Imidacloprid Susceptibility Survey and Selection Risk Assessment in Field Populations of *Nilaparvata lugens* (Homoptera: Delphacidae)

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ABSTRACT Imidacloprid has been used for many years to control planthopper Nilaparvata lugens (Stål) (Homoptera: Delphacidae) in China. To provide resistance assessment for the national insecticide resistance management program, we collected a total of 42 samples of the planthoppers from 27 locations covering eight provinces to monitor their dose responses and susceptibility changes to imidacloprid over an 11-yr period (1996–2006). Results showed that most field populations maintained susceptibility from 1996 to 2003 except for a population from Guilin, Guangxi, in 1997, which showed a low level of resistance to imidacloprid. However, surveys conducted in 2005 indicated that 16 populations from six provinces quickly developed resistance with resistance ratios ranging from 79 to 811. The data collected in 2006 revealed that the resistance levels in 12 populations collected from seven different provinces decreased slightly (RR = 107-316), except the Tongzhou population (Jiangsu Province), which developed 625-fold resistance. Dominant and intensive use of imidacloprid in a wide range of rice, Oryza savita L., growing areas might be a driving force for the resistance development. Migration of the insect also significantly boosted the resistance levels due to extensive and intensive use of imidacloprid in emigrating areas and continuous postmigration sprays of the chemical. In addition, laboratory resistance selection using imidacloprid showed that resistance ratio increased to 14-fold after 27 generations, suggesting that quick resistance development might be associated with more frequent applications of the insecticide in recent years.

KEY WORDS Nilaparvata lugens, imidacloprid, susceptibilities, resistance, risk assessment

The planthopper Nilaparvata lugens (Stål) (Homoptera: Delphacidae) is a serious pest on rice, Oryza sativa L., in Asia. As a monophagous pest, N. lugens limits feeding on *O. sativa* and wild rice species (Pathak and Heinrichs 1982). N. lugens occurs one generation a year in northern rice growing areas and up to 12 generations in southern rice growing areas in China (Ding and Su 2002). Females deposit their eggs in small groups, which may "spread their risks" in space and increase survival (Prestidge 1982). High population density is often seen in fields due to its high mobility and fecundity (Kiritani 1979). In addition to producing typical "hopperburn" feeding damage to the rice, this insect also transmits viral diseases to cause serious stunt in host (Chen et al. 1978, Ling et al. 1978). N. lugens is able to rapidly adapt to resistant plant varieties to produce "biotypes" (Pathak and Heinrichs 1982, Sogawa 1977). Its small body and dimorphic wing types add great flexibility for long distance migration and effective tracking and exploitation of changing hosts (Kisimoto 1965). The migration impels the synchronization between the resource changes and the population up-and-down, and prompts adaptation to the change of environmental conditions, to achieve efficient propagation and multiplication of the population (Denno and Roderick 1990).

Disastrous population outbreaks can easily occur once the conditions become favorable (Heinrichs 1994). Chemical control remains the most effective method for controlling the insect (Endo and Tsurumachi 2001). Resistance development to conventional insecticides, such as organophosphate and carbamate insecticides, was documented in many Asian countries from the 1970s to the late 1990s (Kilin et al. 1981, Hirai 1993). N. lugens is a major migratory insect, and it is able to travel long-distance between southern part and northern China (Cheng et al. 1979). Insecticide resistance levels in destination areas are often seen doubled after the north-bound migration, subjecting to application intensity and resistance nature to a particular insecticide in source locations (Heinrichs 1994).

J. Econ. Entomol. 101(2): 515–522 (2008)

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Fig. 1. Map showing 27 locations in eight major rice growing provinces for collecting the brown planthopper, *N. lugens*, from 1993 to 2006.

Imidacloprid, the first member of the neonicotinyl insecticides, is particularly effective against planthopper Laodelphax striatellus Fallen (Sone et al. 1997). The insecticide was first introduced to China in the early 1990s, and it rapidly became the primary insecticide for controlling N. lugens (Sun et al. 1996). In China, the common formulations of imidacloprid include emulsifiable concentrates (EC) and wettable powders (WP). When being used for seed treatment and foliar application, all formulations are able to consistently maintain a relatively long residual activity against N. lugens. In growing season, three to four sprays of imidacloprid are necessary to bring the population under control. Besides, other insecticides were seldom used due to their relative lower efficacy compared with imidacloprid for controlling N. lugens. The application patterns were almost similar along the migration route. Imidacloprid was also widely used in other Asian countries due to its high efficacy, long residual activity, and environmental compatibility. However, farmers began to switch to other insecticides since 2006 because of decreased efficacy of imidacloprid against N. lugens.

