Screening of chemical insecticides against Diaghania pyloalis

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Abstract. In recent years, a kind of mulberry pests which were known as mulberry pyralid Diaghania pyloalis in different sericultural areas occurred popularly in China. This study aims to screening high efficacy and low toxicity insecticides for controlling Diaghania pyloalis. The current study can help understand integrated pest management (IPM) of Diaghania pyloalis by scientific and reasonable insecticide use. Field experiment was carried out to investigate five insecticides, treated with five concentration gradients. The result shows that Imidacloprid and Chlorfenapyr had higher control effect on Diaghania pyloalis, which was relatively safe to the silkworm. The silkworm was fed until cocooning in the treatment area which sprayed for 15 days and 30 days, investigated cocoon quality achievement, the result shows that 10% Imidacloprid WP and 8% Chlorfenapyr ME had no toxicity to silkworm larvae, there was no significant difference between the economic property index of silkworm in treatment areas and that of the control area. To control Diaghania pyloalis, 2000 dilution of 10% Imidacloprid WP and 8% Chlorfenapyr ME can be used in sericultural production, the leaves should be picked at intervals of 15 days to avoid affecting the quality of cocoons.

1. Introduction
The mulberry moth Diaghania pyloalis Walker, commonly known as mulberry caterpillar, leaf roller, oil insects, etc., belongs to Pyralidae in Lepitoptera[1]. The mulberry moth has gradually become the primary pest in mulberry fields because of its wide spread, strong fecundity capacity, terrible diffusion ability[2-3]. The pest occurred in sericultural areas except Xinjiang, especially in Jiangsu, Zhejiang, Guangdong, Anhui, Jiangxi, Sichuan, Hunan, Hubei, Guangxi and Chongqing[2]. For example in Yizhou District, a city of GuangXi province, the pest damage rate was as high as 86% in 2019 [4]. Study on the regularity and control methods of the pest has become an important topic in the national mulberry field. At present, chemical control is mainly used in the control of mulberry moth[2-5], but the mulberry moths are resistant to pesticides which are commonly used in mulberry fields[2-5], like mulberry moth has developed greater resistance to dichlorvos, phoxim and chlorpyrifos[5]. In addition, the main pesticide methomyl used in mulberry garden has been included in the pesticide list of restricted since 2017. It was very unfavorable to the control of Diaghania pyloalis Walker, so it is urgent to screen suitable pesticides in production. Therefore, this study is carried out screening of high-efficiency and low-toxicity insecticides for controlling Diaghania pyloalis, providing the theory basis for resolving the problem of the pest in scientific, proper, economical and effective ways.
2. Materials and Methods

2.1. Experimental materials

The experiment was carried out at the Ganning Base of the Chongqing Three Gorges Academy of Agricultural Sciences. The experimental site is 325 m above sea level. Flat terrain, uniform fertility and consistent field management were chosen for experiments. The test soil was sandy loam soil, with thickness equal to or higher than 1.0 m.

The mulberry variety used in this test was Hu mulberry with planting density of 9 000 plants /hm². The silkworm species tested in this experiment were Chuanbai×Shufang, which are spotted double-limited varieties. The original species were supplied by Sichuan Santai silkworm seed farm.

2.2. Experimental treatments

In May 2020, insecticides were selected for testing listed in table 1. The test was carried out to investigate different insecticides, each of which was treated with 5 concentration gradients. Control was sprayed by clean water. The shape of the plot was square with basically the same area, around which guardrows established. The insecticides were sprayed thoroughly and evenly on the mulberry trees by using electric sprayers after the dew dried up in the morning. 30 mulberry moth larvae and 50 molted silkworms of 3 ages were fed with suitable ripe mulberry leaves in each treatment area, and fed continuously for 7 days. Each treatment was repeated 3 times. The number of poisoning deaths of mulberry moth larvae and silkworm larvae was investigated daily.

In June 2020, Residual toxicity tests of insecticides on mulberry leaves were carried out, Bidirectional experiment was employed, which combined survey results from pesticide disposal and interval time of leaf picking for sericulture. Pesticide disposal was shown in table 1, the concentration of pesticides was 2000 times diluted solution, clear water was blank control, there were 6 treatments in total. The interval times of leaf picking for sericulture were 15 days, 20 days, 30 days and 35 days after spraying on the mulberry trees. 50 molted silkworms of 3 ages were tested in each processing area, and repeated 3 times. Silkworms of the first and second ages were fed routinely, silkworms of the 3rd age began to feed mulberry leaves with different treatments, and fed continuously for 1 instar, and then the number of dead silkworms in each treatment was calculated. The larvae were fed to spinning cocoons in treatment areas which were 15 days and 30 days after spraying on the mulberry trees. Cocoon quality achievement of each district was investigated.

