

## Resistance of *Spodoptera exigua* to ten insecticides in Shandong, China

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**Abstract** *Spodoptera exigua* (Hübner) collected from three regions (two scallion-producing regions and one ginger-producing region) in Shandong, China and a laboratory colony of Wuhan (WHLC) were evaluated for their susceptibilities to ten insecticides (emamectin benzoate, chlorfenapyr, indoxacarb, spinosyn, tebufenozide, methoxyfenozide, chlorfluazuron, beta-cypermethrin, chlorpyrifos and methomyl) in 2008, 2009 and 2010 using a leaf-dip bioassay method. The results indicate that the resistance ratios of *S. exigua* to newer insecticides such as emamectin benzoate, chlorfenapyr and indoxacarb were all below 20-fold, with no obvious change in all 3 years. *S. exigua* exhibits moderate resistance to spinosyn, and its resistance ratios increased from 1.98–5.31-fold in 2008 to 14.31–64.20-fold in 2010 from three regions as compared with WHLC. *S. exigua* showed moderate to high resistance to insect growth regulators such as tebufenozide, methoxyfenozide, chlorfluazuron in 2008, 2009 and 2010, in which resistance to chlorfluazuron was rapidly increased from 31.49–88.19-fold

in 2008 to 1184.39–2789.67-fold in 2010. Resistance of *S. exigua* to beta-cypermethrin and chlorpyrifos varied greatly among the three regions, ranging from 95.31–437.97-fold and 25.05–40.64-fold in 2008, 951.81–1304.40-fold and 44.91–186.33-fold in 2009, to 27.27–1095.31-fold and 19.12–267.98-fold in 2010. In contrast, *S. exigua* showed low resistance to methomyl, and the resistance ratio was below 5-fold in 3 years. There are several reasons accounting for varying degrees of resistance, including selection pressure, cropping structure and migration, in which the migration of *S. exigua* may play an important role.

**Keywords** Beet armyworm · Beta-cypermethrin · Chlorfenapyr · Chlorfluazuron · Chlorpyrifos · Emamectin benzoate · Indoxacarb · Insecticide susceptibility · Methomyl · Methoxyfenozide · Resistance monitoring · Spinosyn · Tebufenozide

### Introduction

The beet armyworm (*Spodoptera exigua* Hübner), with a distribution within the temperate zone and tropical range, is well known as one of the most destructive agricultural lepidopteran pests worldwide. The beet armyworm is polyphagous, living on a wide range of crops such as cotton, soybean, groundnut, tobacco and vegetables including scallion, ginger and pimiento. Since the pest was found in the 1950s in Shandong province of China, its damage has increased continually. Especially in

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1997 and 1999, *S. exigua* attacked numerous economically important crops and caused great losses to agriculture (Ren and Sun 2000). The reasons for the outbreak were complex. Firstly, it was promoted by the climate of high temperature and drought. For another, *S. exigua* obtained sufficient food thanks to the change of planting structure, particularly due to the increased cultivation of vegetables since the 1990s. Third, the prevention and control of the pest became more difficult because of its developed resistance to chemical insecticides. The problem of resistance is one of the most important reasons for the outbreak (Jiang and Luo 1999; Ren and Sun 2000).

Resistance to insecticides is a major problem associated with the chemical control of insect pests. At present, the extensive use of conventional insecticides such as beta-cypermethrin, chlorpyrifos and methomyl against *S. exigua* has produced prevalent resistance in China, in which the resistance ratio was 1536.0-fold for beta-cypermethrin, 1208.6-fold for chlorpyrifos and 532-fold for methomyl in Taian (Xia et al. 2003). With high resistance to conventional insecticides, the insect growth regulators (IGRs) and newer insecticides were introduced to control *S. exigua* (Zhou et al. 2008). In the case of IGRs, chlorfluazuron was used to control *S. exigua* in Shandong, Jiangsu, Shanghai, Hubei, Fujian and the resistance has been increasing since 1991 (Zhou et al. 2008). Tebufenozide and methoxyfenozide had high toxicity to *S. exigua*, in which resistance to tebufenozide was barely expressed (Liu et al. 2008). In addition, the newer insecticides bearing novel modes of action such as emamectin benzoate, chlorfenapyr, indoxacarb, spinosyn were recently introduced into Shandong province for management of the pests. While the newer insecticides had high toxicity, the resistance to them has received considerable attention, and *S. exigua* was found to have inherent risks for resistance to indoxacarb and spinosyn (Wang et al. 2006, 2008).

