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Comparative ecotoxicity of insecticides with different modes of action to *Osmia excavata* (Hymenoptera: Megachilidae)

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ABSTRACT

Osmia excavata is an important pollinator in commercial fruit orchards. Little information has been published about ecotoxicity to *O. excavata*, especially the larvae. To clarify the risk of commonly used insecticides with different modes of action to the larvae of *O. excavata*, six insecticides (clothianidin, acetamiprid, sulfoxaflor, lambda-cyhalothrin, chlorfenapyr and abamectin) were selected for evaluation of their acute lethal toxicity and sublethal effects. Clothianidin and abamectin were the two most toxic insecticides to the larvae of *O. excavata* with LD_{50} values of 0.007 (0.006–0.008) and 0.0004 (0.0003–0.0006) μg active ingredient (a.i.) bee^{-1}, respectively. And their ecological risks were high according to the hazard quotient values (HQ > 2500). Sulfoxaflor was identified as the only safe insecticide for *O. excavata* (HQ < 50) under field conditions. Sublethal toxicity tests showed that larval weight was significantly decreased by ingesting food treated with clothianidin, lambda-cyhalothrin and abamectin (less than the maximum field registered concentrations on fruit trees) due to interference with consumption per larva and reduction of the efficiency of conversion of ingested food. Additionally, above three insecticides significantly prolonged larval developmental duration before cocooning and decreased eclosion rate. Overall, there results suggested that clothianidin and abamectin should not be applied, especially during the flowering phase, the application frequency of lambda-cyhalothrin should be minimized for the purpose of conserving *O. excavata*. Our results provided important evidences for selecting appropriate insecticides for use in fruit orchards.

1. Introduction

Bees in the genus *Osmia* (Hymenoptera: Megachilidae) are univoltine and a typical vernal species that have been successfully used to improve pollination in commercial apple orchards in different parts of the world (Sgolastra et al., 2015). *Osmia excavata* appears in the largest number among five species of *Osmia* (*O. excavata* (Alfken), *Osmia cornifrons* (Radoszkowski), *Osmia taurus* (Smith), *Osmia jacoti* (Cockerell) and *Osmia pedicornis* (Cockerell)) in north and northwest China, and plays an important role in enhancing pollination, increasing fruit diameter and the number of seeds per fruit, and decreasing the percentage of asymmetrical fruit (Shu et al., 2002). However, the extensive use of insecticides to control pests has been identified as an essential factor in the decline of bee abundance and diversity (Lee et al., 2015; Potts et al., 2010; Sgolastra et al., 2017). Rundlöf et al. (2015) reported that *Osmia* populations were more affected by insecticides than honey bees. Arena and Sgolastra (2014) also highlighted the importance and urgency of extending risk assessments to non-Apis bees. However, most risk assessments of pesticides to date have been focused on honey bees, and little research has been conducted on the larvae of solitary bees because no validated test protocol (Sgolastra et al., 2015). Our laboratory research group has found that some commonly used insecticides have been detected in the pollen provisions from apple trees (data not yet published). Pollen grain collected by *Osmia* is one of the most important food source for larvae (Sgolastra et al., 2018), which inevitably ingest any residual pesticide in pollen and might be negatively affected. Therefore, the response of larval *Osmia* should be included when assessing the risk that insecticides pose to bee populations.

Recently, neurotoxic insecticides were widely used as foliar sprays at multiple growing periods of fruit trees, including the flowering period, because of their high efficiency in controlling pests (China Pesticide Information Network, http://www.chinapesticide.org.cn/sjzx4ywb/ind

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ex.jhtml). Clothianidin and acetamiprid, belonging to neonicotinoid insecticides, act as agonists on nicotinic acetylcholine receptors (nAChRs) that open cation channels; these insecticides have high efficiency, systemic action and long persistence (Casida and Durkin, 2013; Simon-Delso et al., 2015). Sulfoxaflor is the first commercially available insecticide from the class of sulfoxamines (Zhu et al., 2011). The mode of action is similar to that of neonicotinoids as it interacts with nAChRs in the nervous system of insects, and it shows low toxicity to mammals (Sparks et al., 2013). Pyrethroids are synthetic chemical analogs of natural pyrethrins and have been used on a range of agricultural crops (Pansa et al., 2015). Lambda-cyhalothrin, a representative pyrethroid insecticide, is often used to control several important pests. This insecticide interferes with the normal functioning of the nervous system by acting on the calcium and chloride channels and the voltage-gated sodium channels, resulting in uncoordinated behavior, hyperactivity, paralysis and eventually death (Burr and Ray, 2004; Ceuppens et al., 2015). Chlorfenapyr, a pyrrole insecticide, is a mitochondrial electron transport inhibitor that can disrupt the conversion of adenosine diphosphate (ADP) to adenosine triphosphate (ATP) in the mitochondria of cells (N'Guessan et al., 2007). Abamectin is a mixture of avermectin B_{1a} and avermectin B_{1b} belonging to a macrocyclic lactone compound (Lasota and Dybas, 1991). Abamectin can act as a neurotoxic effector of γ-aminobutyric acid neurotransmitter and has been widely used as an insecticidal and acaricidal agent (Kwon et al., 2010).