Imidacloprid, like other systemic insecticides, exhibited prolonged residual activity that is likely to generate high selection pressure on the target insect for resistance development (Taylor and Georghiou 1982). Resistance development to imidacloprid was found in field populations of *Leptinotarsa decemlineata* (Say) (Grafius and Bishop 1996), *Myzus persicae* (Sulzer) (Foster et al. 2003), *Bemisia tabaci* (Gennadius) (Cahill et al. 1996), and *Lygus hesperus* (Dennehy and Russell 1996). Moreover, many insects with organophosphate resistance were able to develop cross-resistance to imidacloprid, such as the western flower thrips, *Frankliniella occidentalis* (Pergande), and tobacco budworm, *Heliothis virescens* (F.) (Elzen 1997, Zhao et al. 1995).

Because of the long application history and widespread use of imidacloprid, decreased susceptibility to imidacloprid in *N. lugens* has become a major concern in rice growing regions in China. Besides a highly resistant strain produced from laboratory selection of field-collected *N. lugens* (Liu and Han 2006), very little research has been done for survey and characterization of the imidacloprid resistance in field populations of *N. lugens*. To provide a foundation for areawide resistance management of *N. lugens*, we initiated a study to investigate regional and temporal changes of susceptibilities to imidacloprid in rice production areas. In addition, an imidacloprid-resistant strain was developed in laboratory selection for risk assessment of the resistance development.

Materials and Methods

Insects. The susceptible strain of *N. lugens* was originally collected in 1993 from a rice nursery located at the Plant Protection Station of Jiangpu County (Jiangsu). The insects were reared on hybrid rice (Shanyou 63), and an iso-line was established from a single-pair mating method in the laboratory.

To examine imidacloprid resistance in different rice growing areas, 42 samples in total were collected from 27 locations in eight different provinces or autonomous region (Fig. 1) from 1996 to 2006. Selection of these sites was based on their importance for rice production and their history and intensity of the insecticide applications. Approximately 800 adults, 500~600 nymphs, or sufficient egg masses were collected at each site and transported to the greenhouse on the campus of Nanjing Agricultural University. The insects were reared on insecticide-free hybrid rice (Shanyou 63) before bioassays were performed. The same rice variety at tillering to booting stage was used for maintaining insect colonies and subsequent bioassays. Field-collected insects were mass mated. The third instar nymphs of F_1 progenies were used for bioassays. All treated insects were maintained at a temperature of 27 ± 1°C and a photoperiod of 16:8 (L:D) h.

Nanning Population (NN). A *N. lugens* population of the second generation was collected in 1999 from a rice field in Nanning, Guangxi Autonomous region. This population was reared for ≈ 20 generations and then used for continuous resistance selection with imidacloprid.

Insecticide. Imidacloprid (10% WP) was supplied by Changlong Chemical Industrial Group Co. Ltd., Jiangsu Province, and it was used for testing susceptibility of *N. lugens* during 1996–2003. Imidacloprid (95.3% TC) also was supplied by Changlong Chemical Industrial Group Co. Ltd. Imidacloprid (2.5% EC) was formulated by mixing 2.5% (wt:vol) imidacloprid (TC) with 10% Triton X-100 (wt:vol) and acetone for the laboratory assays in 2005 and 2006.

Bioassay. The rice-stem dipping method (Zhuang et al. 1999) was adopted in this study. The rice was grown in field during the growing season. Otherwise, the rice was planted in pots which were maintained in greenhouse during nongrowing season. Rice stems, including roots, were pulled out and washed thoroughly. The basal 10-cm-long stems were cut and air-dried to remove excess water. Three rice stems were grouped and dipped into appropriate insecticide test solution for 30 s. Three replicates were used per dose and five to six doses, plus a water only control, were used for each chemical. After the rice stems were air-dried for ≈ 1 h, moistened cotton was used to wrap the roots. Treated rice stems were then placed into a 500-ml plastic cup. Twenty third instars were introduced into each plastic cup using a suction device. Each bioassay included five to six concentrations, and three plastic cups were arranged as three repeats for each concentration. The treated insects were maintained at $27 \pm$ 1°C and photoperiod of 16:8 (L:D) h. Mortality was recorded after 4 d. The nymphs were considered dead if they were unable to show movement after gentle prodding with a fine brush.

