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# Regional susceptibilities of *Rhopalosiphum padi* (Hemiptera: Aphididae) to ten insecticides

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#### **Abstract**

Rhopalosiphum padi (L.) (Hemiptera: Aphididae) is one of the most significant cereal pests worldwide. Control of *R. padi* has relied heavily on chemical insecticides. We sampled 12 populations of *R. padi* from 11 provinces in China and analyzed their regional susceptibilities to 10 insecticides by using the leaf dip method. The *R. padi* populations showed susceptibility or minor resistance to chlorpyrifos, malathion, thiamethoxam, beta-cypermethrin, acetamiprid, and pymetrozine, but minor to moderate resistance to bifenthrin, decamethrin, and abamectin. Correlation analysis indicated positive and significant correlations between *R. padi* resistance levels to thiamethoxam and beta-cypermethrin, between *R. padi* resistance levels to chlorpyrifos and 4 other insecticides (decamethrin, abamectin, acetamiprid, and beta-cypermethrin), and between *R. padi* resistance levels to acetamiprid and 3 other insecticides (decamethrin, thiamethoxam, and beta-cypermethrin). Due to the widespread and variable nature of resistance in *R. padi*, we strongly urge rotation of insecticide classes to delay the onset of high levels of resistance.

Key Words: bird cherry-oat aphid; insecticide resistance; susceptibility

#### Resumen

Rhopalosiphum padi (L.) (Hemiptera: Aphididae) es una de las plagas de cereales más significativas en todo el mundo. El control de *R. padi* ha dependido en gran medida de los insecticidas químicos. Se recogieron muestras de 12 poblaciones de campo de *R. padi* de varias provincias de China y se analizaron las susceptibilidades regionales de las poblaciones de campo a 10 insecticidas mediante el método de inmersión foliar. Las poblaciones de *R. padi* mostraron susceptibilidad o resistencia menor al clorpirifos, malatión, tiametoxam, beta-cipermetrina, acetamiprid, y pimetrozina, pero menor resistencia a moderada a la bifentrina, decametrin, y abamectina. El análisis de correlación indicó correlaciones positivas y significativas entre los niveles de resistencia de *R. padi* al clorpirifos y otros 4 insecticidas (decametrin, abamectina, acetamiprid, y beta-cipermetrina), así como los niveles de resistencia de *R. padi* entre acetamiprid y otros 3 insecticidas (decametrin, tiametoxam, y beta-cipermetrina). Debido a la naturaleza generalizada y variable de la resistencia en *R. padi*, instamos firmemente a la rotación de las clases de insecticidas para retrasar la aparición de altos niveles de resistencia.

Palabras Clave: áfido pájaro de cereza de avena; resistencia a los insecticidas; susceptibilidades regionales

Rhopalosiphum padi (L.) (Hemiptera: Aphididae) is a significant wheat pest that causes damage through direct feeding and transmission of plant viruses (Schliephake et al. 2013). This aphid species is distributed worldwide and is responsible for major economic losses. In China, *R. padi* has become the most frequent pest species on wheat and is abundant throughout all developmental stages of the crop (Parizoto et al. 2013). Damage caused by *R. padi* is increasing annually in wheat-growing regions of China, particularly in southern areas (Cao 2006; Zhan et al. 2007). Due to the lack of crop cultivars resistant to cereal aphids, the primary approach used to control these pests has been insecticide application (Ou et al. 2005). Various types of insecticides, including organophosphates, carbamates, pyrethroids, macrocyclic lactones, neonicotinoids, and the pyridine azomethine have been used to control *R. padi* in China.

Insecticide resistance is a major cause of insect pest control failure; multiple treatments and excessive doses of insecticide have resulted in insect resistance to several insecticides and have raised serious human health and environmental concerns (Chang et al. 2010). Resistance

monitoring is an effective component of a resistance management approach as it provides valuable information on the resistance of insect populations to insecticides. The susceptibility of *R. padi* to insecticides has rarely been investigated in China. We collected samples from 12 *R. padi* field populations from 11 provinces in China and tested their susceptibility to 10 insecticides. Lethal concentration (LC50) values were determined for each product, and correlation analyses were conducted. Our objective in this study was to provide practical data for insecticide resistance management and sustainable control of *R. padi*.