Although the potential impacts of some insecticides on adult honey bees have been demonstrated (Chen et al., 2019; Sgolastra et al., 2017), uncertainties remain about their acute toxicity and sublethal effects on the larvae of *Osmia*. Therefore, it is imperative to predict the potential risks of these insecticides to the larvae of solitary bees. Acute lethal toxicity effects based on median lethal dose (the dose of insecticides needed to cause death in half of the insect; LD_{50} value) of insecticides reflect only short-term responses of insects. However, sublethal effects may cause more harm than acute lethal toxicity effects on non-target insects from a demographic perspective (Yao et al., 2015). For example, sublethal concentrations of insecticides decreased the populations of beneficial arthropods (Desneux et al., 2007) and the whitefly predator *Serangium japonicum* (Yao et al., 2015). Thus, the objective of this research was to examine the toxic effects of commonly used insecticides in orchards on *Osmia* in the worst-case scenario and promote the conservation of *Osmia*. To achieve this objective and contribute to the understanding of these toxic effects, the acute lethal toxicity and sublethal effect of commonly used insecticides with different modes of action on the development and the food utilization of the larvae of *O. excavata* were evaluated.

2. Materials and methods

2.1. Insecticides

Technical-grade clothianidin (active ingredient (a.i.) 98%; Shandong Kexin Biochemical Co., Ltd), acetamiprid (active ingredient (a.i.) 99%; Shandong Weifang Rainbow Chemical Co., Ltd), sulfoxaflor (a.i. 95.90%, Dow AgroSciences), lambda-cyhalothrin (a.i. 96%; Shandong United Pesticide Industry Co., Ltd), chlorfenapyr (a.i. 98%; Shandong United Pesticide Industry Co., Ltd) and abamectin (a.i. 96%; Jingbo Agrochemical Technology Co., Ltd) were dissolved in acetone to acquire stock solutions of 1×10^4 μg a.i. mL^{-1} for lethal and sublethal tests.

2.2. Insects

The colonies of *O. excavata* with pollen provisions were acquired from a continuous mass-rearing program in a 6-ha apple orchard in Qixia City, Shandong Province, China. There were no other flowering crops nearby during the bee nesting period and no pesticides were applied 20 days before flowering and throughout the flowering period of apple trees.

2.3. Bioassays

The toxicity experiment was conducted using a previously designed method (Yan and Wang, 2011), with minor modifications. Briefly, seven concentrations of each insecticide and 0.2% acetone were made (Table 1). The newly hatched female larvae with pollen provisions were selected based on the cell position within the nest and provision size according to Bosch et al. (2008). Female larvae were gently removed from the top of pollen provisions with a soft brush to petri dishes (60 mm diameter) and set aside. The pollen provisions of uniform size were individually soaked in the various insecticides for 10 s, and acetone was used as a control check (CK). The volumes of insecticides were measured before and after immersion for each treatment including sixty pollen provisions (Table S1) and then pollen provisions were individually transferred to plastic tubes (2 mL) after being air-dried. The female larvae of uniform size were returned to their original positions on the pollen provisions. For the acute lethal toxicity test and sublethal toxicity test, sixty larvae per concentration treatment with three replications (that is, 180 larvae per concentration for each insecticide) were respectively reared in a darkroom under conditions of 25 ± 2 °C and 65–75% relative humidity. Black-light lamps were used when checking results.

In the acute lethal toxicity test, the mortality of *O. excavata* was observed after 48 h of treatment. Larvae were identified as dead when the individuals did not respond to mild touches using a brush (Yan and Wang, 2011). To determine the amount of insecticides consumed by each *O. excavata*, pollen provisions before the insect rearing trials and the remaining portions after 48 h of treatments were both weighed. Based on the percentage of pollen provisions consumed by *O. excavata*, we calculated the dose of insecticides consumed by *O. excavata* at each concentration (Table S2). The equation is listed as follows.

$$D = (W1 - W2) \times (V1 - V2) \times C/W1/60 \quad (1)$$

In this equation, D is the ingested insecticide dose by each *O. excavata*; W1 is the weight of pollen provisions per treatment (including sixty pollen provisions) before the insect rearing trials; W2 is the remaining weight of pollen provisions per treatment after 48 h; V1 is the volume of insecticides before immersion for each treatment; V2 is the volume of insecticides after immersion for each treatment; C is the concentration of the corresponding treatment.

