon exposure to the five pesticides and two biopesticides, significant impacts on the longevity of all five parasitoid species were observed (*T. mwanzai*: $F_{7, 118}$= 20.70, P < 0.0001; *T. lutea*: $F_{7, 135}$= 20.55, P < 0.0001; *T. dendrolimi*: $F_{7, 119}$= 27.82, P < 0.0001; *T. chilonis*: $F_{7, 133}$= 36.53, P < 0.0001; *T. remus*: $F_{7, 123}$= 7.47, P < 0.0001) (Fig. 2). The longevities of *T. dendrolimi*, *T. chilonis* and *T. remus* adults were significant decreased compared to the control group across all treatments, with a varying extent of decrease for each species. Spinetoram induced the shortest longevity for *T. mwanzai* (1.8 d), *T. dendrolimi* (1.0 d), and *T. chilonis* (1.8 d) in terms of days, followed by emamectin benzoate and lufenuron. MabrNPV and *B. thuringiensis* notably reduced the longevity of all five evaluated parasitoids. Conversely, chlorantraniliprole, lufenuron and chl.+luf. (3:1) displayed no significant influence on the adult longevity of *T. mwanzai* and *T. lutea* when subjected to a 1-h exposure, with longevity rates similar to the control group. Interestingly, *T. lutea* adults experienced an extended longevity of 13.2 days on the chl.+luf. (3:1) residue treatment, compared to 11.5 days in the control group (Fig. 2).

Emergence rates of parasitoids after exposure to the pest control products at field-recommended concentrations

Significant differences were observed in the effect of five pesticides and two pesticides on the emergence rate of all egg parasitoid species across three developmental stages (Table 2). The following emergence rates were recorded: *T. mwanzai*: $F_{7, 37}$ = 9.05–33.56, P < 0.0001; *T. lutea*: egg: $F_{7, 32}$ = 20.00, P < 0.0001, larvae: $F_{7, 37}$ = 2.00, P = 0.0819, pupae: $F_{7, 39}$ = 5.203, P = 0.0003; *T. dendrolimi*: $F_{7, 32}$ = 8.19–41.43, P < 0.0001; *T. chilonis*: $F_{7, 32}$ = 9.85–13.35, P < 0.0001; *T. remus*: egg: $F_{7, 27}$ = 3.37, P = 0.0103, larvae: $F_{7, 33}$ = 1.08, P = 0.4013, pupae: $F_{7, 26}$ = 1.91, P = 0.1092. The treatment with lufenuron, emamectin benzoate, spinetoram, and chl.+luf. (3:1) resulted in increased emergence in four of five studied species, corresponding to the increased age stage. However, *T. remus* exhibited a different response. Conversely, post-treatment with MabrNPV and *B.thuringiensis* resulted in a decline in

Table 1 Acute toxicity (1-h LC_{50}) and risk analysis of seven pest control products on parasitoid adults under laboratory conditions (26 ± 1 °C, 70 ± 5% RH., and L14: D10 of photoperiod)