## **Materials and Methods**

# **INSECT SAMPLING**

Samples from 12 *R. padi* populations were collected from wheat fields in 2013 (Fig. 1). The apterous aphids collected in the field were

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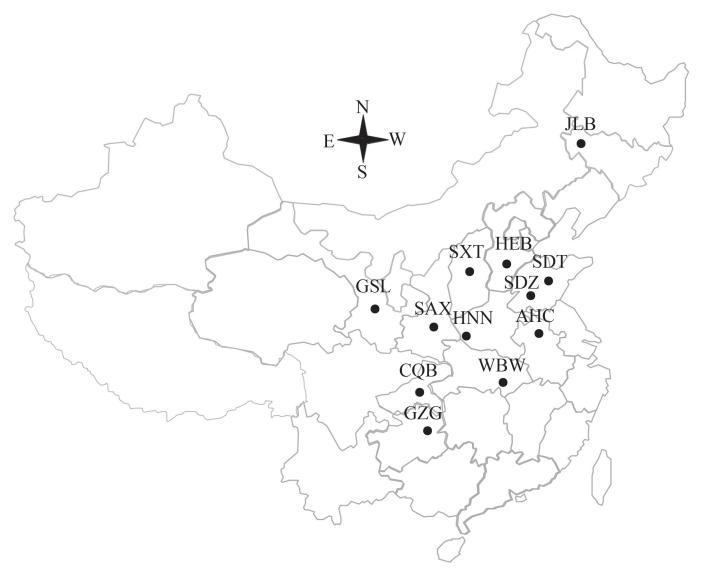


Fig. 1. Sampling regions of *Rhopalosiphum padi* in China. The regions included Baicheng of Jilin Province (the population code was named as JLB), Baoding of Hebei Province (HEB), Lanzhou of Gansu Province (GSL), Taigu of Shanxi Province (SXT), Zibo of Shangdong Province (SDZ), Taian of Shangdong Province (SDT), Xianyang of Shaaxi Province (SAX), Nanyang of Henan Province (HNN), Chuzhou of Anhui Province (AHC), Wuhan of Hubei Province (HBW), Beibei of Chongqing Province (CQB), and Guiyang of Guizhou Province (GZG).

taken to the laboratory and reared on wheat seedlings in cages (41 cm L  $\times$  41 cm W  $\times$  41 cm H). First-generation (F1) apterous adult aphids from each population were used for the bioassay. The relatively susceptible clone Rp-SS had been reared in the laboratory for >3 yr without exposure to any insecticide. Both the field populations and Rp-SS strain were maintained on wheat plants in a rearing room (22  $\pm$  1 °C with a 16:8 h L:D photoperiod and relative humidity >80%).

#### **INSECTICIDES**

The susceptibility of *R. padi* adults to 10 insecticides from 5 chemical classes was tested using analytical-grade insecticides. The products and their sources are shown in Table 1.

#### INSECTICIDE BIOASSAYS

A leaf dip method (Liu et al. 2001; Han et al. 2007) was used for the insecticide bioassays. Serial dilutions of the active ingredients from the test insecticides were prepared using 0.1% Triton X-100 in water. Wheat leaves containing apterous adult aphids were dipped in the insecticide dilutions for 10 s each. Then, the leaves were removed from the solution, and residual droplets on the leaves were adsorbed with clean, dry filter paper. Three replicates of 30 to 50 aphids were used for each insecticide concentration, and 5 or 6 serial concentrations were used for each insecticide. Leaves dipped in 0.1% Triton X-100 were used as a control. The aphids were kept at a constant temperature of  $25 \pm 1$  °C under a 16:8 h L:D photoperiod during and after treatment.

#### STATISTICAL ANALYSES

Mortality was scored 24 h after the aphids had been immersed in the test solutions. Aphids were considered dead if they did not move after gentle prodding. The data were corrected for mortality in the controls using Abbott's formula (Abbott 1925), and mortality responses at different concentrations, LC50 values, 95% confidence intervals (CIs), and slopes were calculated by probit analysis using SPSS software (IBM SPSS Statistics for Windows version 20.0.; IBM Corp., Armonk,

Table 1. Insecticides evaluated in this study.

Group	Insecticide	Purity (%)	Producer				
Organophosphates	chlorpyrifos	96	Tsingtao Meidelong Chemical Co., Ltd., Shandong, China				
	malathion	95	Tianjin Aigefu Co., Ltd., Tianjin, China				
Pyrethroids	bifenthrin	98	Nanjing Red Sun Co., Ltd., Jiangsu, China				
	beta-cypermethrin	96	Yancheng Nongbo Bio-technology Co., Ltd., Jiangsu, China				
	decamethrin	99	Nanjing Red Sun Co., Ltd., Jiangsu, China				
Macrocyclic lactone	abamectin	92	Shandong Sino-Agri United Biotechnology Co., Ltd., Shandong, China				
Neonicotinoids	imidacloprid	97	Shandong Sheda Crop Science Co., Ltd., Shandong, China				
	thiamethoxam	96	Shandong Sino-Agri United Biotechnology Co., Ltd., Shandong, China				
	acetamiprid	96	Shandong Sheda Crop Science Co., Ltd., Shandong, China				
Pyridine azomethine	pymetrozine	97	Rudong County Lianfeng Chemical Industry Co., Ltd., Jiangsu, China				