In the sublethal toxicity test, the larval development and the number of eclosions were examined daily. The newly hatched larvae bodies and larvae bodies after 14 d of treatments were weighed. Pollen provisions per treatment before the insect rearing trials and the remaining portions of pollen provisions after 14 d of treatments were weighed. The food utilization index (i.e., the efficiency of conversion of ingested food (ECI)) was measured according to Chen et al. (2005).

2.4. Risk assessment

The risk of insecticides to bees was assessed using two common methods.

First, the toxicity grades of insecticides to bees were classified based on the acute toxicity (i.e., the median lethal dose, LD_{50}) under laboratory conditions (Li et al., 2017): (1) low toxicity ($LD_{50} > 11.0$ μg a.i. bee^{-1}); (2) medium toxicity (2.0 μg a.i. $bee^{-1} < LD_{50} \leq 11.0$ μg a.i. bee^{-1}); (3) high toxicity (0.001 μg a.i. $bee^{-1} < LD_{50} \leq 2.0$ μg a.i. bee^{-1}); and (4) extremely high toxicity ($LD_{50} \leq 0.001$ μg a.i. bee^{-1}).

Second, the ecological risks of six insecticides to *O. excavata* were determined using the hazard quotient (HQ) method based on the acute toxicity under laboratory conditions and the maximum field recommended rate (EPPO, 2000). The HQ values for each insecticide were calculated using the following equation.

$$\text{Hazard Quotient(HQ)} : HQ = AR/LD_{50} \quad (2)$$

In this equation, AR is the maximum field recommended application

Table 1
The concentrations of insecticides used in this study.

Bioassay	Test concentration (μg a.i. mL^{-1})					
	Clothianidin	Acetamiprid	Sulfoxaflor	Lambda-cyhalothrin	Chlorfenapyr	Abamectin
Acute lethal toxicity test	0.5	50	100	5	100	0.02
	1	100	200	10	200	0.04
	2	200	400	20	400	0.08
	4	400	800	40	800	0.16
	8	800	1600	80	1600	0.32
	16	1600	3200	160	3200	0.64
	32	3200	6400	320	6400	1.28
Sublethal toxicity test	0.5	50	100	5	100	0.02
	1	100	200	10	200	0.04
	2	200	400	20	400	0.08
	4	400	800	40	800	0.16

rate (g a.i. ha^{-1}) registered for use of an insecticide on apple trees and pear trees, as stipulated by the Institute for Control of Agrochemicals, Ministry of Agriculture, Beijing, China; and LD_{50} is the median lethal dose of each insecticide to *O. excavata* (μg a.i. bee^{-1}). The insecticides were considered "safe", "slightly to moderately toxic" and "dangerous" when the calculated HQ values were ≤ 50, $50 < HQ \leq 2500$, and >2500, respectively (EPPO, 2000; Ji et al., 2015).

2.5. Data analysis

All data were analyzed using the R software version 3.0.3. Prior to statistical analysis, the Shapiro–Wilk and Levene tests were applied to evaluate data normality and homogeneity, respectively. Data were transformed (log_{10} or arcsine square-root) for further statistical analysis when necessary, but untransformed means are presented in the following figures and tables. The LD_{50} was determined by a log-probit regression analysis. An analysis of covariance (ANCOVA) was conducted to evaluate the impacts of the six insecticides on the development and food utilization of *O. excavata*, while initial provision mass was a covariate. One-way analysis of variance (ANOVA) was used to evaluate the impacts of insecticides on eclosion rate. Pearson's correlation was performed to analyze the relationships between ingested dosages and eclosion rates of *Osmia excavata*. The Tukey's honestly significant difference (HSD) test was used to analyze the significant differences between treatments at $P < 0.05$.

3. Results

3.1. Acute lethal toxicity and risk assessment

The LD_{50} values of clothianidin, acetamiprid, sulfoxaflor, lambda-cyhalothrin, chlorfenapyr and abamectin to *O. excavata* were 0.007 (0.006–0.008), 0.91 (0.72–1.19), 1.46 (1.18–1.87), 0.047 (0.032–0.070), 2.28 (1.75–3.17) and 0.0004 (0.0003–0.0006) μg a.i. bee^{-1}, respectively. According to the LD_{50} values, the toxicity grades of clothianidin, acetamiprid, sulfoxaflor and lambda-cyhalothrin to the larvae of *O. excavata* were "high", while the toxicities of chlorfenapyr and abamectin were "medium" and "extremely high", respectively. On the basis of LD_{50} and the maximum field application recommended rate, the ecological risks of clothianidin (HQ = 42,857) and abamectin (HQ = 480,000) to the larvae of *O. excavata* were "dangerous". Similarly, acetamiprid (HQ = 206), lambda-cyhalothrin (HQ = 1596) and chlorfenapyr (HQ = 253) were classified as "moderately toxic" to the larvae of *O. excavata*, and sulfoxaflor (HQ = 45) was classified as "safe" (Table 2).