Parasitoids	Pesticides	Regression equation	R	LC_{50} (95% FI) (mL/L)	χ^2	Risk quotient[a]	Category[b]
T. mwanzai	Chlorantraniliprole	$-4.944+2.264x$	0.979	246.77	0.235	1.216	1
	Lufenuron	$0.759+1.288x$	0.863	19.70 (7.87-7105.93)	9.591	40.617	1
	Emamectin benzoate	$-1.253+4.154x$	0.935	0.32 (0.29–0.35)	7.588	375.000	2
	Spinetoram	$-5.066+2.702x$	0.988	0.75 (0.60–1.22)	8.886	893.000	2
	Chl. + Luf. (3:1)	$-2.608+3.223x$	0.9923	2.29 (1.87–3.50)	0.586	4.370	1
	MabrNPV	–	–	–	–	0.000	1
	Bt	–	–	–	–	0.000	1
T. lutea	Chlorantraniliprole	$-0.557+0.981x$	0.998	4610	0.006	0.0651	1
	Lufenuron	$0.512+0.971x$	0.985	421	0.204	1.900	1
	Emamectin benzoate	$0.487+3.071x$	0.896	0.29 (0.08–0.44)	71.086	407.000	2
	Spinetoram	$-3.438+4.138x$	0.7816	1.09 (0.70–3.77)	3.857	676.000	2
	Chl. + Luf. (3:1)	$-3.352+3.488x$	0.985	2.48 (1.97–4.04)	0.963	4.030	1
	MabrNPV	–	–	–	–	0.000	1
	Bt	–	–	–	–	0.000	1
T.dendrolim	Chlorantraniliprole	$3.197+0.218x$	0.988	1.93×10^{6}	0.021	0.00016	1
	Lufenuron	$2.349+0.427x$	0.977	1.60×10^{4} ($162-3.23\times10^{100}$)	0.158	0.050	1
	Emamectin benzoate	$3.276+1.681x$	0.900	0.11 (0.007–0.21)	18.208	1090.910	2
	Spinetoram	$1.144+2.323x$	0.920	0.46 (0.35–0.61)	22.142	1471.740	2
	Chl. + Luf. (3:1)	$-9.011+6.052x$	0.976	2.07 (1.77–2.77)	4.259	4.830	1
	MabrNPV	–	–	–	–	0.000	1
	Bt	–	–	–	–	0.000	1
T. chilonis	Chlorantraniliprole	$2.835+0.402x$	0.960	2460	0.289	0.120	1
	Lufenuron	$-1.325+2.126x$	0.998	9.45	0.195	84.660	2
	Emamectin benzoate	$1.329+2.626x$	0.917	0.25 (0.12–0.35)	23.710	480.000	2
	Spinetoram	$2.207+1.325x$	0.939	1.27 (0.60–295)	14.876	533.070	2
	Chl. + Luf. (3:1)	$-1.769+2.291x$	0.905	9.02	0.346	1.110	1
	MabrNPV	–	–	–	–	0.000	1
	Bt	–	–	–	–	0.000	1
T. remus	Chlorantraniliprole	$-0.54+0.872x$	0.879	2.25×10^{4}	0.049	0.013	1
	Lufenuron	$0.312+0.920x$	0.990	1250	0.019	0.640	1
	Emamectin benzoate	$1.999+2.590x$	0.952	0.14 (0.10–0.18)	7.224	857.14	2
	Spinetoram	$2.170+0.992x$	0.944	7.11 (2.12–443)	3.140	95.200	2
	Chl. + Luf. (3:1)	$-3.381+2.916x$	0.996	7.48	0.008	1.340	1
	MabrNPV	–	–	–	–	0.000	1
	Bt	–	–	–	–	0.000	1

[a] Risk quotient = field-recommended concentration (g a.i. ha^{-1})/LC50 of each parasitoid (mg a.i. L^{-1})

[b] Categories follow the IOBC classification44: 1 = safe; 2 = slightly to moderately toxic; 3 = dangerously toxic

emergence rates for three species (*T. mwanzai*, *T. dendrolimi*, and *T. chilonis*) as the age stage increased.

Upon treatment at the egg stage, the highest emergence rates were observed for *T. mwanzai*, *T. lutea*, and *T. remus* with the chlorantraniliprole treatment (94.3%, 99.6%, and 99.6% respectively). This contrasts with *T. dendrolimi* and *T. chilonis* which exhibited the highest emergence rates in the MabrNPV and Bt treatments (98.7% and 100.0% respectively). However, the emergence rates considerably diminished (53.8–54.7%) for emamectin benzoate and spinetoram treatments, except for *T. remus*. During the larval stage treatment, *T. mwanzai*, *T. lutea*, *T. dendrolimi*, and *T. chilonis* showed increased emergence rates with emamectin benzoate, spinetoram, and chl. + luf. treatments. In contrast, at the pupal stage and immature stages demonstrated higher emergence rates with lufenuron treatment. Remarkably, *T. remus* consistently exhibited an emergence rate exceeding 96.0% across all treatments and developmental stages. There were significant disparities among the

Table 2 Percent emergence (mean ± SE) of parasitoids after exposure to seven pest control products at field-recommended concentrations during the egg, larvae, and pupae stages