The following is the calculation formula: Death rate (%) = the number of death larvae / Total number of larvae before spraying ×100; Correction control effect (%) = (pest quantity in control group - pest quantity in treatment group)/pest quantity in control group ×100.

| Insecticides  | Content/Formulation | Manufacturer                      |
|---------------|---------------------|-----------------------------------|
| Imidacloprid  | 10% WP              | Shandong Jiacheng Crop Science Co. Ltd |
| Chlorfenapyr  | 8% ME               | Guangxi pastoral biochemistry co. Ltd |
| Cyromazine    | 80% WDG             | Guangdong Zhongxun Agricultural Science Co. Ltd |
| Bifenthrin    | 2.5% EW             | Chengdu keilong biochemical co. Ltd |
| Thiamethoxam  | 25% WDG             | Jiangsu Changqing Agrochemical Co. Ltd |

2.3. Data analysis

Statistical analysis of data was performed by using Microsoft Office 2010 and IBM SPSS 16.0. One-Way ANOVA was used to conduct difference analysis.

3. Results and analysis

3.1. Effects of different insecticides against Diaghania pyloalis

In May 2020, the control effect of 5 insecticides on Diaghania pyloalis was tested. The results showed that all insecticides had certain control effect on Diaghania pyloalis, all the mulberry moth larvae died after continuous feeding mulberry leaves for 96 hours (shown in table 2). In general, the control
efficiency increased with the dosage increased. 2.5% Bifenthrin EW and 25% Thiamethoxam WDG, gained the best control effects against Diaghania pyloalis, the control correction effects were significantly higher than that of other groups, no significant difference between different concentrations, all larvae died after feeding for 24 hours. 8% Chlorfenapyr ME, 10% Imidacloprid WP, and 80% Cyromazine WDG, gained the second effective, and there were significant differences in the control effects of different concentrations of insecticides. The mulberry moth larvae died after feeding for 48 hours with mulberry leaves, which sprayed 8% Chlorfenapyr ME, 1000~1500 times solution of 10% Imidacloprid WP and 1000 times solution of 80% Cyromazine WDG. The mulberry moth larvae died after feeding for 72 hours with mulberry leaves, which sprayed 2000~2500 times solution of 10% Imidacloprid WP and 1500 times solution of 80% Cyromazine WDG. Although these five insecticides had better control effect on Diaghania pyloalis, they also had great influence on the mortality of silkworm larvae (shown in table 3). The mortality of silkworm larvae was consistent with that of Diaghania pyloali, that is, mortality rate of silkworm larvae increased with the increase of the control effects against Diaghania pyloalis.

### Table 2  Control effects of different insecticides dosages on *Diaghania pyloalis*

| Fungicides | Dilution times | Medication time 24h | Medication time 48h | Medication time 72h | Medication time 96h |
|------------|----------------|---------------------|---------------------|---------------------|---------------------|
|            | Number of larvae /heads | control larvae effect % | Number of larvae /heads | control larvae effect % | Number of larvae /heads | control larvae effect % | Number of larvae /heads | control larvae effect % |
| Imidacloprid | 1000 0.33 a 98.89 a | 0.00 a 100.00 a | - | - | - |
|             | 1500 4.33 b 85.56 b | 0.00 a 100.00 a | - | - | - |
|             | 2000 5.67 c 81.11 c 1.67 ab | 94.44 ab 0.00 a 100.00 a | - | - | - |
|             | 2500 5.67 c 81.11 c 1.00 a 96.67 a 0.00 a 100.00 a | - | - | - | - |
|             | 3000 12.67 e 57.78 g 3.67 c 87.78 c 1.67 b 94.44 a 0.00 100.00 | - | - | - | - |
|             | 1000 0.67 a 97.78 a | 0.00 a 100.00 a | - | - | - |
|             | 1500 3.33 b 88.89 b | 0.00 a 100.00 a | - | - | - |
|             | 2000 11.33 f 62.22 f | 0.00 a 100.00 a | - | - | - |
| Chlorfenapyr | 2500 3.67 b 87.78 b | 0.00 a 100.00 a | - | - | - |
|             | 3000 7.67 d 74.44 d | 0.00 a 100.00 a | - | - | - |
|             | 1000 0.67 a 97.78 a | 0.00 a 100.00 a | - | - | - |
|             | 1500 9.00 e 70.00 e 3.00 bc 90.00 bc 0.00 a 100.00 a | - | - | - | - |
|             | 2000 19.33 h 35.56 h 13.67 d 54.44 d 9.67 c 67.78 b 0.00 100.00 | - | - | - | - |
|             | 2500 21.67 i 27.78 i 18.00 e 40.00 e 13.67 d 54.44 c 0.00 100.00 | - | - | - | - |
|             | 3000 24.00 j 20.00 j 21.33 f 28.89 f 18.00 e 40.00 d 0.00 100.00 | - | - | - | - |
|             | 1000 0.00 a 100.00 a | - | - | - | - |
|             | 1500 0.00 a 100.00 a | - | - | - | - |
|             | 2000 0.00 a 100.00 a | - | - | - | - |
|             | 2500 0.00 a 100.00 a | - | - | - | - |
|             | 3000 0.00 a 100.00 a | - | - | - | - |
|             | 1000 0.00 a 100.00 a | - | - | - | - |
|             | 1500 0.00 a 100.00 a | - | - | - | - |
|             | 2000 0.00 a 100.00 a | - | - | - | - |
|             | 2500 0.00 a 100.00 a | - | - | - | - |
|             | 3000 0.00 a 100.00 a | - | - | - | - |
| Thiamethoxam | CK 30.00 k | - | - | - | - |