3.2. Effect of insecticides with different modes of action on the development of O. excavata

Compared to that in the CK, the larval weight gain of *O. excavata* was significantly lower after 14 d of treatments with the insecticides when the ingested dosages exceeded different thresholds: clothianidin (0.02–0.07 μg a.i. bee^{-1}), lambda-cyhalothrin (0.33–0.65 μg a.i. bee^{-1}) and abamectin (0.0003–0.0023 μg a.i. bee^{-1}). The corresponding inhibition rates of larval weights of *O. excavata* were 9.67–20.23% (clothianidin), 9.17–12.84% (lambda-cyhalothrin) and 5.85–22.29% (abamectin) (Fig. 1). Although the minimum test concentrations of acetamiprid and sulfoxaflor were both more than the maximum field registered concentrations (MFRC), the two insecticides have no significant effects on the larval weight gain of *O. excavata* (Table S3 and Fig. S1).

Compared to that in the CK treatment, decreased development speed of *O. excavata* before cocooning was observed after treatments with the insecticides when the ingested dosages exceeded different thresholds: clothianidin (0.01–0.07 μg a.i. bee^{-1}), lambda-cyhalothrin (0.09–0.65 μg a.i. bee^{-1}), chlorfenapyr (3.65–15.23 μg a.i. bee^{-1}), and abamectin (0.0003–0.0023 μg a.i. bee^{-1}; Fig. 1). The corresponding larval developments took 7.35–18.81% (clothianidin), 26.79–35.59% (lambda-cyhalothrin), 8.10–15.91% (chlorfenapyr), and 35.24–48.38% (abamectin) longer, respectively. However, sulfoxaflor with the minimum test concentration (>MFRC) has no significant effects on the development speed of *O. excavata* (Table S3 and Fig. S1).

Significantly negative linear relationships were observed between the eclosion rates of *O. excavata* and the dosages of clothianidin ($R^2 = 0.97$, $P = 0.002$), acetamiprid ($R^2 = 0.96$, $P = 0.004$), sulfoxaflor ($R^2 = 0.92$, $P = 0.01$), lambda-cyhalothrin ($R^2 = 0.89$, $P = 0.02$), chlorfenapyr ($R^2 = 0.95$, $P = 0.005$), and abamectin ($R^2 = 0.94$, $P = 0.006$). The eclosion rates of *O. excavata* treated with the insecticides were

Table 2
Toxicity of insecticides with different modes of action to *Osmia excavata* after 48 h of treatments.

Insecticides	Slope ± SE	Df	χ^2 (*P*)	LD_{50} (95% CI) (μg a.i. bee^{-1})	LD_{90} (95% CI) (μg a.i. bee^{-1})	Toxicity grade	HQ
Clothianidin	1.37 ± 0.10	5	0.47 (0.99)	0.007 (0.006–0.008)	0.059 (0.041–0.096)	High	42,857
Acetamiprid	1.10 ± 0.09	5	1.12(0.95)	0.91 (0.72–1.19)	13.17 (7.76–27.31)	High	206
Sulfoxaflor	1.11 ± 0.10	5	5.38 (0.37)	1.46 (1.18–1.87)	19.54 (12.07–37.59)	High	45
Lambda-cyhalothrin	1.07 ± 0.09	5	8.49 (0.13)	0.047 (0.032–0.070)	0.73 (0.35–2.64)	High	1596
Chlorfenapyr	1.01 ± 0.09	5	1.73 (0.89)	2.28 (1.75–3.17)	42.64 (22.60–105.05)	Medium	253
Abamectin	1.15 ± 0.10	5	3.78 (0.58)	0.0004 (0.0003–0.0006)	0.005 (0.003–0.011)	Extremely high	480,000

Notes: SE – standard error; DF – degree of freedom; χ^2 – values of Chi-square; CI – confidential interval; HQ – hazard quotient.