In order to supply accurate information for management of resistance and prevent its outbreak in the future, resistance investigations of *S. exigua* were carried out in Shandong province against newer insecticides, including abamectin, pyrrole, oxadiazine and IGRs, as well as conventional insecticides including the organophosphates, carbamates and synthetic pyrethroids.

## Materials and methods

**Insects** A laboratory colony of Wuhan (WHLC): the *S. exigua* susceptible colony was obtained from the Wuhan Institute of Vegetable Science where it had been maintained in a mass rearing environment for 30 years. The field populations: three strains of *S. exigua* from different vegetable crops were collected for insecticide resistance bioassays between July 2008 and September 2010. The samplings from Zhangqiu and Anqiu were concentrated because of their key roles in agriculture, in which Anqiu is the important ginger-producing area and Zhangqiu is the biggest scallion production area. Additionally, the population of Taian was collected for its various vegetables.

All strains were collected as 3rd–5th instar larvae by walking through a 3-ha block of a particular host crop. Larvae were fed in the laboratory on a semi-synthetic diet, slightly modified from Mu et al. (2002). Diet was replaced after 24 h and pupae were collected on alternate days. The emerged adults were kept in cages with meshed sides to maintain ventilation. The adults were fed on a solution containing sucrose (100 g l<sup>-1</sup>) and a vitamin solution (20 ml l<sup>-1</sup>) in a soaked cotton wool ball. The hatched eggs were fed with semi-synthetic diet. All instars of the insects were reared in the laboratory at 27±1°C and 50–75% relative humidity (r.h.) with a 14 h:10 h L:D photoperiod.

**Insecticides** Ten insecticides were used in this study: 90% emamectin benzoate (Hebei Veyong Bio-Chemical Co., Ltd., Hebei, China); 94.5% chlorfenapyr (BASF (China) Co., Ltd., Beijing, China); 94% indoxacarb (E.I. DuPont de Nemours & Co., Inc., Wilmington, DE, USA); 98.2% spinosyn (Dow AgroSciences LLC., Indianapolis, IN, USA); 95% tebufenozide (Dow AgroSciences LLC.); 97.6% methoxyfenozide (Dow AgroSciences LLC.); 90% chlorfluazuron (Ishihara Sangyo Kaisha, Ltd., Osaka, Japan); 95% beta-cypermethrin (Nanjing Redsun Co., Ltd., Nanjing, China); 97% chlorpyrifos (Jinan Luba Chemicals Co., Ltd., Jinan City, China); 98% methomyl (Shandong Huayang Technology Co., Ltd., Ningyang, China).

**Bioassays** Bioassays were conducted on newly 3rd instars larvae of *S. exigua* from the first filial generation of laboratory cultures using a standard leaf-dip bioassay method (Ahmad et al. 2007). Serial dilutions as mg l<sup>-1</sup> of the active ingredient of the test

compounds were prepared using 0.1% Tween-80 in water from Jiangsu Hai'an Petrochemical Plant, Nantong, China. Cabbage leaf discs (1 cm diam) were cut and dipped into the test solutions for 10 s with gentle agitation, and then allowed to dry in between two pieces of paper toweling. At least six concentrations and four replications (20 larvae per replication) were used to estimate each concentration-mortality line. Controls for each insecticide were treated with 0.1% Tween-80 in water only. Before and after treatment, larvae were maintained at a constant temperature of  $27\pm 1^{\circ}\text{C}$  and r.h. of 50–75% with a photoperiod of 14 h: 10 hL:D. Mortality was assessed after 48 h for general insecticides and after 72 h for IGR insecticides. Larvae were considered dead if they were unable to move in a coordinated manner when disturbed with the point of a pencil (Liu and Shen 2003).