Values followed by different letters at the same column indicate significant difference (P<0.05), and those followed by the same letters indicate no significant difference (P≥0.05), the same as below.

### Table 3  Effects of insecticides on mortality of silkworm larvae

| Fungicides | Dilution times | Medication time 24h | Medication time 48h | Medication time 72h | Medication time 96h |
|------------|----------------|---------------------|---------------------|---------------------|---------------------|
|            | Number of larvae /heads | control larvae effect % | Number of larvae /heads | control larvae effect % | Number of larvae /heads | control larvae effect % | Number of larvae /heads | control larvae effect % |
| Imidacloprid | 1000 0.00 a 100.00 a | - | - | - | - |
|             | 1500 8.67 c 82.67 c | 0.00 a 100.00 a | - | - | - |
|             | 2000 15.00 e 64.00 e | 6.67 b 86.67 b | 1.00 a 98.00 a | 0.00 a 100.00 a | - | - |
|             | 2500 24.00 e 52.00 e | 1.10 c 78.00 c | 0.00 a 100.00 a | - | - | - |
|             | 3000 35.00 g 30.00 g | 23.67 e 52.67 e | 5.00 b 90.00 b | 0.00 a 100.00 a | - | - |
|             | 1000 0.00 a 100.00 a | - | - | - | - |
| Chlorfenapyr | 1500 6.00 b 88.00 b | 0.00 a 100.00 a | - | - | - |
|             | 2000 19.00 d 62.00 d | 0.00 a 100.00 a | - | - | - |
|             | 2500 25.33 c 49.33 e 13.33 c 73.33 c | 0.00 a 100.00 a | - | - | - |
3.2. Residual toxicity of insecticides on mulberry leaves

Because mulberry leaves sprayed by insecticides had a great influence on the mortality of silkworm larvae, residual toxicity tests of insecticides on mulberry leaves were carried out in June 2020. The results (shown in table 4) showed that two insecticides had no toxicity to silkworm larvae, larvae did not die and showed toxic symptoms, which fed with mulberry leaves in the treated area sprayed 8% Chlorfenapyr ME and 10% Imidacloprid WP. 80% Cyromazine WDG, 2.5% Bifenthrin EW and 25% Thiamethoxam WDG had great influence on silkworm. Mulberry leaves were fed to silkworms of the third age, sprayed 80% Cyromazine WDG interval times from 15 days to 35 days. All the silkworms showed symptoms of poisoning including shortening body, and some of them resisting food, vomiting fluid. Although silkworm mortality was low after continuously feeding with mulberry leaves for 96 hours, silkworm bodies were significantly smaller than those in the control group at the fifth instar in the processing area, in which sprayed insecticides for 15 days or 30 days, and silkworms wouldn't spin silk cocoons. Sprayed 2.5% Bifenthrin EW and 25% Thiamethoxam WDG had great influence on the mortality rate of silkworm, all larvae died after feeding mulberry leaves which sprayed 2.5% Bifenthrin EW for 24 hours. Sprayed 25% Thiamethoxam WDG on mulberry leaves, all silkworms died when mulberry leaves were fed for 24 hours with spraying 15 days interval, and for 72 hours with spraying 30 days interval, and for 96 hours with spraying 30~35 days interval. Silkworm were fed until cocooning in treatment areas, which interval times of leaf picking for sericulture were 15 days and 30 days after spraying on the mulberry trees. Silkworm were successfully fed to cocoon forming only in treatment areas, in which sprayed 8% Chlorfenapyr ME and 10% Imidacloprid WP. The results of silkworm breeding achievements were shown in table 5. There was no significant difference between the economic property index of silkworm which included whole cocoon weight, cocoon shell weight, cocoon layer ratio, pupa death rate and pupa weight in treatment areas and that of the control area. It can be seen that the mulberry leaves sprayed with 2000 times solution of 8% Chlorfenapyr ME and 10% Imidacloprid WP for 15 days, had little influence on the survival and economic properties of silkworm. To control Diaghania pyloalis, 10% Imidacloprid WP and 8% Chlorfenapyr ME can be used in sericultural production, the leaves should be picked at intervals of 15 days.

