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Establishment of Baseline Toxicity Data to Different Insecticides for *Aphis craccivora* Koch and *Rhopalosiphum maidis* (Fitch) (Homoptera: Aphididae) by Glass Tube Residual Film Technique

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Abstract.- *Aphis craccivora* Koch is one of the most important pests of legumes, and *Rhopalosiphum maidis* (Fitch), is a serious pest of Gramineous crops. Susceptible strains of these two species were obtained and the baseline toxicity of 15 commonly used insecticides was established by a standard glass tube residual film method. The results showed that LC_{50} values of 15 tested insecticides against *A. craccivora* and *R. maidis* varied from 0.79 to 52.23 mg/L and 1.03 to 39.20 mg/L, respectively. Chlorpyrifos was proved to be the most toxic insecticide against both the species whereas Abamectin was least toxic. Susceptible toxicity baselines data of both *A. craccivora* and *R. maidis* to 15 insecticides established in this study could be used as a reference for resistance monitoring or other related researches.

Key words *Aphis craccivora* Koch, *Rhopalosiphum maidis* (Fitch), toxicity, insecticides, residual film method

INTRODUCTION

The corn leaf aphid, *Rhopalosiphum maidis* (Fitch) and the black cowpea aphid, *Aphis craccivora* Koch (Homoptera: Aphididae), are serious insect-pests of many crops throughout the world (Hill, 1987; Blackman and Eastop, 2000; Al-Eryan and El-Tabbakh, 2004; Kuo *et al.*, 2006). Apart from directly sucking the sap from various plant parts, these aphids can damage-the crops by producing plentiful honeydew which may result in deformed leaves, stunting and premature plant death, growth of sooty mold, reduction in photosynthesis as well as the sterilization of inflorescences (Hill, 1987; Bing *et al.*, 1991; Blackman and Eastop, 2000; Flint, 2000; Gonzáles *et al.*, 2001). In addition, they are a vector of plant viruses and may transmit dozens of viral diseases to different crops (Hill, 1987; Blackman and Eastop, 2000; Kaiser, 1979; Thottappilly and Rossel, 1985).

The current aphid management strategies heavily relied on the use of various synthetic chemical insecticides such as organophosphates, carbamates, pyrethroids, and neonicotinoids (Jackai and Daoust, 1986; Shetlar, 2001). The indiscriminate and large-scale use of synthetic chemical insecticides to control these aphids has resulted in the development of insecticide resistance (Hollingsworth, 1994; Han and Li, 2004). Increasing levels of resistance to the most commonly used insecticides have resulted in increased human health and environmental concerns (Holland *et al.*, 2000; Jansen, 2000). These problems indicate the need to establish an efficient resistance management strategies based on information available about the extent and nature of resistance.

Resistance monitoring can be an effective component of the resistance management approach by providing valuable information on responses of insect-pests populations to currently used insecticides. Detection of changes in field resistance can facilitate the use of alternative control measures, including use of synergists, rotational use of various insecticides, and reduced insecticide application (Yilma *et al.*, 1991; Lee *et al.*, 1997). Any bioassays for field resistance monitoring necessitates establishment of reliable susceptible toxicity baselines, as a standard of control. In present studies, the susceptible toxicity baselines of *A. craccivora* and *R. maidis* to 15 insecticides were established by method of residual film in glass tube, which can provide a basic information for the evaluation of standard for resistance monitoring in the future.

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MATERIALS AND METHODS

Insects

A. craccivora was collected from the experimental area of the South China Agricultural University, Guangzhou, Guangdong, China. *A. craccivora* was reared on the seedlings of broad bean (*Vicia faba* L.) in a plastic cage (60×40×10 cm) at 25±5°C and photoperiod of 14:10 (L:D) h for more than two years and there was no contact with any kind of insecticide. *R. maidis* was obtained form Zhejiang Academy of Agricultural Sciences, reared on the seedlings of wheat (*Triticum aestivum* L.) in a plastic cage (38×25×4 cm) at 20±1°C, under a photoperiod of 14:10 (L: D) h.

Insecticides

The insecticides used in this study were as follows: abamectin (94.4% purity), imidacloprid (96.4% purity), acetamiprid (96.4% purity), beta-cypermethrin (97% purity), lambda-cyhalothrin (96.5% purity), deltamethrin (97% purity), bifenthrin (97% purity), chlorpyrifos (96% purity), dimethoate (96% purity), profenofos (89% purity), malathion (90% purity), phoxim (90% purity), pymetrozine (95% purity), methomyl (98% purity) and pirimicarb (95% purity) provided by Academy of Agricultural Sciences, Guangdong Province, China.