**Data analysis** Data were corrected for control mortality using Abbott's (1925) formula before analysis, and data analyzed with SAS/STAT® version 6.12 (SAS Institute Inc. 1997). Statistical differences between LC's were determined using the presence or absence of overlap in the 95% confidence limits.

## Results

### Resistance of four new insecticides in Shandong, China

**Emamectin benzoate** Emamectin benzoate resistance of *S. exigua* (Table 1) was evaluated in Taian, Zhangqiu and Anqiu. Compared with the WHLC, the insect had already become less resistant in 2008, with resistance ratios from 3.77- to 4.93-fold in Taian, Zhangqiu and Anqiu. The resistance ratios in Taian and Zhangqiu dropped slightly from 4.64- and 4.93-fold in 2008 to 3.77- and 1.45-fold in 2009 and then increased to 10.16- and 5.37-fold in 2010. However, no particular trend was discernible in Anqiu in the extent of resistance during the 3 years.

**Chlorfenapyr** *S. exigua* had exhibited very low resistance to chlorfenapyr in 2008 and no resistance in 2009 and 2010 in Taian, Zhangqiu and Anqiu. Table 1 shows a slight decrease in resistance levels to

chlorfenapyr in Anqiu from 2.91-fold in 2008 to 1.03-fold in 2009. However, the resistance level of *S. exigua* populations from Taian and Zhangqiu in 2010 remained at levels similar to those in 2009 and 2008.

**Indoxacarb** Tests with indoxacarb (Table 1) revealed a moderate resistance level in Taian, Zhangqiu and Anqiu in 2008, with resistance ratios of 15.48-, 17.12-, and 12.25-fold, respectively. There was also a moderate resistance level in Taian and Anqiu strains, the resistance ratios being 18.00- and 13.51-fold in 2009. However, the resistance ratio of Zhangqiu was at a low level in 2009. In 2010, the resistance levels in three regions ranged from 8.70- to 11.26-fold, which coincides with the previous year. For Taian and Anqiu strains, the resistance levels toward indoxacarb appeared to be similar from 2008 to 2010, whereas in the Zhangqiu strain the resistance levels fluctuated slightly from 17.12-fold in 2008 to 6.60-fold in 2009, and to 10.54-fold in 2010.

**Spinosyn** Tests with spinosyn (Table 1) revealed great changes in all three populations from 2008 to 2010. The *S. exigua* population from Taian became moderately resistant to spinosyn, and resistance ratios ranged from 5.13-fold in 2008, increasing to 17.82-fold in 2009, and to 44.60-fold in 2010. Similar trends of resistance toward spinosyn were also demonstrated in Zhangqiu (ranging from 2.81-fold in 2008, to 24.30-fold in 2009, to 64.20-fold in 2010). *S. exigua* in Anqiu had a low resistance level in 2008, and increased to moderate resistance levels in 2009 and 2010.

### Resistance of three IGRs in Shandong, China

**Tebufenozide** The data in Table 2 reveal moderate levels of resistance to ecdysone insecticides, such as tebufenozide in 2008 with resistance ratios of 30.11-, 12.58- and 20.10-fold in Taian, Zhangqiu and Anqiu, respectively. The *S. exigua* population in Taian had the highest resistance ratios among the regions in all 3 years, increasing from 30.11-fold in 2008, to 20.94-fold in 2009, to 85.64-fold in 2010. A similar trend of resistance toward tebufenozide was also demonstrated in Zhangqiu. The resistance ratios in Anqiu ranged from 20.10-fold in 2008, to 37.83-fold in 2009 and to 14.93-fold in 2010.