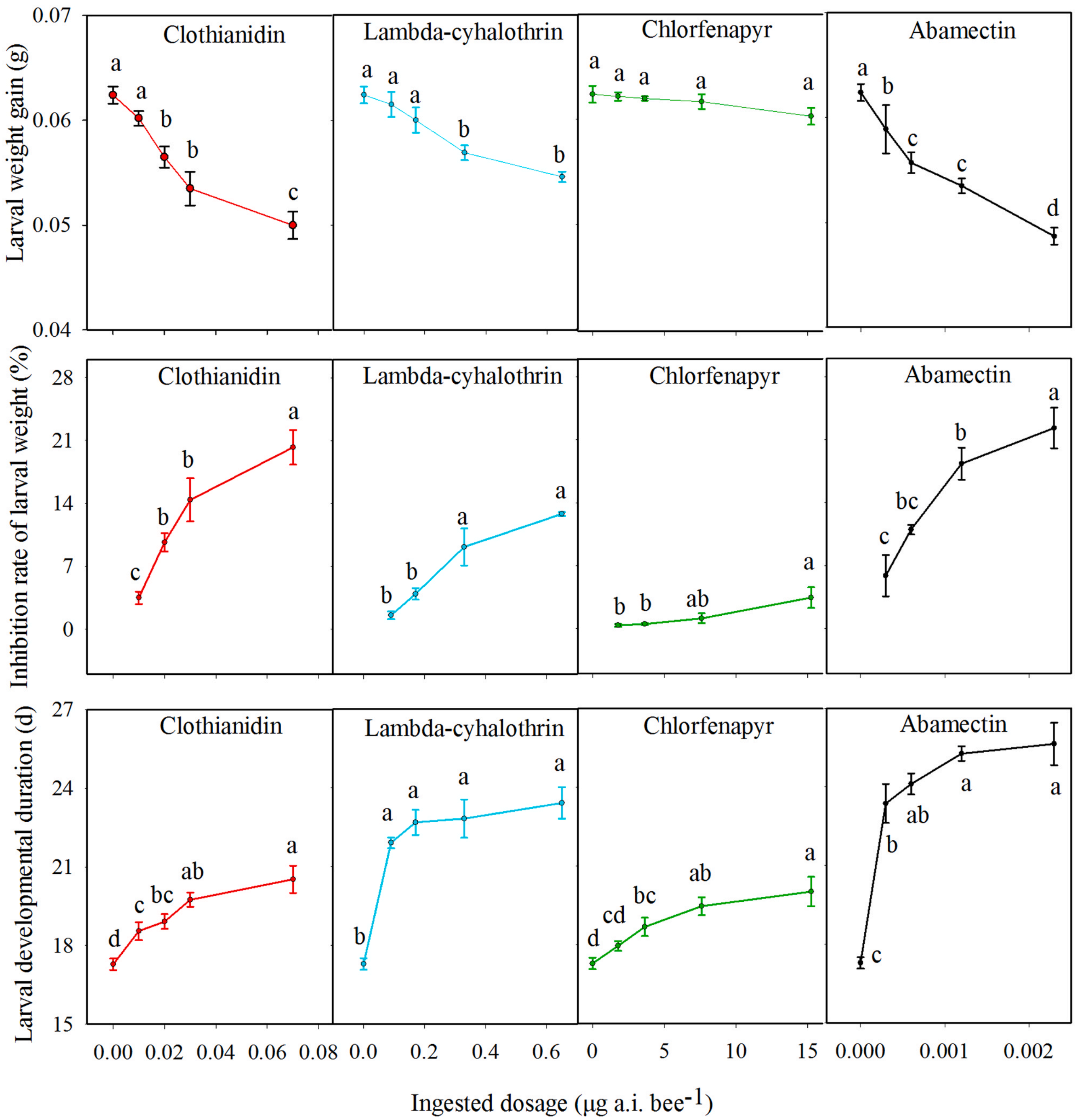

Fig. 1. Effect of insecticides with different modes of action on the larval weight gain, inhibition rate of larval weight after 14 d of treatment and the larval developmental duration before cocoon of *O. excavata*. (Different letters were significantly different within each insecticide according to Tukey's honestly significant difference (HSD) test at $P < 0.05$; the same as in the following figures).

significantly lower than those in the CK treatment although the ingested dosages for each insecticide differed by *O. excavata* during the whole larval stage: clothianidin (0.02–0.08 μg a.i. bee^{-1}), lambda-cyhalothrin (0.1–0.8 μg a.i. bee^{-1}), chlorfenapyr (4.0–16.00 μg a.i. bee^{-1}), and abamectin (0.4–3.2 ng a.i. bee^{-1}). The corresponding eclosion rates decreased 9.72–31.94% (clothianidin), 17.64–53.72% (lambda-cyhalothrin), 12.38–27.06% (chlorfenapyr), and 14.33–40.43% (abamectin), respectively (Fig. 2). However, sulfoxaflor with the minimum test concentration (> MFRC) has no significant effects on the eclosion rates of *O. excavata* (Table S3 and Fig. S2).