New York). Bioassays in which the control mortality rate exceeded 10% were discarded and repeated. LC50 values were calculated, and the 2 compared values were considered significantly different if their respective 95% CIs did not overlap (Litchfield & Wilcoxon 1949; Wolfe & Hanley 2002). Resistance ratios (RR) were calculated by dividing the LC50 value for each field population by that for the Rp-SS strain. We classified the resistance level as susceptible (RR  $\leq$  3), minor resistance (3 < RR  $\leq$  5), low resistance (5 < RR  $\leq$  10), moderate resistance (10 < RR  $\leq$  40), high resistance (40 < RR  $\leq$  160), and extremely high resistance (RR > 160-fold) based on the standards described by Shen & Wu (1995). Pairwise correlation coefficients of the log LC50 values for the field populations and each insecticide were calculated using Pearson's correlation analysis and SPSS software (IBM Corp.) to interpret crossresistance spectra among the insecticides tested.

### Results

#### EFFECTS OF THE INSECTICIDES ON THE LABORATORY STRAIN

The relatively susceptible strain Rp-SS showed the lowest LC50 values for all insecticides, except for chlorpyrifos and acetamiprid (Table 2). For chlorpyrifos, the GZG population showed the lowest LC50 value, whereas with acetamiprid, the SXT strain showed the lowest LC50 value. Among the 10 insecticides tested, bifenthrin was found to be the most toxic to the susceptible strain of *R. padi* with an LC50 value of 0.29 mg/L, followed by decamethrin (0.46 mg/L), imidacloprid (0.73 mg/L), thiamethoxam (0.74 mg/L), acetamiprid (0.98 mg/L), malathion (1.06 mg/L), beta-cypermethrin (1.10 mg/L), abamectin (1.11 mg/L), chlorpyrifos (1.13 mg/L), and pymetrozine (10.76 mg/L). Thus, the pyrethroids, neonicotinoids, organophosphates, and abamectin insecticides were more toxic than pymetrozine to the laboratory strain of *R. padi*.

# SUSCEPTIBILITY OF THE FIELD POPULATIONS TO 10 INSECTICIDES

Twelve *R. padi* populations sampled from 11 provinces were examined for their susceptibility to 10 insecticides as compared with that of the laboratory-maintained strain Rp-SS (Table 2). Four populations (SXT, GZG, HEB, and AHC) were susceptible to chlorpyrifos, and the SDT population showed low resistance to it, whereas the others had minor resistance (Table 2). However, the resistance level to chlorpyrifos in SXT, GZG, and HEB populations was less than that of the Rp-SS strain. The SAX, CQB, SDZ, and HBW populations were susceptible to malathion, the HEB, AHC, GSL, SDT, and JLB populations showed minor

resistance to malathion, and the other 2 populations (SXT and GZG) displayed low resistance to malathion.

The SDT and JLB populations had low levels of resistance to acetamiprid. Seven populations (SXT, GZG, AHC, SAX, CQB, HNN, and HBW) were susceptible to acetamiprid, and the remaining 3 populations (HEB, GSL, and SDZ) showed minor resistance to acetamiprid. The GZG, GSL, HNN, and CQB populations were susceptible to thiamethoxam, and the other populations showed minor resistance to thiamethoxam, except for the JLB population, which displayed a low level of resistance to this insecticide. In addition, minor levels of resistance to imidacloprid were found in the SXT, SAX, CQB, and HUB populations, whereas the other populations such as GZG, HEB, AHC, HNN, and SDT were susceptible to imidacloprid. The field-sampled aphid populations displayed less than moderate resistance for all the neonicotinoids tested, except in the case of the SDZ population. Additionally, the JLB population showed low resistance to 3 neonicotinoids.

Three field populations (HNN, SDZ, and JLB) showed low resistance to beta-cypermethrin, 5 populations (SXT, GZG, AHC, SAX, and HBW) were susceptible, and 4 populations (HEB, GSL, SDT, and CQB) showed minor resistance to beta-cypermethrin. No population was susceptible to bifenthrin, with 2 populations (SXT and HEB) showing moderate resistance, 5 populations (GZG, AHC, SAX, GSL, and SDT) showing minor resistance, and 5 populations (HNN, CQB, SDZ, JLB, and HBW) showing low resistance to this insecticide. Four populations (GSL, SDT, SDZ, and JLB) were moderately resistant to decamethrin, 4 populations (HEB, AHC, SAX, and HBW) showed low resistance, 2 populations (SXT and HNN) showed minor resistance, and 2 populations (GZG and CQB) were susceptible to this pyrethroid.