**Table 1** Comparison of resistance of four new insecticides against 3rd instars of *Spodoptera exigua* from three regions in Shandong, China, in 2008, 2009 and 2010

Insecticide	Location	Host	2008			2009			2010		
			Slope±SE	LC <sub>50</sub> (mg/L <sup>-1</sup> ) (95%CL)	RR <sup>z</sup>	Slope±SE	LC <sub>50</sub> (mg/L <sup>-1</sup> ) (95%CL)	RR <sup>z</sup>	Slope±SE	LC <sub>50</sub> (mg/L <sup>-1</sup> ) (95%CL)	RR <sup>z</sup>
Emamectin Benzoate	Taian	Scallion	1.96±0.13	0.16 (0.14–0.18)	4.64	1.93±0.18	0.13 (0.10–0.17)	3.77	1.82±0.08	0.35 (0.33–0.37)	10.16
	Zhangqiu	Scallion	1.71±0.14	0.17 (0.14–0.20)	4.93	1.10±0.06	0.05 (0.04–0.07)	1.45	1.71±0.18	0.19 (0.15–0.23)	5.37
	Anqiu	Ginger	1.73±0.17	0.13 (0.11–0.15)	3.77	1.62±0.08	0.16 (0.14–0.18)	4.64	1.41±0.25	0.12 (0.06–0.24)	3.51
	WHLC <sup>y</sup>	–	–	–	–	1.04±0.18	0.03 (0.03–0.04)	1.00	–	–	–
Chlorfenapyr	Taian	Scallion	1.59±0.10	24.46 (21.08–28.38)	1.98	2.55±0.02	15.67 (15.42–15.84)	1.27	1.42±0.26	17.12 (12.43–23.57)	1.39
	Zhangqiu	Scallion	1.76±0.08	15.38 (13.33–17.74)	1.25	1.92±0.05	6.31 (5.91–6.75)	0.51	1.00±0.05	10.11 (9.05–11.28)	0.82
	Anqiu	Ginger	1.90±0.05	35.86 (33.51–38.38)	2.91	1.51±0.18	12.70 (9.86–16.35)	1.03	1.52±0.18	7.90 (5.90–10.58)	0.64
	WHLC	–	–	–	–	0.50±0.08	12.33 (7.18–21.17)	1.00	–	–	–
Indoxacarb	Taian	Scallion	2.74±0.21	9.10 (7.78–10.65)	15.48	2.94±0.16	10.58 (9.69–11.54)	18.00	1.76±0.03	10.14 (9.73–10.57)	17.26
	Zhangqiu	Scallion	1.56±0.08	10.06 (8.81–11.49)	17.12	2.86±0.09	3.88 (3.61–4.16)	6.60	1.76±0.21	6.19 (4.73–8.11)	10.54
	Anqiu	Ginger	1.82±0.13	7.20 (6.13–8.45)	12.25	1.57±0.27	7.94 (4.87–12.97)	13.51	1.44±0.18	5.11 (3.96–6.60)	8.70
	WHLC	–	–	–	–	0.86±0.22	0.59 (0.28–1.25)	1.00	–	–	–
Spinosyn	Taian	Scallion	1.97±0.16	6.29 (5.30–7.46)	5.13	1.54±0.08	21.86 (20.41–23.42)	17.82	1.29±0.28	54.71 (36.88–81.75)	44.60
	Zhangqiu	Scallion	1.53±0.11	3.45 (2.95–4.04)	2.81	1.52±0.09	29.81 (27.89–31.86)	24.30	0.97±0.21	78.75 (49.28–125.86)	64.20
	Anqiu	Ginger	1.25±0.12	2.43 (1.90–3.12)	1.98	1.50±0.12	20.96 (17.25–25.49)	17.09	0.96±0.14	17.55 (10.82–28.46)	14.31
	WHLC	–	–	–	–	0.94±0.14	1.23 (0.86–1.74)	1.00	–	–	–

<sup>z</sup> Resistance ratio (RR) estimated as RR=LC<sub>50</sub> of field strain in 2008 or 2009 or 2010/LC<sub>50</sub> of WHLC in 2009<sup>y</sup> WHLC: laboratory colony of Wuhan

**Table 2** Comparison of resistance of three insect growth regulators against 3rd instars of *Spodoptera exigua* from three regions in Shandong, China, in 2008, 2009 and 2010