| Fungicides | Interval times/d | Medication time 24h Mortality /% | Medication time 48h Mortality /% | Medication time 72h Mortality /% | Medication time 96h Mortality /% |
|------------|-----------------|---------------------------------|---------------------------------|---------------------------------|---------------------------------|
|            | Number of silkworms / heads |                    |                                   |                                   |                                   |
| Imidacloprid | 15              | 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a | | | |
|            | 20              | 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a | | | |
|            | 30              | 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a | | | |
|            | 35              | 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a | | | |
| Chlorfenapyr | 15              | 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a | | | |
|            | 20              | 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a | | | |
|            | 30              | 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a 50.00 a 0.00 a | | | |
Table 5. Effects of insecticides on economic property indexes of Silkworms

| Fungicides   | Interval times/d | Whole cocoon weight/g | Cocoon shell weight/g | Cocoon layer ratio/% | Pupa death rate/% | Pupa weight/g |
|--------------|------------------|------------------------|-----------------------|----------------------|-------------------|---------------|
| Cyromazine   | 15 20 30 35      | 50.00 a 0.00 a 49.33 a | 49.33 a 1.33 a 49.33 b | 1.33 b 49.33 b      |                   |               |
| Bifenthrin   | 20 30 35         | 0.00 e 100.00 e       | -                     | -                    | -                 | -             |
| Thiamethoxam | 20 30 35         | 3.67 d 92.67 d 1.00 d | 98.00 d 0.00 e 100.00 e | -                    | -                 | -             |
| CK           | 50.00 a 0.00 a   | 50.00 a 0.00 a 50.00 a | 0.00 a 50.00 a 50.00 a | 0.00 a              |                   |               |

4. Conclusion and Discussion

Five insecticides had a certain control effect on Diaghania pyl oalis, but three of them had more toxicity on silkworm. 2.5% Bifenthrin EW and 25% Thiamethoxam WDG had highly toxic to silkworm and had a long residual period, all silkworms died when mulberry leaves were fed for 96 hours with spraying 35 days interval, therefore, they have been forbidden to use in mulberry fields for sericultural production. Although silkworm mortality was low after continuously feeding with mulberry leaves sprayed 80% Cyromazine WDG for 96 hours, the toxic effects of cyromazine were accumulative and slow to appear by continuous feeding, then silkworm bodies were significantly smaller than those in the control group, and silkworms wouldn't spinning silk cocoons, so cyromazine should be banned too.

Imidacloprid and Chlorfenapyr had higher control effects on Diaghania pyl oalis, which were relatively safe to the silkworm. Silkworm larvae did not die and showed on toxic symptoms fed with mulberry leaves sprayed 8% Chlorfenapyr ME and 10% Imidacloprid WP for 15 days or 30 days. The silkworm was fed until cocooning in the treatment area which sprayed for 15 days and 30 days, the results of sericulture quality achievement show that 2.5% Bifenthrin EW, 25% Thiamethoxam WDG and 80% Cyromazine WDG should be forbidden to use in mulberry fields for sericultural production. 10% Imidacloprid WP and 8% Chlorfenapyr ME had no toxicity to silkworm larvae, there was no significant difference between the economic property index of silkworm in treatment areas and that of the control area. To control Diaghania pyl oalis, 2000 dilution of 10% Imidacloprid WP and 8% Chlorfenapyr ME can be used in sericultural production, the leaves should be picked at intervals of 15 days.

Acknowledgment

These experiments were supported by two projects, one of them is technical innovation and application development special general project of Chongqing "Research and Application of Prevention and Control Technology of Diaghania pyl oalis" (cstc2019jscx-msxmX0359), and the other one is Wan Zhou extension test station of modern mountain characteristic efficient agricultural (sericulture) technology system in Chongqing.

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