Selection of Resistance to Imidacloprid. Rice stems were treated with imidacloprid using a dipping method, and they were transferred into a cage (57 by 57 by 57 cm). Approximately 1,000 third instars of *N. lugens* were introduced into the cage and subsequently maintained at $27 \pm 1^{\circ}$ C and with a photoperiod of 16:8 (L:D) h for 4 d. Survivors were transferred to another cage containing fresh rice seedlings. Pretrials were conducted to obtain an optimal mortality level for resistance selection. The mortality was controlled to range between 40 and 70% to ensure sufficient survivors to develop and reproduce enough progeny for the insecticide selection of subsequent generations (The treatment concentration was approximately the same LC_{50} value for each generation). The insect received imidacloprid treatment every generation for a total of 27 generations.

To assess risk of imidacloprid selection on resistance development, realized heritability (h^2) was estimated by using a method described by Tabashnik (1992) as $h^2 = R/S$, where R is the response to selection and S is the selection differential. Response to selection (R) was estimated as $R = [\log (\text{final LC}_{50}) - \log (\text{initial})]$ LC_{50}]/n (where the final LC_{50} is the LC_{50} of offspring after n generations of selection, the initial LC₅₀ is the LC₅₀ of the parental generation before the selections start, n is the number of generations selected. The selection differential (S) was estimated as $S = i \times i$ σ_{p} , where *i* is the intensity of selection and σ_{p} is the phenotypic standard deviation). Intensity of selection (i) was estimated as i = 1.583 - 0.0193336p + $0.0000428p^2 + 3.65194/p$, where p is the average percentage of surviving rate (Tabashnik and McGaughey 1994). The phenotypic standard deviation (σ_p) was estimated as $\sigma_p = [1/2(\text{initial slope} + \text{final slope})]^{-1}$, where initial slope is the slope of the probit regression lines from the parental generation before selection and final slope is the slope of the probit regression lines from offspring after n generation selection.

Statistical Analysis. Mortality was corrected using Abbott's formula (Abbott 1925) for each probit analysis. The POLO program was used for probit analysis of dose–response data (LeOra Software 1997), unless otherwise stated. The resistance ratio (RR) was calculated by dividing the LC_{50} of a field population by the LC_{50} of the susceptible strain. Resistance levels were classified based on the standard described by Shen and Wu (1995): susceptible, <3-fold; minor resistance, \approx 3–5-fold; low resistance, \approx 5–10-fold; medium resistance, \approx 10–40-fold; high resistance level, \approx 40–160-fold; and extremely high resistance level, >160-fold.

Results

Toxicity of Two Imidacloprid Formulations against the Susceptible Strain of *N. lugens*. The LC₅₀ values of imidacloprid WP (10%) and imidacloprid EC (2.5%) in the susceptible strain were 0.09 (0.08–0.11) and 0.08 (0.05–0.11) mg (AI)/liter, respectively (Tables 1 and 2). The overlapped 95% CL of the LC₅₀ values indicated that two different imidacloprid formulations had the same toxicity against the susceptible strain of *N. lugens*.

Dose Response and Resistance Level in Field Populations. We collected 42 field samples in total from eight provinces (autonomous region) from 1996 to 2006 to examine their susceptibility to imidacloprid. The results showed that most of the *N. lugens* populations from Jiangsu Province (Nanjing, Yizheng, and Nantong), Anhui Province (Anqing), and Guangxi Autonomous region (Nanning and Guilin) remained susceptible (RR < 5-fold) to imidacloprid from 1996