Bioassays

Bioassays were conducted with apterous adult of *A. craccivora* and *R. maidis* by using a standard glass tube residual film method. Glass tubes (1.5cm diameter, 10 cm length) were rotated rapidly with 2 mL of 5-7 serial concentrations of respective insecticides dissolved in acetone, which contained a 0.1%(v/v) Trion X-100 and then poured off the excess solution, inverted on a wire rack and allowed to air dry for about 12 h at room temperature. Controls for each replicate were treated with acetone alone. For each aphid bioassay, at least 360 apterous adult aphids were treated per insecticide, usually with 3 replicates of 20 adults at each of 5 to 7 insecticide concentrations. Then transferred to a climatic cabinet, with conditions of 25±1°C, 14:10 (L:D) h photoperiod. After 12h, mortality was assessed. Adults were considered dead if they were only one leg moved, or unable to move when disturbed with a soft brush (Chen *et al.*, 2007). Bioassays with control mortality exceeding 10% were discarded and repeated.

Statistical analysis

Mortality data were analyzed by probit analysis (SPSS 10.0 Institute, 2000) to estimate the median lethal concentrations (LC_{50}) (Finney, 1971).

RESULTS

The LC_{50} values were used to evaluate the insecticides toxicity. Susceptible toxicity baseline data for 15 insecticides against apterous adult of *A. craccivora* are shown in Table I. The results showed that chlorpyrifos was the most toxic insecticide followed by methomyl, bifenthrin, deltamethrin, dimethoate, lambda-cypermethrin, profenofos, pirimicarb, beta-cypermethrin, phoxin, imidacloprid, malathion, acetamiprid, pymetrozine and abamectin. The acute toxicity of chlorpyrifos for *A. craccivora* was very high, and thus the LC_{50} value obtained was very low. The LC_{50} value for chlorpyrifos was only 0.79 mg/L. The abamectin and pymetrozine were slightly toxic with the LC_{50} values of 52.23 and 13.60 mg/L, respectively. The other insecticides tested in this study showed moderate toxicity and the LC_{50} values varied from 1.03 to 7.58 mg/L (Table I).

Susceptible toxicity baseline data for 15 insecticides against apterous adult of *R. maidis* are shown in Table II. Among the 15 insecticides tested, chlorpyrifos was found to be the most toxic to *R. maidis* with the LC_{50} value of 1.03 mg/L, followed by lambda-cypermethrin (1.68 mg/L), pirimicarb (1.72 mg/L), phoxin (1.86 mg/L), profenofos (2.26 mg/L), deltamethrin (2.27 mg/L), methomyl (2.84 mg/L), bifenthrin (3.67 mg/L), dimethoate (4.30 mg/L), beta-cypermethrin (5.06 mg/L), malathion (7.18 mg/L), imidacloprid (8.15 mg/L), acetamiprid (10.19 mg/L), pymetrozine (11.15 mg/L) and abamectin (39.20 mg/L). This result indicated that organophosphate (OP) and pyrethroid insecticides were more toxic to *R. maidis* than neonicotinoid insecticides (imidacloprid and acetamiprid) and abamectin.

Table I.- Susceptible toxicity baselines of 15 insecticides against *A. craccivora* by glass tube residual film method.

Insecticide	N[a]	Intercept	Slope ± SE	χ2	LC_{50}(95%FL[b])(mg/L)	LC_{90}(mg/L)
Imidacloprid	360	-0.91	1.13±0.20	0.47	6.33(3.88-8.82)	86.07
Acetamiprid	360	-1.12	1.27±0.20	0.46	7.58(4.36-10.71)	77.30
Pymetrozine	360	-1.49	1.32±0.20	1.14	13.60(9.64-17.96)	128.13
Methomyl	360	-0.02	1.50±0.21	4.49	1.03(0.78-1.32)	7.39
Pirimicarb	360	-0.82	1.44±0.20	0.70	3.73(2.76-4.82)	28.87
Dimethoate	360	-0.46	1.79±0.22	3.20	1.81(1.42-2.25)	9.38
Profenofos	360	-0.83	1.58±0.21	2.42	3.39(2.56-4.31)	22.00
Malathion	360	-1.34	1.43±0.19	6.37	7.24(3.75-14.72)	48.13
Phoxin	360	-1.36	1.99±0.23	1.33	4.83(3.92-5.90)	21.24
Chlorpyrifos	360	0.21	2.09±0.24	2.92	0.79(0.64-0.96)	3.26
Lambda-cypermethrin	360	-0.43	1.42±0.20	0.76	2.02(1.50-2.62)	16.14
Beta-cypermethrin	360	-1.37	2.01±0.24	4.58	4.81(3.89-5.87)	20.91
Bifenthrin	360	-0.07	1.51±0.22	1.75	1.11(0.76-1.45)	7.82
Deltamethrin	360	-0.09	0.97±0.18	0.87	1.24(0.71-1.79)	25.89
Abamectin	360	-2.08	1.21±0.20	2.43	52.23(37.12-70.53)	594.80

[a] number of adult aphids assayed.
[b] 95% fiducial limits estimated using SPSS10.0 Institute (2000).