Our bioassay results showed that 4 populations (SXT, GZG, HEB, and GSL) were susceptible to abamectin, whereas 2 populations (CQB and HNN) showed moderate resistance, 3 populations (AHC, JLB, and HBW) showed low resistance, and 3 populations (SAX, SDT, and SDZ) showed minor resistance to abamectin. All field populations were susceptible to pymetrozinine, except for 2 populations (HEB and SAX) that showed minor resistance to this insecticide.

#### CORRELATIONS BETWEEN THE LC50 INSECTICIDE VALUES

Paired comparisons of the log LC50 values for the insecticides tested showed positive and significant correlations between  $R.\ padi$  resistance levels to chlorpyrifos and 2 other insecticides (decamethrin and abamectin) (P < 0.05), between  $R.\ padi$  resistance levels to acetamiprid and 2 other insecticides (decamethrin and thiamethoxam), and between  $R.\ padi$  resistance levels to beta-cypermethrin and thiamethoxam (Table 3). There also was a highly positive correlation between each pair of the 3 insecticides: chlorpyrifos, acetamiprid, and beta-cy-

**Table 2.** Susceptibility of 12 R. padi field populations collected in China to 10 insecticides.

Insecticide	Location	n	Slope ± SE	X2(df)	LC50	95% CLa	RRb
Chlorpyrifos	Rp-SS	623	3.29 ± 0.25	1.45(3)	1.13	0.98-1.28	1.0
	SXT	710	$2.20 \pm 0.19$	3.28(4)	0.45	0.40-0.50	0.4
	GZG	722	2.60 ± 0.24	5.82(4)	0.39	0.35-0.43	0.3
	HEB	723	2.08 ± 0.14	7.01(4)	0.63	0.56-0.73	0.6
	AHC	687	1.65 ± 0.15	3.80(3)	4.05	3.35-5.14	3.6
	SAX	641	2.36 ± 0.17	3.32(3)	2.81	2.48-3.18	2.5
	GSL	690	2.19 ± 0.25	7.89(3)	4.04	3.48-4.58	3.6
	HNN	642	2.93 ± 0.36	1.91(3)	4.74	4.02-5.33	4.2
	SDT	653	3.26 ± 0.25	7.55(3)	5.68	5.14-6.35	5.0
	CQB	592	2.53 ± 0.19	5.30(3)	3.57	3.13-4.14	3.1
	SDZ	620	2.36 ± 0.19	0.95(3)	4.54	3.99-5.26	4.0
	JLB	596	2.87 ± 0.21	4.57(3)	4.70	4.20-5.31	4.1
	HBW	616	2.43 ± 0.18	1.57(3)	3.07	2.72-3.49	2.7
Malathion	Rp-SS	618	2.38 ± 0.18	2.82(3)	1.06	0.94-1.21	1.0
	SXT	713	1.98 ± 0.23	2.56(3)	5.91	5.22-6.82	5.6
	GZG	746	1.99 ± 0.22	2.98(3)	6.07	5.34-6.91	5.7
	HEB	730	3.06 ± 0.29	2.13(4)	3.95	3.64-4.32	3.7