	Test pop		Dose response					
Yr	Province	Location	Slope	$\begin{array}{c} {\rm LC}_{50} \ ({\rm mg} \ [{\rm AI}]/{\rm liter}) \\ (95\% \ {\rm CL}) \end{array}$	RR ^a			
	Suscepti	ole strain	2.0792	0.09 (0.08-0.11)	1.0			
1996	Anhui	Anging	3.5949	0.33 (0.29-0.36)	3.7			
	Guangxi	Guilin	2.3891	0.37 (0.32-0.43)	4.1			
	Guangxi	Nanning	2.8584	0.40 (0.35-0.44)	4.4			
	Jiangsu	Nanjing	2.3661	0.37 (0.32-0.42)	4.1			
1997	Anhui	Anging	2.6178	0.33 (0.28-0.37)	3.7			
	Guangxi	Guilin	2.6217	0.57(0.51-0.64)	6.3			
	Guangxi	Nanning	3.0210	0.42(0.39-0.44)	4.7			
	Jiangsu	Nanjing	3.2496	0.36 (0.32-0.40)	4.0			
1998	Guangxi	Nanning	3.3481	0.21 (0.01-0.22)	2.3			
	Jiangsu	Yizheng	4.1345	0.22 (0.19-0.24)	2.4			
1999	Guangxi	Nanning	3.3536	0.12 (0.10-0.14)	1.3			
	Jiangsu	Nantong	4.5779	0.11 (0.09-0.13)	1.2			
2002	Guangxi	Nanning	1.9867	0.08 (0.07-0.10)	0.9			
2003	Jiangsu	Nanjing	2.2620	0.29 (0.25–0.35)	3.2			

Table 1. Dose response and resistance ratio to imidacloprid (WP) in field populations of N. lugens collected from 1996 to 2003

^{*a*} Resistance ratio (RR) = LC_{50} of field pop/ LC_{50} of the susceptible colony (generation 30).

to 2003 except for the Guilin population with minor resistance in 1997 (RR = 6.3-fold; Table 1). However, high to extremely high resistance levels to imidacloprid (RR = 79.1-551.8-fold) were found in August 2005 (Table 2) in the three populations from Guilin, Nanning, and Nanjing, where the insects were repeatedly assayed from 1996 to 2003, and their resistance ratios were no greater than 6.3-fold. The other 13 populations from Jiangsu, Zhejiang, Anhui, and Jiangxi provinces also developed extremely high resistance to the chemical (RR = 206.5-811) during September and October 2005.

Further resistance surveys between June and August 2006 (Table 3) indicated that five field populations of *N. lugens* from Nanning (Guangxi), Shanggao (Jiangxi), Nanjing (Jiangsu), Xiaogan (Hubei), and Jinhua (Zhejiang) developed high to extremely high resistance levels to imidacloprid (RR = 107.1-232.3fold), which did not increase substantially but maintained median levels similar to those during the same period of 2005. In general, the other seven field populations from four different provinces developed or maintained extremely high resistance levels from September to October in 2006, especially the Tongzhou population (Jiangsu Province), which developed the highest resistance level (625.1-fold) to the chemical. The surveys conducted in 2006 showed that the resistance ratio of the Nanning population relatively declined from 206.5-fold in 2005–107.1-fold, and resistance level of the Nanjing population also decreased from 551.8-fold to 162.3-fold.

Resistance Selection. In the continuous resistance selection tests, imidacloprid was used to treat every generation of Nanning population for 27 generations. The results indicated that the resistance level to imidacloprid increased substantially from 1.6-fold to 13.9-fold (Fig. 2). In the first three generations, selection with imidacloprid did not reveal a distinct increase in resistance ratios, which only fluctuated between 1.6–1.7-fold. From generation four to generation 6, the

Table 2. Dose response and resistance ratio to imidacloprid (EC) in field populations of N. lugens collected in 2005