Insecticide	Location	Host	2008			2009			2010		
			Slope±SE	LC <sub>50</sub> (mg.l <sup>-1</sup> ) (95%CL)	RR <sup>z</sup>	Slope±SE	LC <sub>50</sub> (mg.l <sup>-1</sup> ) (95%CL)	RR <sup>z</sup>	Slope±SE	LC <sub>50</sub> (mg.l <sup>-1</sup> ) (95%CL)	RR <sup>z</sup>
Tebufenozide	Taian	Scallion	1.92±0.12	50.36 (43.39–58.46)	30.11	1.30±0.13	34.28 (27.35–42.95)	20.49	0.67±0.08	143.26(143.26–177.22)	85.64
	Zhangqiu	Scallion	1.40±0.05	21.05 (19.12–23.18)	12.58	1.20±0.04	21.50 (19.22–24.05)	12.85	1.11±0.13	114.56 (91.40–143.58)	68.48
	Anqiu	Ginger	1.21±0.14	33.63 (23.08–49.00)	20.10	1.29±0.02	63.29 (62.19–64.41)	37.83	1.36±0.33	24.98 (10.27–60.77)	14.93
	WHLC <sup>y</sup>	–	–	–	–	0.92±0.02	1.67 (1.61–1.74)	1.00	–	–	–
Methoxyfenozide	Taian	Scallion	2.65±0.13	37.73 (34.04–41.82)	191.91	1.51±0.13	111.81(101.30–123.41)	568.72	1.01±0.03	86.87 (80.33–93.95)	441.88
	Zhangqiu	Scallion	1.69±0.09	14.28 (12.52–16.28)	72.63	1.18±0.12	16.70 (11.15–24.99)	84.94	1.22±0.08	88.36 (74.88–104.27)	449.46
	Anqiu	Ginger	1.87±0.17	16.49 (13.52–20.12)	83.88	1.99±0.14	56.63 (52.24–61.38)	288.05	1.76±0.45	27.46 (12.38–60.93)	139.70
	WHLC	–	–	–	–	1.52±0.45	0.20 (0.11–0.37)	1.00	–	–	–
Chlorfluazuron	Taian	Scallion	2.09±0.13	7.41 (6.45–8.52)	36.92	0.77±0.04	122.37(106.66–140.38)	609.72	0.87±0.12	558.55(410.32–760.32)	2783.00
	Zhangqiu	Scallion	1.62±0.08	17.70 (15.36–20.39)	88.19	1.24±0.11	466.59(423.63–513.89)	2324.80	0.67±0.07	559.89(458.53–683.65)	2789.67
	Anqiu	Ginger	1.67±0.09	6.32 (5.27–7.59)	31.49	1.52±0.12	380.17 (337.28–428.53)	1894.20	0.44±0.06	237.71(170.14–332.11)	1184.39
	WHLC	–	–	–	–	0.42±0.01	0.20 (0.18–0.23)	1.00	–	–	–

<sup>z</sup> Resistance ratio (RR) estimated as RR=LC<sub>50</sub> of field strain in 2008 or 2009 or 2010/LC<sub>50</sub> of WHCL in 2009<sup>y</sup> WHLC: laboratory colony of Wuhan

**Table 3** Comparison of resistance of three conventional insecticides against 3rd instars of *Spodoptera exigua* from three regions in Shandong, China, in 2008, 2009 and 2010

