

# Genealogical Analysis of Resistance to the Whitebacked Planthopper, *Sogatella furcifera* in Chinese Japonica Rice Chunjiang 06

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## 中国粳稻春江 06 抗白背飞虱的系谱分析

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**摘要:**对中国粳稻春江 06 的抗白背飞虱特性进行系谱分析表明, 春江 06 对白背飞虱的拒取食和杀卵抗性均来自于秀水 620。秀水 620 的亲本中, 只有秀水 04 具有较强的拒取食抗性, 但没有杀卵抗性。在祥湖 24 中检测到明显的杀卵反应。亲本秀水 04、单 209 和辐农 709 具有拒取食抗性, 但测 21 没有。单 209 和辐农 709 的共同亲本农虎 6 号也具有拒取食抗性。然而, 农虎 6 号、农垦 58(日本粳稻)和老虎稻(中国粳稻地方品种)不具有拒取食抗性, 在田间表现出感虫性。农虎 6 号、单 209、辐农 709 和秀水 04 表现出稳定的田间抗性。春江 06 育种中的两个籼稻品种 IR26 和 IR28 高感白背飞虱, 既无拒取食抗性, 也无杀卵作用。

**关键词:** 白背飞虱; 品种抗性; 粳稻; 系谱分析; 拒取食抗性; 杀卵抗性

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**Abstract:** Genealogical analysis was conducted to study the resistance to the whitebacked planthopper (WBPH), *Sogatella furcifera*, in a Chinese japonica rice variety Chunjiang 06 (CJ-06). Results indicated that both the sucking inhibitory- and ovicidal resistance to WBPH were inherited from Xiushui 620 (XS-620). Among the parental lines of XS-620, only Xiushui 04 (XS-04) had a strong sucking inhibitory resistance, but no ovicidal resistance. Significant ovicidal response was detected in Xianghu 24. Of the 3 parental lines of XS-04, Dan 209 (D-209) and Funong 709 (FN-709) had sucking inhibitory resistance, whereas Ce 21(C-21) did not. Nonghu 6 (NH-6), a common parent of D-209 and FN-709, also inhibited sucking. However, the parents of NH-6, Nongken 58 (Japanese japonica) and Laohudao (LHD, Chinese japonica landrace), had no sucking inhibitory resistance, and were susceptible to WBPH under the field conditions. NH-6, D-209, FN-709 and XS-04 expressed definite resistance to WBPH in the fields. Two indica varieties IR26 and IR28, which were used to breed CJ-06, were highly susceptible to WBPH, and had neither sucking inhibitory- nor ovicidal resistance.

**Key words:** whitebacked planthopper; varietal resistance; japonica rice; genealogical analysis; sucking inhibition; ovicidal reaction

## 1 Introduction

A Chinese japonica rice variety Chunjiang 06 (CJ-06) was found to be resistant to the whitebacked planthopper (WBPH), *Sogatella furcifera*. The WBPH resistance in CJ-06 is mediated by two independent antibiotic mechanisms<sup>[8]</sup>. One is an ovicidal resistance, by which WBPH eggs suffer high mortality in the watery lesions induced at oviposition sites<sup>[6,7,9]</sup>. The other is sucking inhibitory resistance, which strongly restricts planthopper feeding and colonization on CJ-06. The dual mechanisms of varietal resistance provide a stable and durable field resistance to WBPH in CJ-06.

The ovicidal resistance to WBPH is a unique self-defense response of japonica rice against the planthopper infestations,

while the sucking inhibitory resistance has been known to be as a common basic mechanism for varietal resistance to the planthoppers in indica rice<sup>[2-4]</sup>. The present experiments were conducted to identify the genetic donors of compound WBPH resistance traits in CJ-06.