	AHC	644	4.28 ± 0.33	10.66(3)	4.02	3.75-4.30	3.8
	SAX	637	2.59 ± 0.19	1.09(3)	2.48	2.21-2.79	2.3
	GSL	629	2.93 ± 0.37	3.74(3)	4.08	3.30-4.69	3.8
	HNN	645	4.45 ± 0.40	5.22(3)	5.48	5.02-5.89	5.2
	SDT	619	2.70 ± 0.20	9.75(3)	4.48	4.01-5.00	4.2
	CQB	594	2.25 ± 0.18	11.69(3)	2.84	2.18-3.83	2.7
	SDZ	638	1.68 ± 0.15	1.27(3)	2.62	2.23-3.10	2.5
	JLB	601	2.72 ± 0.20	4.42(3)	4.89	4.35–5.56	4.6
	HBW	621	2.42 ± 0.17	0.73(3)	3.00	2.64-3.41	2.8
Acetamiprid	Rp-SS	589	3.57 ± 0.36	3.44(3)	0.98	0.90-1.06	1.0
octampria	SXT	760	2.42 ± 0.24	3.58(4)	0.40	0.36-0.46	0.4
	GZG	746	$2.59 \pm 0.33$	4.64(4)	0.56	0.47-0.72	0.6
	HEB	768	1.71 ± 0.17	3.11(4)	3.24	2.31–5.21	3.3
	AHC	706	1.83 ± 0.15	1.32(3)	1.73	1.50-2.00	1.8
	SAX	706	1.56 ± 0.14	2.91(3)	2.64	2.24–3.18	2.7
	GSL	656	1.95 ± 0.16	2.77(3)	3.16	2.74–3.70	3.2
	HNN	606	1.83 ± 0.16	1.22(3)	2.90	2.44–3.40	3.0
	SDT	607	2.30 ± 0.20	16.66(3)	5.51	3.88-7.43	5.6
	CQB	605		2.12(3)	2.74	2.30–3.21	2.8
	SDZ	616	1.84 ± 0.16 2.16 ± 0.17	0.59(3)	3.04	2.66–3.50	3.1
	JLB	620	$2.30 \pm 0.17$ $2.30 \pm 0.18$	1.46(3)	5.11	4.42-5.83	5.2
	HBW	660	2.06 ± 0.16	8.62(3)	1.95	1.69-2.23	2.0
midaalaarid			2.20 ± 0.18				
nidacloprid	Rp-SS	606		0.58(3)	0.73	0.63-0.86	1.0
	SXT GZG	841	1.66 ± 0.13	6.08(4)	2.32	2.02-2.67	3.2
		701	2.08 ± 0.15	0.80(4)	1.86	1.62-2.12	2.5
	HEB	615	2.27 ± 0.18	0.15(3)	2.08	1.82-2.38	2.8
	AHC	762	1.59 ± 0.14	6.62(3)	1.90	1.63-2.21	2.6
	SAX	690	1.58 ± 0.15	1.41(3)	3.61	3.00-4.55	4.9
	GSL	626	1.67 ± 0.15	5.63(3)	2.30	1.92-2.71	3.1
	HNN	637	2.47 ± 0.19	0.54(3)	1.90	1.66-2.15	2.6
	SDT	606	1.99 ± 0.17	4.55(3)	1.81	1.51-2.12	2.5
	CQB	586	2.24 ± 0.18	4.56(3)	3.41	2.98–3.93	4.7
	SDZ	620	3.59 ± 0.30	2.67(3)	9.98	8.82-11.05	13.6
	JLB	596	2.13 ± 0.17	1.60(3)	6.56	5.56–7.57	8.9
	HBW	612	1.85 ± 0.15	0.86(3)	2.25	1.92-2.62	3.1
hiamethoxam	Rp-SS	585	$3.52 \pm 0.33$	5.53(3)	0.74	0.66-0.81	1.0
	SXT	835	$1.32 \pm 0.11$	4.00(4)	2.35	1.96-2.92	3.2
	GZG	684	$2.04 \pm 0.24$	3.31(3)	1.42	1.26-1.63	1.9
	HEB	754	2.35 ± 0.22	3.36(4)	3.49	2.81-4.40	4.7
	AHC	734	1.52 ± 0.14	3.73(3)	2.76	2.33-3.34	3.7