Insecticide	Location	Host	2008			2009			2010		
			Slope±SE	LC <sub>50</sub> (mg.l <sup>-1</sup> ) (95%CL)	RR <sup>z</sup>	Slope±SE	LC <sub>50</sub> (mg.l <sup>-1</sup> ) (95%CL)	RR <sup>z</sup>	Slope±SE	LC <sub>50</sub> (mg.l <sup>-1</sup> ) (95%CL)	RR <sup>z</sup>
Beta-cypermethrin	Taian	Scallion	1.81±0.07	62.21 (57.20–67.67)	95.31	2.27±0.30	808.42 (694.86–940.54)	1238.60	2.36±0.38	437.00 (358.79–532.25)	669.52
	Zhangqiu	Scallion	1.74±0.12	285.86 (231.27–353.33)	437.97	1.63±0.11	851.41 (788.09–919.82)	1304.40	1.93±0.39	714.91 (536.17–953.22)	1095.31
	Anqiu	Ginger	1.72±0.08	64.08 (55.59–73.88)	98.18	3.85±0.36	624.51 (553.44–704.70)	956.81	2.27±0.30	17.80 (13.81–22.94)	27.27
Chlorpyrifos	WHLC <sup>y</sup>	–	–	–	–	0.54±0.20	0.65 (0.19–2.20)	1.00	–	–	–
	Taian	Scallion	2.49±0.15	330.80 (293.01–373.47)	40.64	1.29±0.02	1516.82 (1490.32–1543.76)	186.33	1.93±0.13	1433.45 (1291.28–1591.28)	176.09
	Zhangqiu	Scallion	1.73±0.07	203.92 (184.03–225.95)	25.05	0.65±0.03	1179.33 (1091.62–1274.10)	144.87	1.06±0.13	2181.43 (1907.46–2494.76)	267.98
Methomyl	Anqiu	Ginger	1.64±0.07	247.06 (222.68–274.10)	30.35	1.54±0.16	365.57 (320.98–416.36)	44.91	1.58±0.12	155.61 (126.05–192.09)	19.12
	WHLC <sup>y</sup>	–	–	–	–	1.08±0.23	8.14 (6.16–10.76)	1.00	–	–	–
	Taian	Scallion	1.84±0.08	367.40 (332.44–406.04)	2.60	2.04±0.20	361.43 (322.25–405.37)	2.56	2.77±0.62	677.97 (513.89–894.44)	4.80
	Zhangqiu	Scallion	1.84±0.08	183.70 (166.22–203.02)	1.30	2.19±0.27	313.68 (234.03–420.44)	2.22	2.31–0.21	362.33 (300.44–436.96)	2.57
	Anqiu	Ginger	1.72±0.14	236.28 (184.94–301.88)	1.67	1.93±0.18	866.69 (680.66–1103.56)	6.14	1.95±0.23	368.20 (286.88–472.57)	2.61
	WHLC <sup>y</sup>	–	–	–	–	1.07±0.31	141.16 (70.18–283.92)	1.00	–	–	–

<sup>z</sup> Resistance ratio (RR) estimated as RR=LC<sub>50</sub> of field strain in 2008 or 2009 or 2010/LC<sub>50</sub> of WHCL in 2009<sup>y</sup> WHLC: laboratory colony of Wuhan

**Methoxyfenozide** In the case of methoxyfenozide, another ecdysone insecticide just like tebufenozide, *S. exigua* in all three regions revealed moderate to high resistance in the 3 years, with the highest resistance level in Taian. In Zhangqiu, the resistance of *S. exigua* to methoxyfenozide did not change significantly from 2008 to 2009, but increased to 449.46-fold in 2010. In the Anqiu population, the resistance increased from 83.88-fold in 2008 to 288.05-fold in 2009, and then decreased to 139.70-fold in 2010 (Table 2).

**Chlorfluazuron** In 2008, *S. exigua* showed moderate resistance to chlorfluazuron with a resistance ratio ranging from 31.49- to 88.19-fold in the three regions (Table 2). The resistance ratios of *S. exigua* populations from Taian and Zhangqiu exhibited a similar trend, and ranging from 36.92- to 88.19-fold in 2008, increasing to 609.72- to 2324.80-fold in 2009, and to 2783.00- and 2789.67-fold in 2010. However, for the *S. exigua* populations from Anqiu, the resistance was 31.49-fold in 2008, and had a slight fluctuation from 1894.20-fold in 2009 to 1184.39-fold in 2010.

#### Resistance of three conventional insecticides in Shandong, China

**Beta-cypermethrin** *Spodoptera exigua* became highly resistant to beta-cypermethrin in 2008 with resistance ratios ranging from 95.31-, 437.97-, 98.18-fold for the Taian, Zhangqiu, Anqiu populations, respectively (Table 3). The resistance of *S. exigua* varied greatly among the 3 years and regions. The resistance of *S. exigua* in Taian increased from 95.31-fold in 2008 to 1238.60-fold in 2009 and to 669.52-fold in 2010. A similar trend of resistance pattern toward beta-cypermethrin was also demonstrated in Zhangqiu (from 437.97-fold in 2008 to 1304.40-fold in 2009, and to 1095.31-fold in 2010). However, resistance ratios for the *S. exigua* populations from Anqiu increased from 98.18-fold in 2008 to 956.81-fold in 2009, and then significantly decreased to 27.27-fold in 2010.