Table II.- Susceptible toxicity baselines of 15 insecticides against *R. maidis* by glass tube residual film method

Insecticide	N[a]	Intercept	Slope ± SE	χ2	LC_{50}(95%FL[b])(mg/L)	LC_{90}(mg/L)
Imidacloprid	360	-2.10	2.31±0.28	4.15	8.15(6.61-9.83)	29.28
Acetamiprid	360	-1.22	1.21±0.21	1.48	10.19(7.29-14.08)	116.47
Pymetrozine	360	-1.06	1.01±0.20	1.52	11.15(7.29-16.21)	206.54
Methomyl	360	-0.60	1.33±0.21	1.26	2.84(2.10-3.85)	26.12
Pirimicarb	360	-0.39	1.64±0.22	1.46	1.72(1.29-2.19)	10.47
Dimethoate	360	-0.96	1.52±0.22	1.06	4.30(3.23-5.54)	29.93
Profenofos	360	-0.42	1.18±0.20	0.25	2.26(1.34-3.17)	27.45
Malathion	360	-1.17	1.36±0.22	2.22	7.18(5.38-9.97)	62.71
Phoxin	360	-0.38	1.40±0.21	3.76	1.86(1.21-2.49)	15.38
Chlorpyrifos	360	-0.02	2.13±0.25	0.66	1.03(0.83-1.25)	4.10
Lambda-cypermethrin	360	-0.20	0.90±0.19	0.61	1.68(0.94-2.51)	44.57
Beta-cypermethrin	360	-1.41	2.00±0.25	2.71	5.06(4.06-6.23)	22.15
Bifenthrin	360	-0.83	1.47±0.21	0.17	3.67(2.69-4.76)	27.33
Deltamethrin	360	-0.56	1.59±0.22	0.40	2.27(1.73-2.90)	14.58
Abamectin	360	-1.86	1.17±0.20	0.63	39.20(25.46-54.04)	492.17

[a] number of adult aphids assayed.
[b] 95% fiducial limits estimated using SPSS10.0 Institute (2000).

DISCUSSION

Susceptible toxicity baseline is the basic data required for insecticide resistance monitoring. It also provides guide for resistance management strategy in IPM. In previous works, most resistance monitoring data of insect pests reported were obtained by various bioassays, and the resistance levels were normally evaluated by comparing data tested in different years, only few susceptible strains and baseline data were used. For aphid resistance monitoring, Zhu *et al.* (2000) established the susceptible toxicity baselines of seven organophosphate (OP) insecticides against the greenbug, *Schizaphis graminum* by a residue contact bioassay (glass tube residual film method) for 8h mortality. Liu *et al.* (2001) established the toxicity baselines of four selected aphicides against *Lipaphis erysimi* (Kaltenbach). Lowery *et al.* (2003, 2005) reported the baseline susceptibilities to imidacloprid for *Aphis pomi* De Geer and *Aphis spiraecola* Patch by using a dip bioassay technique. Chen *et al.* (2007) reported a topical application bioassay with 48h mortality to establish the susceptible toxicity baselines of different insecticides against the wheat aphid, *Sitobion avenae* (Fabricius). Han *et al.* (2007) also established the susceptible toxicity baselines of chloronicotinyl insecticides to *S. avenae* by using the dip bioassay. Lu *et al.* (2009) reported a baseline toxicity data of 22 insecticides to *Rhopalosiphum padi* (Linnaeus) and *S. avenae* by the method of residual film in glass tube. No toxicity baseline data of *A. craccivora* and *R. maidis* have been reported. In this study, we determined that LC_{50} values varied from 0.79 to 52.23 mg/L and 1.03 to 39.20 mg/L for *A. craccivora* and *R. maidis*, respectively. Since the LC_{50} values are low the selected susceptible strains can be used with confidence to establish the baseline toxicity data of insecticides.

Comparison with topical application and dip bioassay, which are the generally which are generally used as bioassays for monitoring resistance to aphids, glass tube residual film bioassay is more simple, though not as accurate as topical application bioassay (Huang *et al.*, 2006). Besides bioassay, exposure time to insecticide is another important factor influencing bioassay against aphids (Huang *et al.*, 2006; Lu *et al.*, 2009). Most insecticides gave stable mortality after 48 or 72h (Huang *et al.*, 2006). Considering the characteristics of aphids and control mortality, we chose 12h exposure to score mortality. With this toxicity baseline data of 15 insecticides, monitoring resistance of these two aphids will be easier for resistance management of these pests.

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