## 2 Materials and Methods

### 2.1 Rice varieties used

Totally 16 varieties involved in the pedigree of CJ-06

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**Table 1 . Rice varieties used in the present experiment and their abbreviations used in the text, figures and tables.**

Rice variety	Abbreviation	Type of rice
Aijing 23	AJ-23	indica/japonica
Belila	BLL	Italian japonica
Ce 21	C-21	japonica
C81-40	C81-40	japonica
Chunjiang 06	CJ-06	japonica
CP	CP	japonica
Dan 209	D-209	japonica
Funong 709	FN-709	japonica
Hulei	HL	japonica
IR26	IR26	indica
IR28	IR28	indica
Jinlei 440	JL-440	japonica
Jinyin 154	JY-154	Japanese japonica
Laohudao	LHD	japonica
Lingfeng	LF	Japanese japonica
Nonghu 6	NH-6	japonica
Nongken 57	NK-57	Japanese japonica
Nongken 58	NK-58	Japanese japonica
Xiushui 02	XS-02	japonica
Xiushui 04	XS-04	japonica
Xiushui 620	XS-620	japonica

were selected to evaluate WBPH resistance. Most of them were supplied by Jiaying Academy of Agricultural Sciences, while the others were obtained from China National Rice Research Institute (CNRRRI) and International Rice Research Institute (IRRI). The rice varieties used in the present experiments were listed in Table 1.

## 2.2 Evaluation of WBPH resistance

All the varieties were individually planted in disposable plastic cups (7 cm in diameter, 9 cm in height) until early tillering stage under open conditions. Gravid females were individually confined onto the upper portion of leaf sheaths with parafilm sachets (2 cm×2 cm), and allowed to suck and lay eggs for a day at room temperature (26–30°C).

Sucking inhibitory resistance was evaluated quantitatively by weighing honeydew excreted in the parafilm sachets. Ovicidal resistance was assessed by calculating egg mortality. This was done by counting the number of live and dead eggs at 5–6 days after oviposition by dissecting the leaf sheath tissues at oviposition sites. The eggs with reddish eye-spots were categorized as developing live eggs, and white opaque eggs as dead ones.

## 2.3 Evaluation of field performance of WBPH resistance

Ten selected CJ-06 family varieties were separately planted in a paddy plot (35 m×20 m). One-month seedlings were individually transplanted at 25 cm×20 cm intervals on June 21, 2002. The rice plants were grown under the standardized practices without using pesticides. Densities of female adults of WBPH were counted every week by selecting 100 hills for the first four weeks and 50 hills after five weeks.

Density of the first- and second-generation nymphs was estimated by a tapping method. Nymphs on each hill were tapped down in a 40 cm×28 cm rectangular shallow tray placed at the base of hill, and the number of nymphs that fell into the tray was quickly counted. Ten hills were randomly selected for each variety. Intensity of necrotic symptoms due to ovicidal response in rice plants was also recorded twice on July 17 and August 3 by assigning scores of 0 to 3.

## 3 Results

### 3.1 WBPH resistance in intermediate parents

CJ-06 and its 8 intermediate parental varieties including two indica varieties were subjected to identification of intermediate donors of WBPH resistance traits (Fig. 1). Both the sucking inhibitory- and ovicidal resistance of CJ-06 were inherited from Xiushui 620 (XS-620) (Fig. 2). Another immediate parental line C81-40 showed definite ovicidal response, but was lacking in sucking inhibitory resistance. XS-620 was bred by crossings of the 4 intermediate parental lines, namely Xiushui 04 (XS-04), Xiushui 02 (XS-02), Xianghu 24 (XH-24) and CP. CP is no longer conserved. Among the three remaining parental lines, only XS-04 showed a strong sucking inhibitory resistance, causing daily honeydew excretion to be suppressed to below 5 mg/(female·day) (Fig. 2). However, XS-04 did not have ovicidal resistance. Significant ovicidal response was detected in XH-24. WBPH excreted 9.4 mg/(female·day) honeydew, and the egg mortality was 38% on an average in XS-02, indicating no significant sucking inhibition and ovicidal response.

Two indica varieties IR26 and IR28, which were respectively used to breed XS-04 and CP, were highly susceptible to WBPH, and had neither sucking inhibitory- nor ovicidal mechanisms of resistance (Fig. 2). An indica/japonica hybrid variety Aijing 23 (AJ-23), which was an intermediate parent of XS-02, suppressed WBPH sucking to some extent [6.7 mg/(female·day) on an average], and caused only a weak ovicidal response.