Parasitoids	Pesticides	Emergence rates at different developmental stages		
		Egg	Larvae	Pupae
T. mwanzai	Chlorantraniliprole	94.3 ± 3.2a A	84.6 ± 4.1bc B	90.8 ± 3.8a AB
	Lufenuron	70.4 ± 5.6bc B	68.9 ± 4.8d B	94.6 ± 1.6a A
	Emamectin benzoate	54.7 ± 6.9d B	66.6 ± 2.1d AB	73.9 ± 4.3b A
	Spinetoram	53.8 ± 7.2d A	52.6 ± 4.4e A	51.7 ± 3.3c A
	Chl. + Luf. (3:1)	65.9 ± 3.5c B	82.4 ± 3.3bc A	94.7 ± 3.7a A
	MabrNPV	77.0 ± 6.1b A	76.0 ± 3.4 cd A	32.4 ± 1.3d B
	Bt	84.3 ± 2.9b A	76.0 ± 3.0 cd B	76.6 ± 1.5b B
	Untreated control	97.3 ± 1.8a A	93.4 ± 3.3a A	96.6 ± 2.2a A
T. lutea	Chlorantraniliprole	99.6 ± 0.4a A	88.2 ± 9.7abc AB	93.2 ± 3.4ab B
	Lufenuron	79.1 ± 5.3c B	80.3 ± 4.3c B	95.5 ± 2.3ab A
	Emamectin benzoate	53.0 ± 2.4e B	89.6 ± 5.7bc A	83.9 ± 5.3c A
	Spinetoram	65.4 ± 6.7d B	77.6 ± 2.9c A	95.1 ± 4.9ab A
	Chl. + Luf. (3:1)	92.4 ± 2.2b A	91.7 ± 2.9b A	88.9 ± 4.4bc A
	MabrNPV	96.8 ± 2.4ab A	97.9 ± 1.1ab A	89.7 ± 6.9bc A
	Bt	97.3 ± 1.8ab A	95.6 ± 3.4b A	98.2 ± 1.1a A
	Untreated control	98.9 ± 0.7a A	98.6 ± 0.9a A	99.3 ± 0.7a A
T. dendrolimi	Chlorantraniliprole	97.6 ± 1.4a A	98.4 ± 0.6ab A	94.8 ± 3.4b A
	Lufenuron	78.5 ± 5.2b B	62.3 ± 7.7d B	93.9 ± 2.6b A
	Emamectin benzoate	57.0 ± 4.3c B	92.7 ± 1.6c A	92.7 ± 1.5b A
	Spinetoram	42.8 ± 5.6d B	70.0 ± 2.9d A	72.5 ± 6.7c A
	Chl. + Luf. (3:1)	79.6 ± 4.0b B	97.5 ± 0.5b A	99.8 ± 0.2a A
	MabrNPV	98.7 ± 1.1a A	95.1 ± 1.1b B	98.4 ± 1.6ab A
	Bt	98.2 ± 0.6a A	92.3 ± 2.3c B	95.2 ± 1.3b B
	Untreated control	99.5 ± 0.3a A	99.6 ± 0.4a A	98.7 ± 0.5a A
T. chilonis	Chlorantraniliprole	95.4 ± 2.3b A	100.0 ± 0.0a A	96.3 ± 2.1b A
	Lufenuron	81.8 ± 5.1c B	88.2 ± 2.4c AB	95.3 ± 2.1b A
	Emamectin benzoate	87.2 ± 1.9c B	97.7 ± 1.5b A	95.8 ± 2.1b A
	Spinetoram	64.7 ± 5.1d B	76.1 ± 2.1d AB	84.4 ± 4.4c A
	Chl. + Luf. (3:1)	84.9 ± 5.9c B	97.0 ± 2.2b A	100.0 ± 0.0a A
	MabrNPV	97.9 ± 1.3b A	97.6 ± 1.0b A	74.7 ± 5.4d B
	Bt	100.0 ± 0.0a A	91.3 ± 4.2c B	99.3 ± 0.7a A
	Untreated control	100.0 ± 0.0a A	100.0 ± 0.0a A	98.4 ± 1.6ab A
T. remus	Chlorantraniliprole	99.6 ± 0.2a A	96.5 ± 0.8b B	97.3 ± 0.5a B
	Lufenuron	96.2 ± 1.1c A	97.6 ± 0.8a A	97.8 ± 0.8a A
	Emamectin benzoate	96.9 ± 0.4c A	96.2 ± 1.1b A	96.4 ± 3.4a A
	Spinetoram	97.5 ± 0.6c A	96.4 ± 0.9b A	96.0 ± 2.3a A
	Chl. + Luf. (3:1)	98.8 ± 0.5ab A	97.2 ± 1.2ab A	97.4 ± 1.2a A
	MabrNPV	97.1 ± 0.6c A	97.5 ± 0.5b A	97.8 ± 0.6a A
	Bt	97.7 ± 0.9bc A	97.1 ± 0.7b A	97.7 ± 0.8a A
	Untreated control	99.3 ± 0.4a A	98.9 ± 0.7a A	98.0 ± 0.6a A