**Chlorpyrifos** Tests with chlorpyrifos (Table 3) revealed a moderate resistance level in the Taian, Zhangqiu and Anqiu populations in 2008 and resistance ratios were 40.64-, 25.05- and 30.35-fold, respectively. The

resistance ratios of *S. exigua* in Taian increased from 40.64-fold in 2008 to 186.33-fold in 2009, and then did not increase significantly in 2010. The resistance level for *S. exigua* in Taian increased from 25.05-fold in 2008, to 144.87-fold in 2009, to 267.98-fold in 2010. In contrast, the *S. exigua* in Anqiu had a moderate resistance level during the 3 years, ranging from 30.35-fold in 2008, to 44.91-fold in 2009, to 19.12-fold in 2010.

**Methomyl** Although the  $LC_{50}$ s of methomyl to *S. exigua* from all three regions were significantly greater than the WHLC in 2008, resistance ratios were generally low (no more than 3-fold) (Table 3). The resistance ratios of *S. exigua* from Zhangqiu did not change significantly from 2008, 2009 to 2010. The resistance levels for the *S. exigua* populations in Taian did not increase from 2008 to 2009, but increased to 4.80-fold in 2010. In contrast, for the Anqiu population, resistance ratios increased from 1.67-fold in 2008 to 6.14-fold in 2009, and then decreased to 2.61-fold in 2010.

#### Discussion

The present studies, conducted in 2008, 2009 and 2010, demonstrate that the *S. exigua* populations in three regions of Shandong province have shown varying degrees of resistance to four newer insecticides, three insect growth regulators (IGRs) and three conventional insecticides. At present, this pest was found resistant to emamectin benzoate, chlorfenapyr, indoxacarb, spinosyn, tebufenozide, chlorfluazuron, beta-cypermethrin, chlorpyrifos, methomyl (Lan et al. 2005; Liu and Shen 2002; Liu et al. 2004, 2007; Si et al. 2009). This suggests that populations of *S. exigua* have the potential to develop resistance to a wide range of chemicals.

Of all the insecticides evaluated, the newer insecticides with novel modes of action including emamectin benzoate, chlorfenapyr, indoxacarb and spinosyn showed high toxicity to *S. exigua*. In addition, the IGRs including tebufenozide, methoxyfenozide and chlorfluazuron showed varying toxicity to *S. exigua*, in which the toxicity of tebufenozide and methoxyfenozide to the pest was high. Some conventional insecticides that are the most commonly used against

insects including beta-cypermethrin, chlorpyrifos and methomyl had a certain degree of toxicity to *S. exigua*, but the toxicity of those conventional insecticides was the lowest of all ten insecticides. This might be associated with the susceptible target of action in different insecticides.

This study with ten insecticides indicates that some insecticide resistance in *S. exigua* does occur in Shandong, China. However, the resistance levels of *S. exigua* to most insecticides did not show obvious trends from 2008, to 2009 to 2010. The phenomenon in *S. exigua* is often due to more than one reason. The development of insecticide resistance is primarily a result of the selection pressure exerted on sprayed populations increasing the frequency of resistant individuals (Torres-Vila et al. 2002). In three regions, the conventional insecticides were widely used to control several pests including *S. exigua* for many years. A few years ago, the IGRs were seldom used by farmers to control *S. exigua*, as those insecticides need a longer time to achieve their best effect. Recently, farmers increased the application with greater recognition of those insecticides. In Taian and Zhangqiu, farmers used the IGRs more than five times a month and usually rotated with conventional insecticides. However, the IGRs, especially tebufenozide, were forbidden for use in vegetables for export in Anqiu. The low application of newer insecticides is also associated with their high price, which many farmers could not afford. However, this cannot explain why some newer insecticides which are seldom used in fields (such as spinosyn) resulted in moderate resistance. Ahmad et al. (2008) suggested that resistance to newer insecticides is due to a possible cross-resistance mechanism to conventional insecticides. Shen and Wu (1995) suggested that it was practical to rely more on insecticide application history in a field when cross-resistance needs to be determined. However, it is not common practice to mix conventional and newer insecticides to control this pest in the three regions. Hence, we cannot conclude that the cases of resistance to newer insecticides were caused by cross-resistance existing between conventional and newer insecticides.