	Test pop		Dose response							
Мо	Province	Location	n	Slope (SE)	$\begin{array}{c} {\rm LC}_{50} \ ({\rm mg} \ [{\rm AI}]/{\rm liter}) \\ (95\% \ {\rm CL}) \end{array}$	χ^2 (df)	RR^{a}			
	Susceptible strain		420	1.52 (0.17)	0.08 (0.05-0.10)	7.1 (4)	1.0			
Aug.	Guangxi	Guilin	420	1.76 (0.21)	6.33 (4.88-7.90)	0.6 (3)	79.1			
8.	Guangxi	Nanning	420	1.26 (0.16)	16.52 (12.39-22.03)	1.7(4)	206.5			
	Hunan	Changde	420	1.32 (0.15)	16.03 (8.69-29.52)	8.5 (4)	200.4			
	Jiangsu	Nanjing	420	1.47(0.21)	44.14 (31.68-59.18)	0.6(3)	551.8			
Sept.	Anhui	Caohu	420	1.76 (0.21)	22.16 (17.40-27.95)	5.3 (4)	277			
-	Jiangsu	Gaochun	420	1.64(0.18)	26.37 (17.10-38.46)	5.7(4)	329.6			
	Jiangsu	Suzhou	420	2.36(0.30)	63.97 (49.82-78.29)	4.0 (3)	799.6			
	Jiangsu	Wuxi	420	1.60(0.19)	32.69 (24.85-42.05)	2.8(4)	408.6			
	Zhejiang	Haiyan	420	1.23(0.15)	46.67 (35.85-63.45)	1.2(4)	583.4			
	Zhejiang	Jiaxing	420	1.40(0.16)	29.03 (21.89-37.91)	1.6(4)	362.9			
	Zhejiang	Shaoxing	420	1.48(0.17)	27.38 (17.85-40.43)	4.8 (4)	342.3			
	Zhejiang	Tongxiang	420	1.69(0.24)	64.88 (49.24-85.78)	1.1(4)	811			
	Zhejiang	Yuyao	420	1.34(0.18)	45.35 (32.87-62.35)	0.8(4)	566.9			
Oct.	Anhui	Hexian	420	1.30 (0.20)	50.01 (34.86-70.73)	2.5 (4)	625.1			
	Jiangxi	Nanchang	420	2.53 (0.35)	36.72 (28.41-44.73)	2.6 (4)	459			
	Jiangxi	Xinjian	420	1.44 (0.21)	60.02 (43.72-82.96)	1.7 (4)	750.3			

^{*a*} Resistance ratio (RR) = LC_{50} of field popultion/LC50 of the susceptible colony (generation 121).

Table 3. Dose response and resistance ratio to imidacloprid (EC) in field populations of N. lugens collected in 2006

	Test	Pop		Dose response						
Month	Province	ovince Location		Slope (SE)	$\begin{array}{c} {\rm LC}_{50} \ ({\rm mg} \ [{\rm AI}]/{\rm liter}) \\ (95\% \ {\rm CL}) \end{array}$	χ^2 (df)	RR^{a}			
	Susceptible strain		420	1.52 (0.17)	0.08 (0.05-0.10)	7.1 (4)	1.0			
June	Guangxi	Nanning	420	1.68 (0.17)	8.57 (6.62-10.63)	1.5(4)	107.1			
July	Jiangxi	Shanggao	420	2.22 (0.24)	14.31 (11.55–17.19)	1.2 (3)	178.9			
Aug.	Hubei	Xiaogan	420	1.81 (0.18)	18.58 (14.63-22.81)	2.2(4)	232.3			
0	Jiangsu	Nanjing	420	1.49 (0.20)	12.98 (0.31-32.12)	12.3 (3)	162.3			
	Zhejiang	Jinhua	420	1.61(0.17)	14.05 (10.33-17.89)	2.0(4)	175.6			
Sept.	Anhui	Hexian	420	1.52 (0.16)	19.77 (15.30-24.85)	4.0 (4)	247.1			
	Anhui	Ningguo	420	1.93 (0.19)	24.59 (20.18-29.81)	1.3 (4)	307.4			
	Anhui	Qianshan	420	2.88(0.34)	15.01 (12.87-17.73)	5.6(2)	187.6			
	Fujian	Fuging	420	2.05 (0.20)	19.79 (16.11-23.81)	3.6(4)	247.4			
	Jiangsu	Tongzhou	420	2.42 (0.27)	50.01 (42.09-61.41)	2.4(3)	625.1			
	Zhejiang	Haiyan	420	1.64 (0.17)	25.21 (20.30-31.19)	8.6 (4)	315.1			
Oct.	Jiangsu	Dongtai	420	2.08 (0.20)	25.35 (21.01-30.47)	5.2(4)	316.9			

^{*a*} Resistance ratio (RR) = LC_{50} of field population/ LC_{50} of the susceptible colony (generation 121).

resistance ratios quickly increased to 7.6-fold. After that, the resistance ratios showed a decline trend until generation 12 with the resistance ratio of 3.0-fold. But the resistance ratio of generation 13 quickly reached 7.8-fold, after which the resistance continued to increase at a relatively slow speed until generation 22 with the resistance ratio of 9.0-fold. When the experiment ended at generation 27, the resistance increased to 13.9-fold, the highest resistance ratio obtained in the test generations. The average mortality and the estimate of realized heritability (h^2) during the process of resistance selection were 63.57 ± 11.32% and 0.0825, respectively (Table 4). It was likely that the resistance level of *N. lugens* to imidacloprid would increase further if the selection had continued.