Values followed by different lower-case letters within the same column and capital letters within the same row indicate significant differences at $P < 0.05$ level by Tukey's HSD test

seven pest control products in the egg and larval stages, but no impact was observed at the pupal stage. The exception was for chlorantraniliprole, where no significant difference was observed in the emergence rate of *T. remus* when treated with any of the five pesticides or two biopesticides during the three developmental stages.

Significant effects of the seven pest control product residues on parasitism were observed for each parasitoid

Table 3 *S. frugiperda* parasitism (mean ± SE) by parasitoids after exposure to different pest control products

Pesticide	Number of eggs parasitized (± SE)				
	T. mwanzai	*T. lutea*	*T. dendrolimi*	*T. chilonis*	*T. remus*
Chlorantraniliprole	7.3 ± 1.05aD	15.2 ± 0.66aD	44.6 ± 3.39aB	28.2 ± 2.74aC	85.1 ± 5.24abA
Lufenuron	6.6 ± 0.73aD	15.7 ± 1.67aC	40.8 ± 4.85aB	22.0 ± 3.30bC	69.2 ± 2.52bA
Emamectin benzoate	2.1 ± 0.46bC	9.3 ± 1.04dBC	18.6 ± 3.66bB	7.4 ± 1.56cC	43.8 ± 5.56cA
Spinetoram	2.0 ± 0.52bB	6.9 ± 1.15dB	8.8 ± 1.79bB	7.5 ± 1.09cB	24.5 ± 0.50dA
Chl. + Luf. (3:1)	6.7 ± 0.57aD	14.3 ± 1.21abC	46.5 ± 2.32aB	19.4 ± 1.41bC	79.8 ± 3.62abA
MabrNPV	6.4 ± 1.02aD	10.4 ± 0.99cCD	46.2 ± 3.29aB	17.4 ± 2.24bC	73.3 ± 2.14abA
Bt	5.6 ± 0.50aD	11.0 ± 1.41bcCD	45.4 ± 2.94aB	16.4 ± 2.00bC	76.8 ± 3.75abA
Untreated control	7.9 ± 0.88aE	16.3 ± 0.80aD	47.8 ± 3.46aB	29.0 ± 0.45aC	88.8 ± 1.53aA

Values followed by different lower-case letters within the same column and capital letters within the same row indicate significant differences at $P < 0.05$ level by Tukey's HSD test

species 24 h post-treatment (*T. mwanzai*: $F_{7, 63} = 5.418$, $P < 0.0001$; *T. lutea*: $F_{7, 84} = 8.871$, $P < 0.0001$; *T. dendrolimi*: $F_{7, 72} = 5.12$, $P < 0.0001$; *T. chilonis*: $F_{7, 75} = 5.12$, $P < 0.0001$; *T. remus*: $F_{7, 43} = 5.12$, $P < 0.0001$) (Table 3). Once again, the exception was for chlorantraniliprole where all parasitoids showed a reduced number of parasitized eggs compared to the control. Emamectin benzoate (2.1, 9.3, 18.6, 7.4, 43.8 respectively) and spinetoram (2.0, 6.9, 8.8, 7.5, 24.5 respectively) resulted in significantly fewer parasitized eggs of *T. mwanzai*, *T. lutea*, *T. dendrolimi*, *T. chilonis* and *T. remus* compared to the control (7.9, 16.3, 47.8, 29.0, 88.8 respectively). Furthermore, *T. lutea* and *T. chilonis* exhibited reduced parasitism of host eggs with MabrNPV (10.4, 11.0 respectively) and Bt (17.4, 16.4 respectively) compared to the control (16.3, 29.0 respectively). In contrast, the parasitism of *T. mwanzai*, *T. dendrolimi*, and *T. remus* showed no significant difference compared to the control (MabrNPV: $F_{4, 48} = 95.609$, $P < 0.0001$; Bt: $F_{4, 49} = 110.827$, $P < 0.0001$).