3.3. *Effect of insecticides with different modes of action on the food utilization by O. excavata*

Compared to that in the CK treatment, the food consumption per larva of *O. excavata* was significantly lower after 14 d of treatments with the insecticides when the ingested dosages exceeded different thresholds: clothianidin (0.03–0.07 μg a.i. bee^{-1}), lambda-cyhalothrin (0.17–0.65 μg a.i. bee^{-1}), and abamectin (0.0003–0.0023 μg a.i. bee^{-1}). The corresponding food consumption per larva decreased 5.23–6.76% (clothianidin), 3.55–7.76% (lambda-cyhalothrin), and 3.26–16.79% (abamectin). In contrast, the food consumption per larva was significantly higher than that in the CK treatment when the application of chlorfenapyr was in the range of 3.65–15.23 μg a.i. bee^{-1}

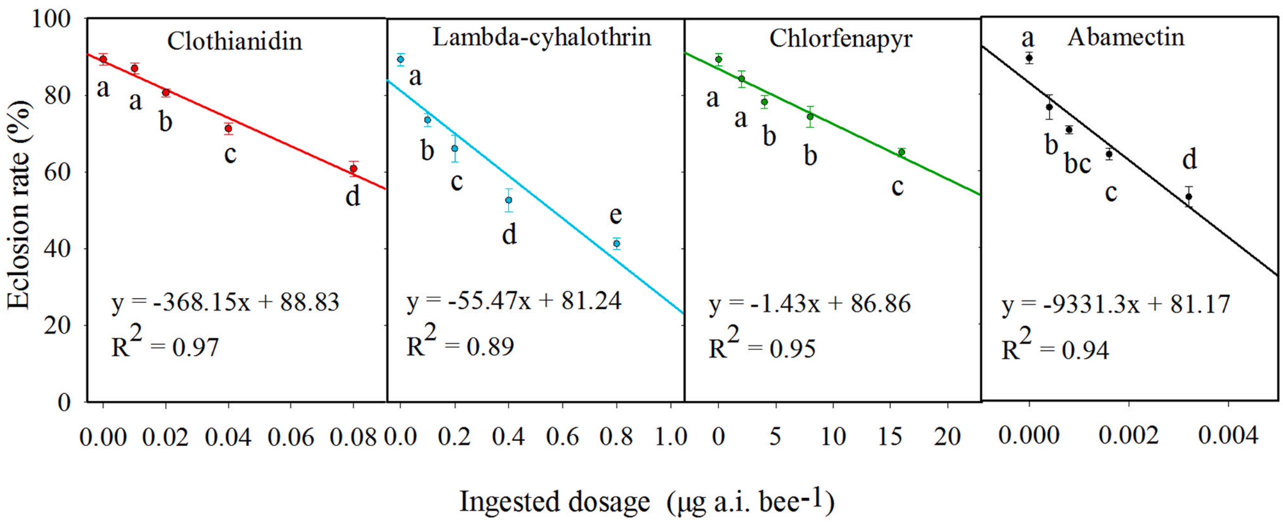

Fig. 2. Relationship between ingested dosages and eclosion rates of *Osmia excavata*.

(4.25–8.81%; Fig. 3). However, acetamiprid and sulfoxaflor with the minimum test concentrations (>MFRC) have no significant effects on the food consumption per larva of *O. excavata* (Table S3 and Fig. S3).

Relative to those in the CK treatment, the ECI values for *O. excavata* were also significantly lower after 14 d of treatments with the insecticides when the ingested dosages exceeded different thresholds: clothianidin (0.02–0.07 μg a.i. bee^{-1}), lambda-cyhalothrin (0.33–0.65 μg a.i. bee^{-1}), chlorfenapyr (3.65–15.23 μg a.i. bee^{-1}), and abamectin (0.0023 μg a.i. bee^{-1}). The corresponding ECI value decreased 9.99–14.50% (clothianidin), 4.68–5.54% (lambda-cyhalothrin), 4.59–11.30% (chlorfenapyr), and 6.71% (abamectin) (Fig. 3). However, acetamiprid with the minimum test concentration (>MFRC) has no significant effects on the ECI values for *O. excavata* (Table S3 and Fig. S3).