Discussion

Imidacloprid Use History and Resistance Development. Resistance development to imidacloprid in *N. lugens* may be closely associated with the application history of the chemical. Since the first introduction in the early 1990s, imidacloprid has been an important chemical for controlling *N. lugens* in China (Qiu et al. 1997, Liu et al. 2003). The increase of imidacloprid applications was a direct consequence of the decrease in buprofezin applications because buprofezin had little effect on adult and egg stages, though it had relatively high efficacy against the nymphs of *N. lugens* (Asai et al. 1983).

Our resistance monitoring data indicated that *N. lugens* had maintained susceptibility to imidacloprid for >10 yr since the first introduction of the chemical. Before 2003, no distinct resistance was detected. Populations collected from southern and southeast rice growing areas generally remained susceptible from 1996 to 2003. This phenomenon indicated that initial imidacloprid-resistant genes might exist in the populations at a very low level. Continuous and dominant use of imidacloprid for up to 13 yr has allowed the resistant gene frequency to build up to a critical turning point. Within only a 2-yr period, the Nanning population showed a sharp increase in resistance by

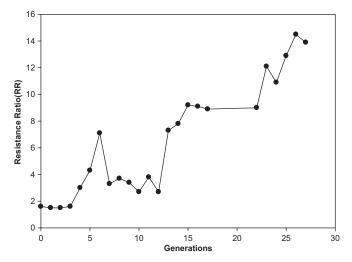


Fig. 2. Assessment of resistance development speed to imidacloprid in *N. lugens* which received insecticide treatment every generation for a total of 27 generations.

Table 4. Estimation of realized heritability (h^2) of resistance to imidacloprid in N. lugens from laboratory-selection experiment

Рор	No. of generations selected	Estimate of mean response per generation			Est	Estimate of mean selective differential per generation					
(selection scheme)		Initial LC ₅₀ (log)	Final LC ₅₀ (log)	R	p	i	Initial slope	Final slope	σ_p	S	h^2
Nanning	27	-0.8359	0.0964	0.0345	36.4	1.0363	1.8218	3.1307	0.4038	0.4185	0.0825

>200-fold. In many other areas, especially in southeast rice growing areas, *N. lugens* developed >800-fold resistance to imidacloprid. Resistance development to certain insecticides due to their long application history also can be seen in many other insects (Denholm et al. 1998, Mohan and Gujar 2003, Sayyed et al. 2005, Zhao et al. 2006, Yu and McCord 2007).

Resistance Development Due to Extensive and Intensive Use of Imidacloprid. One important factor for imidacloprid resistance development in N. lugens is due to its wide spread adoption in almost all rice growing areas in China, which applied heavy selection pressure on the target insects and accelerated resistance development in N. lugens. Since the early 1990s, imidacloprid has been used for control of N. lugens because of its systemic nature and high efficacy against sucking insects (Liu et al. 2003). Continuous use of imidacloprid as a dominant insecticide for planthopper control resulted in a gradual decrease of its efficacy against target insects (efficacy drop from 95 to 60% control in the Yangtze River Delta areas such as Jiangsu and Zhejiang provinces and other areas; data not shown). To maintain effective control, the application dose was increased from 15 g (AI)/ha to ≈ 60 -120 g (AI)/ha. In many rice growing areas, farmers sprayed every generation of N. lugens to prevent its outbreaks. Intensive applications of imidacloprid applied heavy selection pressure on N. lugens and resulted in a dramatic increase of resistance to the chemical. Similar situations also have yielded an increase in resistance in many other insects, such as imidacloprid resistance development in L. decemlineata and B. tabaci, triazophos resistance in Chilo suppressalis (Walker), and pyrethroids resistance in Helicoverpa armigera (Hübner) (Shen and Wu 1995, Elbert and Nauen 2000, Qu et al. 2003, Cao et al. 2004, David et al. 2006).

Therefore, it is likely that the long history of intensive use of imidacloprid is the major factor for facilitating high resistance development in many populations of N. lugens. This hypothesis was proven to be true from our laboratory studies. By measuring realized heritability (h^2) , we demonstrated that N. lugens could develop a certain level of resistance to imidacloprid when the target insect received constant treatment in the laboratory. We also observed that the resistance developed was relatively fast if the target insects were treated more frequently (every generation), otherwise, the speed of resistance development was relatively slow when the insects were treated less frequently (data not shown). In spite of this, resistance development in laboratory selection was relatively slower (14-fold after 27 generations) than that in field populations (up to 207-fold for Nanning 2005 population). Faster resistance development in field populations might be associated with migration which supplied with highly resistant insects carrying diverse resistance gene resources. In addition, relatively longer residual activity and frequent sprays also placed the population under constant selection pressure, and subsequently prompted rapid resistance development in the field populations.