Discussion

The rapid increase in *S. frugiperda* populations has driven widespread pesticide use in China (Kenis et al. 2023). However, limited research exists on their impact on native insect communities, particularly on beneficial parasitoid wasps. Despite their crucial role in pest control, parasitoid wasps often show greater susceptibility to pesticides compared to their host organisms. Assessments of pesticide effects on wasps should therefore consider both acute and residual toxicity (Desneux et al. 2007) and our research revealed significant variations in the toxicity and residual risks posed by five synthetic pesticides and two biopesticides on adult development of five egg parasitoid species at their recommended field concentrations. Of the 9 insecticides tested, spinetoram and emamectin benzoate were harmful for the mortality, emergence (except *T. remus*), lifespan, and reproduction of adults in all five tested parasitoid species, followed by two biopesticides, MabrNPV and Bt. Insect growth regulators (IGRs) including chlorantraniliprole, lufenuron and chl. + luf. had a minimal impact on them, thus being considered safe. Previous studies have indicated that *T. chilonis* adults exhibited a reduced response (LC50 of 1.953 mg a.i. l−1) with no death observed following chlorantraniliprole application at the recommended field dosage (Preetha et al. 2009). Additionally, similar results have been reported in *T. pretiosum*, *Diadegma semiclausum* (Helen) (Hymenoptera: Ichneumonidae), *Dolichogenidea tasmanica* (Cameron) (Hymenoptera: Braconidae), and *T. remus* at exposure levels relevant to their habitat, that did not impact the searching behavior of females (Brugger et al. 2010; Li et al. 2021). The risk quotient analysis of lufenuron indicated that it is safe to adults of five parasitoid species, with risk quotients all below 50. In our study, it does have an impact on the emergence of *T. mwanzai*, *T. lutea*, *T. dendrolimi*, and *T. chilonis* (Table 2), which is consistent with its effect on the emergence of *Trichogramma galloi* Zucchi adults from eggs that were treated and given to parasitoids (Cônsoli et al. 2001). Similar results were reported when adults of *Trichogramma japonicum* Ashmead and *T. dendrolimi* were exposed to lufenuron residues (Yang et al. 2020). However, other related studies indicated that IGRs such as pyriproxyfen was harmful for development of early juvenile stages of *E. formosa* (Wang et al. 2019). The detrimental effects on immature stages could be attributed to the suppression of chitin synthesis and disruption of exoskeleton formation after molting (Devillers 2013; Dhadialla et al. 1998). IGRs often have strong target specificity and high efficiency, while causing minimal harm to pollinators and natural enemies (Bassi et al. 2009, Stevens et al. 2022). Studies have indicated that chlorantraniliprole and lufenuron, together with benzoylphenylurea compounds like diflubenzuron and flufenoxuron, exhibit limited negative