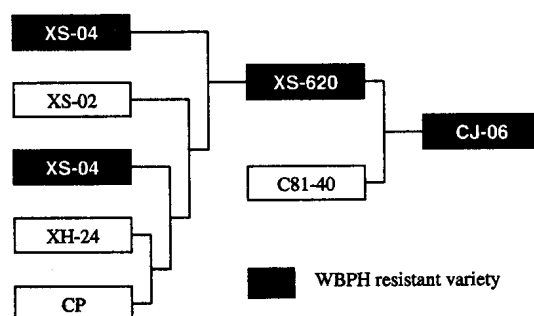


Fig. 1. Intermediate parental varieties of Chunjiang 06(CJ-06).

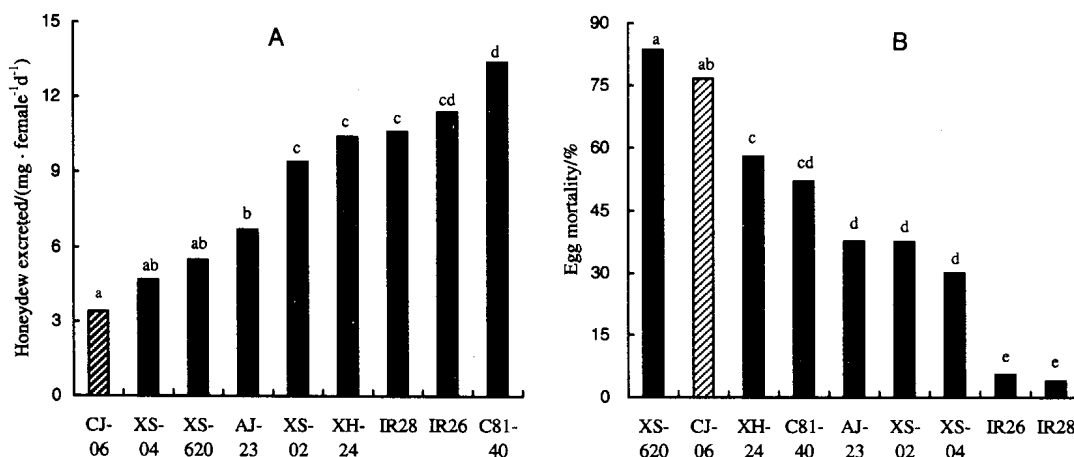


Fig. 2. Honeydew excretion by WBPH females (A) and WBPH egg mortality (B) in intermediate parental varieties of CJ-06 and some related varieties.

Bars marked with the same letters are not significantly different ( $P < 0.05$ , LSD).

### 3.2 Sucking inhibitory resistance in XS-04 family varieties

WBPH resistance in seven varieties involved in the pedigree of XS-04 was tested (Fig. 3). Among the 3 parents of XS-04, Dan 209 (D-209) and Funong 709 (FN-709) inhibited WBPH sucking, while Ce 21 (C-21) did not (Fig. 4). Nonghu 6 (NH-6), which is a common parent of D-209 and FN-709 was also sucking inhibitory. However, sucking inhibitory resistance in the parents of NH-6, Nongken 58 (NK-58, Japanese japonica) and Laohudao (LHD, Chinese japonica landrace), is not clear, although honeydew excretion on LHD was somewhat suppressed.

### 3.3 Field performance of WBPH resistance in selected CJ-06 related varieties

There was a big immigration surge of WBPH on June 19 – 23. Immigrants invaded the experimental plots at a density of 0.4 – 2.2 females/hill. Significantly more gravid immigrant females (1.1 – 1.5 females/hill) inhabited in the plots of C-21, LHD, and XH-24, while their densities on D-209, FN-709, XS-04 and NH-6 were 0.1 – 0.2 females/hill on July 10, when the first generation nymphs started hatching. In this period, conspicuous ovicidal symptoms caused by oviposition by the immigrant females appeared in NK-58, C-21 and XH-24 (Fig. 5). CJ-06 and BLL also exhibited ovicidal symptoms, but to a lesser extent. In mid July, density of the nymphs increased to 25 nymphs/hill in LHD, and about 10 nymphs/hill in BLL and NK-58 (Table 2). In the other varieties, nymphal density did not increase beyond 5 nymphs/hill. In particular, it was suppressed below 1 nymph/hill in FN-709 and NH-6.