The susceptibility survey in 2006 provided evidence of selection impact on resistance change. In comparison with the data of 2005, the resistance ratios in all seven populations dropped slightly in 2006. This phenomenon might be attributed to the temporal suspension of imidacloprid for planthopper control. As a consequence of decreased efficacy due to resistance development to imidacloprid, farmers switched to other insecticides to control the planthopper. Therefore, selection pressure for imidacloprid resistance was temporarily reduced, and the resistance levels to imidacloprid in many N. lugens populations decreased accordingly, as detected in our 2006 survey. Based on these findings, rotating imidacloprid with buprofezin and other insecticides would be the most important practice to slow down resistance development to imidacloprid in N. lugens.

Influence of Migration on Resistance. N. lugens is a migratory insect and resistance development to insecticides was expected to be slow because resistance might be diluted in the process of migration (Zhuang et al. 2004). However, unlike other insecticides, imidacloprid was used to control N. lugens not only in the emigrating region but also in the immigrating region (Liu et al. 2003). In addition, farmers applied imidacloprid to every generation of N. lugens to suppress its damaging population (Liu and Han 2006). The intensive use of imidacloprid also was observed in other southeastern Asian countries, especially Vietnam and Thailand, where the insects continue feeding in winter and become the major source for north-bound migration to China in the following year (Liu et al. 2004, Zhuang et al. 2004). Subsequently, the migration of N. lugens no longer postpones resistance development substantially.

Due to the migrating nature of *N. lugens*, it is possible that a barrier of resistance gene flow does not exist among large-scale geography areas, i.e., distinct geographic genotypes are less likely present (Yao et al. 2002). Once the insect evolved resistance to imidacloprid, the resistance would quickly spread across whole rice production areas. In destination areas, insecticide resistance level is often seen doubled after migration (Heinrichs 1994). Because of the widespread use of imidacloprid in southern China and other southeastern Asian countries, northern migration allowed those highly resistant insects to relocate in northern rice growing areas, where the insects received further treatments of imidacloprid. Therefore, resistance levels to imidacloprid increased dramatically in the destination areas (Cheng and Zhu 2006). Besides this, imidacloprid resistance in *N. lugens* kept increasing as the season progressed after the immigrants settled down in a local area because of frequent use of the chemical and constant selection pressure on target pest. Because of the dynamic nature of the resistance, management of resistance must rely on not only local but also areawide and even international collaborations (Gao et al. 2006).

Considerations for Imidacloprid Resistance Management. Based on large area surveys of imidacloprid resistance in 28 field populations of N. lugens (2005-2006) and laboratory risk assessment, intensive use of the chemical in almost all rice areas for >10 yr might be the reason for the high level of resistance and potential cause for serious population outbreaks. To delay or slow down resistance development, relaxing selection pressure might be the most important strategy for management of imidacloprid resistance in N. lugens. This can be realized through alternation and rotation of insecticides with different modes of action, such as buprofezin, chlorpyrifos, isoprocarb, fipronil, and dichlorvos. Selection of insecticides for rotation must be preferably given to those without cross-resistance (Alyokhin et al. 2007). Therefore, it is very urgent to determine potential cross-resistance to other neonicotinoids in imidacloprid-resistant population of *N. lugens* (Liu et al. 2003). In high resistance risk areas, temporary suspension of imidacloprid use may be a practical way to slow down resistance development (Cheng and Zhu 2006). In addition, a resistance management program must be formulated to include a scheme of insecticide alternation and rotation and must be implemented not only in local areas but also at a nationwide level. An international collaboration and coordination with emigrating source countries is essential for a successful management program (Gao et al. 2006).

Acknowledgments

We thank Prof. Xianian Cheng (Nanjing Agricultural University), Jian Chen and Sandy West (USDA-ARS, Stoneville, MS), and Fangneng Huang (Louisiana State University, Baton Rouge, LA) for critical review of the early version of the manuscript. The study was partially supported by the research project of the Ministry of Agriculture in China for Replacing High Toxicity Pesticides.

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Received 18 August 2007; accepted 7 January 2008.