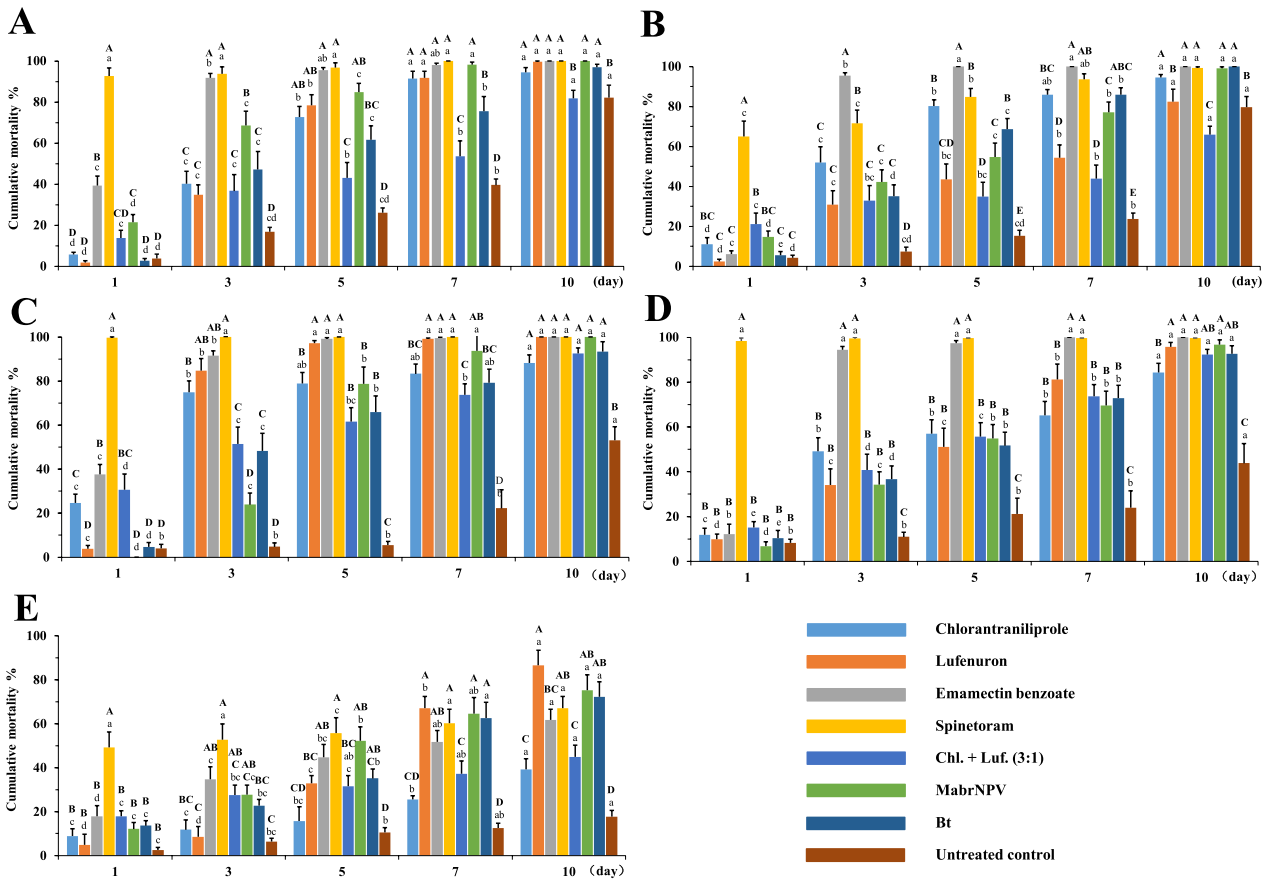

Fig. 1 Cumulative mortality (mean ± SE) of *T. mwanzai* (**A**), *T. lutea* (**B**), *T. dendrolimi* (**C**), *T. chilonis* (**D**), and *T. remus* (**E**) treated with seven pest control products at concentrations recommended for field application. Data were means ± SE. Different upper-case in the same day indicate significant difference at $P < 0.05$ level by Tukey's HSD test. Different lower-case in the same insecticide indicate significant difference at $P < 0.05$ level by Tukey's HSD test