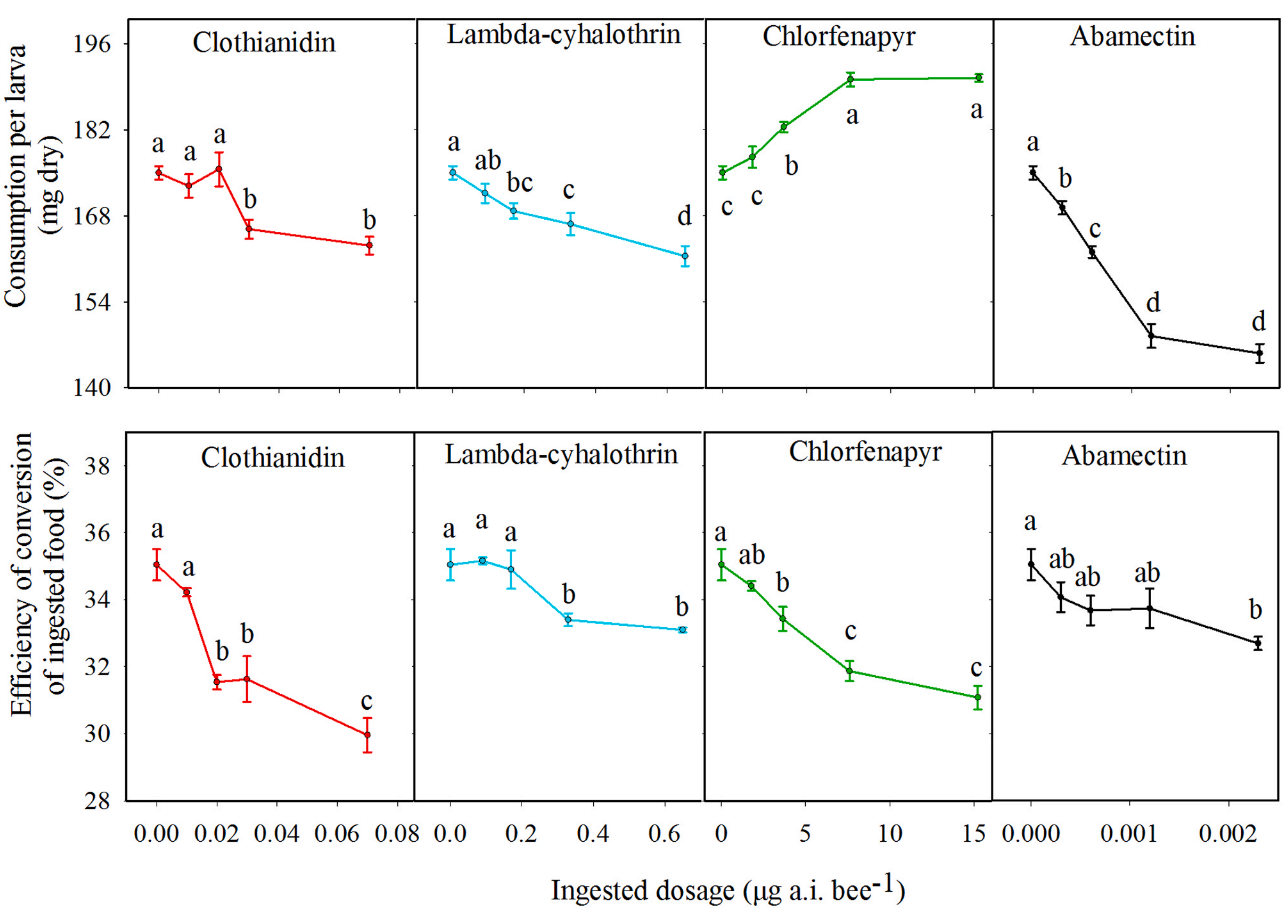

Fig. 3. Effect of insecticides with different modes of action on the consumption per larva and the efficiency of conversion of ingested food by *O. excavata* after 14 d of treatments.

4. Discussion

Pollen grain is one of the most important food source for the larvae of *Osmia*, residual insecticides in the pollen after application would be eaten by the larvae. To date, however, there are still no in-depth investigations of the toxicity of pesticides to solitary bees. In the current study, we evaluated the toxicities of widely used insecticides with different modes of action in fruit trees. The study results demonstrated that among the six insecticides investigated, abamectin (with an LD_{50} value of 0.0004 μg a.i. bee^{-1}) was the most toxic to the larvae of *O. excavata*. Furthermore, the HQ score of abamectin indicated that the ecological risk of this insecticide is dangerous to the larvae of *O. excavata*. Earlier studies about the acute lethal effects of these insecticides corroborated our findings. For instance, Zhang et al. (2017) reported that abamectin (with an LD_{50} value of 0.007 μg a.i. bee^{-1}) posed relatively high toxicity to adult pollinators (*Apis mellifera* L.) that was approximately 17.5-fold greater than that found in the current study. Aljedani and Musleh (2017) also found that abamectin was extremely toxic to foraging honey bee workers (*Apis mellifera jemenatica* (Hymenoptera: Apidae)) under laboratory conditions. Although the toxicity grades of the two neonicotinoid insecticides used in this study were both high, acetamiprid was less toxic to the larva of *O. excavata* than clothianidin (130-fold = LD_{50} (acetamiprid)/LD_{50} (clothianidin)); the ecological risk of clothianidin was found to be high. The diversity of chemical structures in neonicotinoids may be responsible for their different toxicities (Jiang et al., 2019). Jeschke et al. (2011) reported that acetamiprid has a cyano group, in contrast to clothianidin and other neonicotinoid insecticides, which have nitro groups. Overall, the results from the current study suggested that abamectin and clothianidin at the recommended field dosages may cause high toxicity risks to *O. excavata*, and should not be sprayed during the flowering phase. If these two insecticides were to be used at other times, the application rates of abamectin and clothianidin still needs further study to control them within acceptable toxicity risks to *O. excavata*. According to the acute lethal toxicity test, chlorfenapyr was the least toxic to the larvae of *O. excavata*, and sulfoxaflor was safe in terms of ecological risk (i.e. HQ value < 50). These results suggest that chlorfenapyr and sulfoxaflor are suitable options and can be recommended as insecticides for targeted insect control in orchards. The HQ and the trigger values of 50 and 2500 were mainly used for the evaluation of the risk in adult bees. In this study, our aim using above methods was to calculate the relative risk of each insecticide to the larvae of solitary bees considering the amount of product that can be applied in the field (e.g. the application rate) and we were using this information only as a comparative approach among the different insecticides. Additionally, the HQ values may be overestimated by using the application rate under field settings because insecticides may be degraded when exposed to UV, or incorporated systemically in plants, etc. In order to better evaluate the hazard, the concentration of pesticide residue in pollen in relation to the acute toxicity to *O. excavata* instead of the field application rate should be considered in successive researches.