The difference in cropping patterns among the three regions is likely to play an important role in the revolution of insecticide resistance of *S. exigua*. For the regions of Taian and Zhangqiu, winter wheat and scallion rotation system is the main cropping pattern

and the temperature in winter is usually less than  $-10^{\circ}\text{C}$ , whereas in Anqiu there are lots of winter-warm sheds and vegetables including ginger are the main crops, with the temperature in winter-warm sheds ranging from  $15\text{--}25^{\circ}\text{C}$  in winter. Jiang et al. (2001) indicated that the pupal stage of *S. exigua* was the most possible stage to overwinter and was not able to overwinter in areas where temperatures below  $0^{\circ}\text{C}$  exceed 38 days, so Anqiu but not Taian or Zhangqiu could provide a place for *S. exigua* pupae to survive. The surviving pupae could have an effect on the resistance of *S. exigua* in the following year. In addition, a crucial agroecological component determining the extent to which insecticide resistance may evolve is the proportion of the total population sprayed. For instance, the grass near farmland may act as refugia providing less resistant individuals, which have the potential to dilute insecticide resistance after breeding with resistant individuals, with resistant gene frequencies remaining at an acceptable level for successful control.

However, that selective force and cropping pattern do not explain satisfactorily the differences in level of insecticide resistance observed in all three regions in 2008, 2009 and 2010. Resistance levels that do not fit the agroecological context from which the target population was taken are often tentatively explained through a migratory process (Wu and Guo 1997). *S. exigua* exhibits a substantial long-distance migratory potential on the three continents where this moth occurs (Su 1998). Feng et al. (2003), using radar and a simultaneously operated searchlight trap and ground light-trap at a site in Langfang observed the migration of *S. exigua* in 2002 and indicated that the insect was a high-altitude nocturnal windborne migrant in northern China. In Australia, migration of *H. armigera* is considered a key factor responsible for the year-to-year cyclical variation in resistance frequencies (Daly 1993). That means the migratory potential of *S. exigua* has potential effects on insecticide resistance dynamics. Hence, immigration processes may be a major behavioral factor affecting the evolution of insecticide resistance in *S. exigua*. The great changes of resistance level in some insecticides from 2008 to 2010 may suggest the possibility that the *S. exigua* populations in Shandong were immigration populations.

*Spodoptera exigua* has recently emerged as a serious pest of vegetables, cotton and other crops in Shandong, China. The development of a broad-spectrum resistance



to insecticides has complicated its chemical control. However, the control of *S. exigua* has relied mainly on the application of various insecticides. It is very important to select several effective insecticides to control this pest. From the results of this article, we propose emamectin benzoate, chlorfenapyr, indoxacarb, spinosyn, tebufenozide, and methoxyfenozide as effective insecticides. In order to protect those insecticides and postpone the development of resistance, an integrated pest management strategy should be devised for managing this pest. Prognosis on the basis of light- or pheromone-traps and prevailing meteorological conditions may help in determining better timing of control operations. Slow-release pheromone formulations have shown success for mating disruption. Ma et al. (2003), Chen and Gong (2003) and Zhao et al. (2001) all indicated that the number of accumulated eggs of *S. exigua* in crops was significantly reduced when using slow-release pheromone formulations in the field. Preservation of the parasitoids such as *Cantheconidea furcellata* and *Telenomus* sp. (Lu et al. 2004) is necessary to reduce pesticide applications. At the same time, timely monitoring of changes in resistance of the pest and rotating the proposed chemistries is recommended.

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