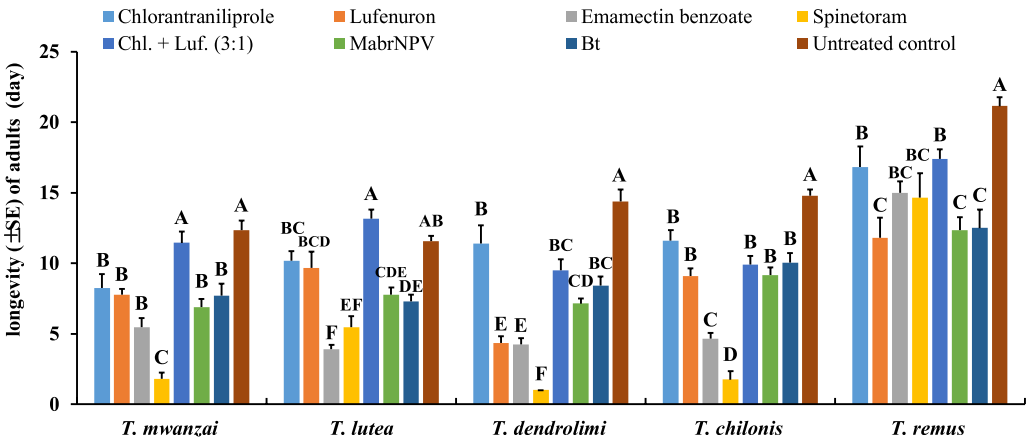

Fig. 2 Longevity (mean ± SE) of parasitoids treated with seven pest control products at concentrations recommended for field application. Data were means ± SE. Different upper-case in the same parasitoid indicate significant difference at $P < 0.05$ level by Tukey's HSD test

effects on egg parasitoids such as *Trichogramma dendrolimi* (Takada et al. 2001) and *Trichogramma cacoeciae* (Hassan et al. 2009). Our study revealed that a combined chlorantraniliprole-lufenuron (C:L=3:1) was safe for adults of five parasitoid species, resulting in a low mortality rate of 13.8–30.7% during a 1-h exposure (Fig. 1). Furthermore, no significant impact on the parasitism of *T. mwanzai*, *T. lutea*, *T. dendrolimi*, and *T. remus* adults was observed (Table 3). These findings match with the study by Kousika et al. (2015) which found that a formula containing higher amounts of chlorantraniliprole 4.3% + abamectin 1.7% SC had a minimal impact on adult emergence and parasitism of *T. pretiosum (but* smaller amounts of this formula were determined to be safe for *T. pretiosum).* Furthermore, the effects on other predator groups were more pronounced. For instance, studies have shown that this pesticide is safe for both larvae and adults of *Harmonia axyridis* (Pallas), *Chrysoperla sinica* (Tjeder), and *Snelleniua manilae* (Ashmead) (Liu et al. 2016). However, it is important to note that our study does not address the potential long-term effects of prolonged exposure of parasitoids to tested pesticides. Although IGRs are relatively safe for parasitic wasps, whether they are used individually or in combination, it is still important to carefully assess their impact on non-target organisms before their adoption in IPM. Risk assessments have indicated that both two biopesticides are relatively safe for the adults of the five parasitoid species, but it has been observed reduced lifespan after treatment with the biopesticides and reproductive capacity of *T. lutea* and *T. chilonis* within 24 h was reduced. While this sustained negative impact may not affect their efficacy in controlling pests, further research is needed to determine if it has any effect on the reproductive capacity and behavior of their offspring.

Our data indicates that the dry residues of spinetoram caused significant mortality rates above 90% for adults of *T. mwanzai*, *T. dendrolimi*, and *T. chilonis* at field-recommended levels. It is consistent with the finding of Sattar et al. (2011), who reported 98.8% residual toxicity to adult *T. pretiosum* and 100% mortality in *T. chilonis* adults treated with spinosad (spinosyn) (Sattar et al. 2011). Other research has also indicated that spinetoram demonstrated a sustained efficacy and direct mortality on *Helicoverpa armigera* Hubner (Abbas et al. 2015). Additionally, our results support the findings of Hernandez et al. (2011), and Visnupriya and Muthukrishnan (2016), who observed the adverse effects of spinetoram on parasitoids at multiple growth stages. However, they did not find increased toxicity compared to other solutions and untreated samples. Similarly, Liu and Zhang (2012) found that higher doses of spinosad significantly reduced the emergence of adults for *Trichogramma pretiosum* Riley and *Trichogramma brassicae* Bezdenko when applied during the older larval and pupal stages. The high mortality rate of spinetoram may be due to the strong inhibitory effects, as a neurotoxin, on neuronal cell conduction and neuromuscular activity in both adults and larvae.