aCL, confidence limit; bRR, resistance ratio.

Table 2. (Continued) Susceptibility of 12 *R. padi* field populations collected in China to 10 insecticides.

Insecticide	Location	n	Slope ± SE	X2(df)	LC50	95% CLa	RRb
	SAX	689	1.72 ± 0.14	3.14(3)	2.36	2.02-2.77	3.2
	GSL	658	$1.89 \pm 0.14$	8.43(4)	1.49	1.20-1.78	2.0
	HNN	593	$2.60 \pm 0.20$	1.37(3)	2.04	1.79-2.30	2.8
	SDT	621	$2.64 \pm 0.20$	2.29(3)	2.29	2.02-2.58	3.1
	CQB	590	2.04 ± 0.17	3.51(3)	2.24	1.91-2.59	3.0
	SDZ	612	$2.34 \pm 0.19$	8.97(3)	2.12	1.84-2.42	2.9
	JLB	615	2.18 ± 0.17	0.49(3)	5.47	4.74-6.24	7.4
	HBW	576	2.21 ± 0.18	8.22(3)	2.33	2.02-2.67	3.1
Beta-cypermethrin	Rp-SS	625	4.90 ± 2.36	5.02(3)	1.10	0.97-1.25	1.0
	SXT	659	1.80 ± 0.15	4.26(3)	1.14	0.97-1.37	1.0
	GZG	658	2.06 ± 0.17	2.12(3)	1.66	1.40-2.07	1.5
	HEB	762	1.45 ± 0.13	3.53(4)	5.01	3.87-7.04	4.6
	AHC	735	$1.23 \pm 0.13$	3.78(3)	3.03	2.47-3.84	2.8
	SAX	629	1.67 ± 0.15	2.09(3)	3.25	2.76-3.90	3.0
	GSL	652	2.08 ± 0.17	2.22(3)	5.34	4.55-6.48	4.8
	HNN	618	2.07 ± 0.17	0.89(3)	6.78	5.80-8.16	6.2
	SDT	618	2.11 ± 0.18	1.87(3)	5.51	4.78-6.48	5.0
	CQB	581	1.84 ± 0.16	2.65(3)	5.52	4.69-6.63	5.0
	SDZ	577	1.97 ± 0.17	3.48(3)	6.33	5.39-7.65	5.7
	JLB	566	1.75 ± 0.18	0.39(3)	8.15	6.62-10.7	7.4
	HBW	736	1.90 ± 0.15	5.17(4)	1.78	1.53-2.05	1.6
Bifenthrin	Rp-SS	623	2.21 ± 0.17	0.75(3)	0.29	0.24-0.34	1.0
	SXT	605	1.52 ± 0.15	0.15(3)	5.39	3.92-8.49	18.2
	GZG	639	1.75 ± 0.15	4.91(3)	1.13	0.91-0.36	3.8
	HEB	801	2.31 ± 0.24	2.21(4)	3.15	2.83–3.47	10.7
	AHC	670	1.77 ± 0.14	7.86(4)	1.07	0.86-1.28	3.6
	SAX	684	1.80 ± 0.15	2.98(3)	1.18	0.98-1.38	4.0
	GSL	651	1.30 ± 0.15	4.45(3)	1.32	0.99-1.64	4.5
	HNN	616	2.11 ± 0.17	0.30(3)	1.73	1.48-2.00	5.9
	SDT	569	1.92 ± 0.17	1.16(3)	1.00	0.82-1.17	3.4
	CQB	634	2.29 ± 0.19	2.70(3)	2.01	1.67-2.77	6.8
	SDZ	583	$2.60 \pm 0.19$	4.75(3)	2.34	2.07–2.65	7.9
	JLB	610	2.60 ± 0.20	1.53(3)	2.72	2.38–3.08	9.2
	HBW	616	2.11 ± 0.17	0.30(3)	1.73	1.48-2.00	5.9
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Decamethrin	Rp-SS	596	2.10 ± 0.17	2.35(3)	0.46	0.40-0.53	1.0
	SXT	652	1.44 ± 0.23	1.74(3)	2.00	1.69-2.37	4.3
	GZG	694	1.61 ± 0.23	1.27(3)	0.48	0.37-0.57	1.0
	HEB	619	1.23 ± 0.14	1.31(3)	3.49	2.81–4.40	7.5
	AHC	685 651	$1.13 \pm 0.13$	1.24(3)	4.35	3.48-5.66 1.71-2.27	9.4 5.6
	SAX GSL	651 631	1.64 ± 0.15	0.43(3)	2.61 6.01	1.71–2.37	5.6 12.0
		621 632	1.74 ± 0.16	0.48(3)		4.96–7.61	12.9 3.3
	HNN		2.21 ± 0.17	3.76(3) 5.06(3)	1.51	1.29–1.74	3.3 10.2
	SDT CQB	593 634	1.72 ± 0.17	5.06(3) 0.71(3)	4.73 1.20	3.86–6.07 0.98–1.42	2.6
	SDZ	607	1.95 ± 0.17	0.71(3) 5.82(3)	6.08	0.98-1.42 5.24-7.24	13.1
	JLB	598	2.17 ± 0.18 2.03 ± 0.20	1.20(3)	5.35	4.43-6.83	11.5
	HBW	629	1.61 ± 0.15	8.76(3)	3.73	3.13-4.57	8.0
Abamectin	Rp-SS	589	1.79 ± 0.16	4.93(3)	1.11	0.97-1.25	1.0
	SXT	669	1.56 ± 0.14	1.02(3)	2.00	1.69-2.37	1.8
	GZG	677	1.72 ± 0.15	3.33(3)	1.94	1.66-2.26	1.8
	HEB	862	1.47 ± 0.11	2.61(4)	2.22	1.87-2.59	2.0
	AHC	642	1.47 ± 0.17	3.39(3)	7.81	6.22–10.50	7.1
	SAX	662	2.05 ± 0.16	10.72(3)	5.55	4.85–6.35	5.0
	GSL	602	1.37 ± 0.15	3.56(3)	2.95	2.26–3.64	2.7
	HNN	620	1.89 ± 0.16	1.41(3)	11.4	9.83–13.37	10.4
	SDT	630	2.37 ± 0.18	1.13(3)	5.17	4.56–5.877	4.7
	CQB	597	$2.18 \pm 0.18$	2.30(3)	13.2	11.40-15.97	12.1

aCL, confidence limit; bRR, resistance ratio.