It is a frequent scenario that pesticides degrade into sublethal doses over time in agroecosystems due to abiotic factors such as temperature, rainfall and sunlight. Generally, sublethal effects of insecticides are important in understanding the impact of pesticides on the population dynamics of insects (Desneux et al., 2007). This study measured the sublethal effects of pesticides in pollen on the development and feeding of *O. excavata*. Larval development of *O. excavata* before cocooning was remarkably prolonged after treatments with the insecticides in this study. These results agree with the finding regarding the effect of sublethal dosages or concentrations of clothianidin on *Coccinella septempunctata* (Jiang et al., 2018). Additionally, compared with those not exposed to the insecticides, larval weights were significantly lower after 14 d of treatments with clothianidin and abamectin (<MFRC), suggesting that the growth and development of *O. excavata* were disturbed by these insecticides. In the current study, significantly negative linear relationships were also observed between eclosion rates and the dosages of insecticides, which may be attributable to dysfunction and deformity of *O. excavata* (e.g., cuticle sclerotization and the formation of wings; Jiang et al., 2018; Desneux et al., 2007) caused by the persistent effects of the pesticides. Further research is necessary to understand the basis of these phenomena.

Interestingly, we further observed that the ECI values (indicating the percentage of food ingested by insect and transformed into body biomass) of *O. excavata* were profoundly lower after treatments with insecticides at sublethal dosages. Accordingly, we believed that nutrition gained from pollen used for the growth and development of larvae was decreased, either through the larvae's reduced feeding activity or lowered efficiency of converting ingested food. Both factors are in accordance with findings in previous studies. For example, neonicotinoids can lower the feeding efficiency of insects by inducing significant neurotoxic symptoms, such as repellency, paralysis, and loss of coordination (Xie et al., 2015). Aljedani and Musleh (2017) found that abamectin may cause digestive disorders because it can exert a cytotoxic effect on midgut cells, which are the most important sites for terminal digestion and absorption of nutrients. However, Jonathan et al. (2008) believed that reduced growth of insects exposed to insecticide-treated food may be a result of energy diversion for detoxification processes rather than food avoidance after insecticide treatment. Although food consumption per larva was significantly increased in the chlorfenapyr treatment, the ECI values of *O. excavata* were profoundly decreased compared to those of larvae in the CK treatment. Possible reasons for this phenomenon may lie in the compensatory response in addition to lower nutrient absorption capacity of insects. Ramalho et al. (2011) reported that insects exhibited overcompensation responses by stimulating consumption as a response to low-quality food.

5. Conclusions

To our knowledge, this study firstly researched the effects of commonly used insecticides on the development (e.g., larval weight, larval development before cocooning and eclosion rate) and the food utilization of the larvae of solitary bees. The results demonstrated that clothianidin and abamectin observably decreased the survival of the larvae of *O. excavata* and posed relatively high ecological risk, and these two insecticides should not be applied, especially during the flowering phase of fruit trees. Chlorfenapyr posed the least acute lethal toxicity to *O. excavata*, sulfoxaflor was identified as the only safe insecticide for *O. excavata* (HQ > 50). Additionally, acetamiprid and sulfoxaflor with the minimum test concentration (>MFRC) have little significant negative effects on the development of larvae and food utilization. However, this study just conducted in the laboratory, appropriate field dosages should be further verified for the purpose of conserving *Osmia*.

CRediT authorship contribution statement

Yingying Song: Investigation, Validation, Formal analysis, Writing - original draft. **Lili Li:** Investigation, Validation, Writing - review & editing, Supervision. **Chao Li:** Investigation, Formal analysis, **Zengbin Lu:** Investigation, Formal analysis, Writing - review & editing. **Fang Ouyang:** Formal analysis, Writing - review & editing. **Li Liu:** Investigation, Formal analysis. **Yi Yu:** Writing - review & editing, Supervision. **Xingyuan Men:** Conceptualization, Resources, Writing - review & editing, Supervision, Project administration, Funding acquisition.

Declaration of Competing Interest

The authors have declared no competing financial interests.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.ecoenv.2021.112015.

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