Emamectin benzoate enhances neural effects by affecting neurotransmitters such as glutamate and GABA, leading larvae cease feeding immediately, causing paralysis and significant mortality within 3–4 days (Fanigliulo and Sacchetti 2008; Gu et al. 2023). This is consistent with our findings, as the main lethal effect of emamectin benzoate on adult parasitoids occurs after 3 days (Fig. 1). Moreover, emamectin benzoate was notably toxic to *T. mwanzai* and *T. lutea* eggs, while it was found to be harmless for all life stages of *T. remus* and pupae of *T. mwanzai* and *T. dendrolimi* (Table 2). Previous studies have indicated that emamectin benzoate has good control effects on the larvae of many Lepidopteran pests, such as *Cydia pomonella* (L.) (Depalo et al. 2022), and *S. frugiperda* (L.) (Liu et al. 2022b). However, this insecticidal effect is often less effective against eggs and pupae. Similar results have been reported in *Trichogramma nr. Brassicae* (Hymenoptera: Trichogrammatidae (Hewa-Kapuge et al. 2003).

To date, IPM strategies have typically involved the use of both biological and chemical agents, as this can effectively control pests while reducing economic and environmental costs (Lacey et al. 2015; Ghidiu et al. 2012). Field-recommended concentrations of biopesticides such as *Bacillus thuringiensis*, have showed relatively low cumulative mortality rate (23–48%) in adults of parasitoid species tested after 3-days treatment with Bt. According to Brunner et al. (2001), B. *thuringiensis* was found to be safe or minimal toxic at the recommended rate for *Colpoclypeus florus* (Walker), but it exhibited high toxicity towards *Trichogramma platneri* Nagarkatti. The mortality of *T. platneri* resulting from *B. thuringiensis* sprays was primarily attributed to the physical characteristics of the spray (Brunner et al. 2001). MabrNPV exhibited low toxicity towards species such as *Eriocheir sinensis* and *Procambarus clarkii*, while providing effective control against *Plutella xylostella* (L.) at the recommended dosage (Wang et al. 2022c). Similarly, our findings indicated decreased mortality rate in parasitoid adults (below 40%), coupled with higher emergence rates and parasitized counts across the three examined immature stages, among the seven pest control products tested. However, both MabrNPV and *B. thuringiensis* significantly shortened the lifespan of five parasitoid adults in comparison to the control group. Risk assessments have indicated that both biopesticides are relatively safe for parasitoid adults, although it has been observed a reduced lifespan after treatment with the biopesticides

(Fig. 2), and their reproductive capacity, as measured by *T. lutea* and *T. chilonis* within 24 h, could be reduced (Table 2). While this sustained negative impact may not affect their efficacy in controlling pests, further research is needed to determine if it has any effects on the reproductive capacity and behavior of their offspring.

In conclusion, our research suggests that extended exposure to these pesticides, even those considered safe, may give rise to subtle effects on the behavior and survival of parasitoids (Desneux et al. 2007). Meanwhile, the test of LC_{50} can help us evaluate and compare the virulence of these pesticides by understanding the optimum concentration, which could guide more precisely of their application. The short-term results obtained after exposure can provide valuable insights to guide the integration of the five parasitoids with the pest control products in an IPM system. Safeguarding generalist natural enemies in agricultural settings can significantly enhance the sustainability of these systems, particularly when paired with compatible pesticides. Furthermore, the preservation of natural enemies can contribute to the sustained effectiveness of potent pesticides. This is because certain natural enemies, such as *T. remus*, are less susceptible to pesticides and prey on resistant pests (Colmenarez et al. 2022), thereby potentially impeding the development of pesticide resistance in pest populations (Gould et al. 1991). More research is needed to determine the efficacy of these insecticides in field environments, as their efficiency may be hindered in uncontrolled environments, in order to avoid causing damage to the parasitoids. In particular, a case-by-case evaluation should be carried out prior to applying any insecticide and/or releasing these parasitoids in future studies.

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s43170-023-00205-y.

Additional file 1: Table S1. The detailed information of the six acaricides.

Author contributions

Conceptualization, PD and ND; methodology, L-SZ, PD; validation, L-SZ, and PD; formal analysis: H-RL, and PD; writing—original draft preparation: PD, H-RL, C-YL; writing—review and editing: YH, X-LH; funding acquisition: WX, L-SZ; supervision: PD. All authors have read and agreed to the published version of the manuscript.

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Declarations

Ethics approval and consent to participate

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Consent for publication

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Competing interests

The authors declared that they have no competing interest in connection with the evaluated manuscript.

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