Table 2. (Continued) Susceptibility of 12 R. padi field populations collected in China to 10 insecticides.

Insecticide	Location	n	Slope ± SE	X2(df)	LC50	95% CLa	RRb
	SDZ	643	2.15 ± 0.18	1.24(3)	3.64	3.09-4.20	3.3
	JLB	612	2.17 ± 0.18	3.47(3)	7.47	6.44-8.59	6.8
	HBW	641	$2.45 \pm 0.18$	4.59(3)	7.77	6.79-8.78	7.1
Pymetrozine	Rp-SS	611	$2.19 \pm 0.17$	2.43(3)	10.76	9.36-12.39	1.0
	SXT	655	2.18 ± 0.26	6.66(3)	14.86	13.11-16.76	1.4
	GZG	737	2.34 ± 0.25	1.32(3)	18.33	16.49-20.69	1.7
	HEB	872	$3.37 \pm 0.33$	6.94(4)	33.81	29.77-40.43	3.1
	AHC	622	$1.18 \pm 0.14$	2.24(3)	24.67	19.51-33.25	2.3
	SAX	666	$1.54 \pm 0.15$	4.77(3)	34.51	28.66-43.34	3.2
	GSL	593	$1.46 \pm 0.15$	1.03(3)	11.45	38.89-14.04	1.1
	HNN	643	$1.44 \pm 0.14$	3.99(3)	25.62	21.34-31.36	2.4
	SDT	610	$1.49 \pm 0.16$	5.51(3)	30.50	25.06-38.59	2.8
	CQB	607	$1.84 \pm 0.16$	5.76(3)	17.17	14.63-20.01	1.6
	SDZ	579	$2.03 \pm 0.19$	1.38(3)	20.03	17.09-23.52	1.9
	JLB	637	$1.99 \pm 0.17$	7.51(3)	18.49	15.94-21.38	1.7
	HBW	616	1.82 ± 0.17	1.81(3)	24.73	21.06-29.45	2.3

aCL, confidence limit; bRR, resistance ratio.

permethrin (P < 0.01). Additionally, a slightly negative correlation was found between chlorpyrifos and bifenthrin, which can be exploited in resistance management. However, there was a lack of significant correlation for malathion, pymetrozine, and imidacloprid in populations of R. padi from China.

# **Discussion**

Although resistance to insecticides has been reported in laboratory strains of *R. padi* (Chen et al. 2007), this is the first study to collect *R. padi* field populations from numerous geographic sites and to investigate the susceptibility and resistance of the insect to 10 insecticides used in China. Our results showed that the *R. padi* field populations had varying degrees of resistance to the insecticides.

Organophosphate insecticides have been used to control aphids for several decades in China (Chen et al. 2007; Lu & Gao 2009), and organophosphate resistance in laboratory strains of *R. padi* has been reported (Chen et al. 2007). Neonicotinoid insecticides have been widely used more recently to control wheat aphids in China, whereas organophosphates have been nearly abandoned, suggesting that selection pressure by organophosphates on field populations of wheat aphids is not strong. As a consequence of the high fitness cost of maintaining resistance, and rapid reproduction of aphids, the resistance levels to organophosphates could decline rapidly after a few generations in the

absence of exposure to the insecticides (Low et al. 2013), which may explain the low levels of resistance in *R. padi* populations to malathion and chlorpyrifos; these organophosphates were used frequently to control the pest for a long time and are nearly abandoned now.

As neonicotinoids have been applied widely to manage wheat aphids in recent years, aphids seem to be developing resistance to these insecticides (Shi et al. 2011). In this study, the SDT population had low resistance to acetamiprid, and the JLB population had low resistance to acetamiprid and thiamethoxam. These results suggest that field populations of R. padi have the potential to develop resistance to neonicotinoids. We also detected a correlation between acetamiprid and thiamethoxam. Cross-resistance between imidacloprid and thiamethoxam has been confirmed for Bemisia tabaci (Gennadius) (Hemiptera: Aleyrodidae) in southern Spain under field conditions (Elbert & Nauen 2000) and for Nilaparvata lugens (Stål) (Hemiptera: Delphacidae) in China (Liu et al. 2010). Similarly, Foster et al. (2008) reported that Myzus persicae (Sulzer) (Aphidomorpha: Aphididae) is crossresistant to 4 neonicotinoid compounds: thiamethoxam, thiacloprid, clothianidin, and dinotefuran. These reports suggest the possibility of cross-resistance to these chemicals (Ahmad et al. 2007). To maintain the effectiveness of neonicotinoids and postpone the development of resistance in R. padi, rotating the application of neonicotinoids and other insecticides would be a productive strategy.

Pyrethroids have been used to control aphids for several decades in China (Wei et al. 1990; Shuai & Wang 2005). We did not find high

Table 3. Pairwise correlation coefficients for log LC50 values of the insecticides tested on populations of R. padi collected in China.

Insecticide	Chlorpyrifos	Malathion	Acetamiprid	Imidacloprid	Thiamethoxam	Beta-cypermethrin	Bifenthrin	Decamethrin	Abamectin
Malathion	-0.05 <sup>ns</sup>								
Acetamiprid	0.75**	0.01 <sup>ns</sup>							
Imidacloprid	0.38 <sup>ns</sup>	-0.14 <sup>ns</sup>	0.35 <sup>ns</sup>						
Thiamethoxam	0.26 <sup>ns</sup>	0.27 <sup>ns</sup>	0.56*	0.40 <sup>ns</sup>					
Beta-cypermethrin	0.73**	0.13 <sup>ns</sup>	0.84**	0.54 <sup>ns</sup>	0.55*				
Bifenthrin	-0.3 <sup>ns</sup>	0.45 <sup>ns</sup>	-0.11 <sup>ns</sup>	0.24 <sup>ns</sup>	0.42 <sup>ns</sup>	0.04 <sup>ns</sup>			
Decamethrin	0.61*	-0.02 <sup>ns</sup>	0.63*	0.55 <sup>ns</sup>	0.44 <sup>ns</sup>	0.53 <sup>ns</sup>	$0.07^{\text{ns}}$		
Abamectin	0.58*	0.01 <sup>ns</sup>	0.30 <sup>ns</sup>	0.07 <sup>ns</sup>	0.26 <sup>ns</sup>	0.45 <sup>ns</sup>	-0.11 <sup>ns</sup>	-0.06 <sup>ns</sup>	
Pymetrozine	0.16 <sup>ns</sup>	0.02 <sup>ns</sup>	0.38 <sup>ns</sup>	-0.04 <sup>ns</sup>	0.31 <sup>ns</sup>	0.17 <sup>ns</sup>	-0.06 <sup>ns</sup>	0.12 <sup>ns</sup>	0.18 <sup>ns</sup>

Superscripts represent significance of the regression.  $^{ns}$  shows that there is no significant correlation between log LC50 values of insecticides. \* indicates significant correlation between log LC50 values of insecticides (P < 0.05). \*\* indicates highly significant correlation between log LC50 values of insecticides (P < 0.05).

resistance (40 < RR ≤ 160) among the R. padi populations to the 3 pyrethroids tested in this study. However, a positive correlation was observed between the log LC50 values for beta-cypermethrin and those for the 2 neonicotinoids (acetamiprid and thiamethoxam), suggesting that the efficiency of beta-cypermethrin decreased due to cross-resistance to neonicotinoids. In contrast to our results, the log LC50 values of bifenthrin and deltamethrin to Spodoptera litura F. (Lepidoptera: Noctuidae) in Hunan were found to be significantly correlated (Tong et al. 2013); this difference may be due to diverse resistance mechanisms in different insect species. In contrast, previous studies reported that enhanced activity of metabolic enzymes, including cytochrome P450 monooxygenases, esterases, and glutathione S-transferases, was the main reason for the development of metabolic resistance in insects to insecticides (Devonshire & Moores 1982; Puinean et al. 2010; Silva et al. 2012). These metabolic enzymes have isoenzymes, all of which use a range of substrates that may play different roles in insect detoxification under the stress of different chemicals in the same insecticide class, resulting in different resistance levels to insecticides of the same insecticide class, such as pyrethroids (Tong et al. 2013).

Abamectin and pymetrozine are relatively new insecticides with a unique mechanism of action to control aphids. Our results indicate that all *R. padi* field populations had low levels of resistance to abamectin, except for 2 populations that showed moderate resistance to this chemical. Our results also showed that pymetrozine was an effective insecticide for controlling *R. padi*. Although these findings suggest that these insecticides remain effective at managing *R. padi* in the sampled areas, resistance management practices for abamectin should be considered because of the occurrence of low resistance levels found in this study.

We conclude that the sampled *R. padi* field populations varied in their resistance levels to the tested insecticides. As known from previous studies, the emergence of resistance in *R. padi* is subject to many factors, including intensity of insecticide application, the types of insecticides applied, and the genetic backgrounds of the populations (Ahmad et al. 2003; Huang et al. 2004). The aim of chemical control is to keep pest numbers below damaging levels. To maintain our ability to produce wheat efficiently, we must have continued access to effective insecticides. Thus, implementation of resistance-delaying tactics should be considered. Foremost among these tactics are minimizing insecticide use, applying insecticides only when truly needed, and rotation of insecticide classes.

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