The first-generation nymphs produced brachypterous females in late July, which stayed in the original habitat and produced second-generation nymphs. Density of brachypterous females was highest in LHD, 6 females/hill on an average. The density was 1.4 – 2.2 females/hill in C-21, BLL

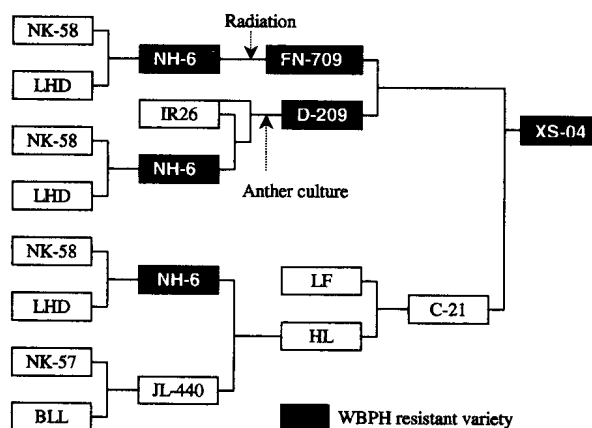


Fig. 3. Genealogy of sucking inhibitory resistant variety XS-04.

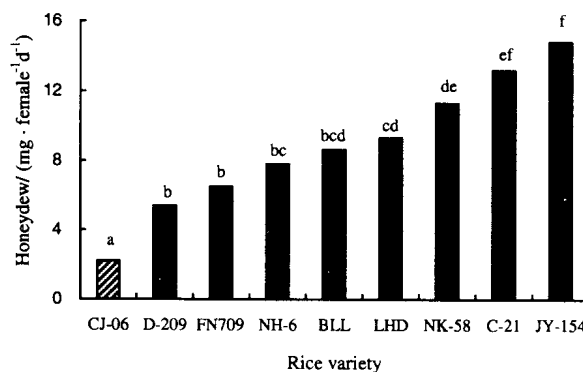


Fig. 4. Honeydew excretion by WBPH on several varieties in the pedigree of CJ-06.

Bars marked with the same letters are not significantly different ( $P < 0.05$ , LSD).

and NK-58. In the other varieties, the density was kept below 0.5 female/hill. The most conspicuous ovicidal symptoms appeared in XH-24, although female density was the lowest on XH-24 (Fig. 6). The symptoms in C-21 and NK-58 were also clear. At the end of July, the second-generation nymphs started to emerge, and grew to the maximum stage in mid August. The nymphal density in LHD was extraordinarily high, loading about 500 nymphs/hill (Table 2). The LHD plants were seriously stunted, and partially destroyed by heavy infestation of nymphs. Nymphal densities in BLL, NK-58 and C-21 were around 50 nymphs/hill. Nymphal populations in the other varieties were very scanty. The second-generation nymphs emerged to macropterous adults, which emigrated from the experimental fields in late August.

## 4 Discussion

### 4.1 Origins of ovicidal resistance

As we have demonstrated, ovicidal resistance is a characteristic defense mechanism to WBPH infestations in japonica rice<sup>[6,7,9]</sup>. The japonica varieties involved in the pedigree of CJ-06 retained rather commonly the ovicidal resistance. NK-58, Japanese japonica, is apparently one of the original donors of the trait. The genetic expression of ovicidal response became inconspicuous in the progenies (NH-6, D-209, FN-709 and XS-04) of NK-58 and LHD. However, the potential ovicidal traits were inherited from XH-24 to XS-620, and CJ-06. C81-40 was also an independent donor of the ovicidal resistance to CJ-06.

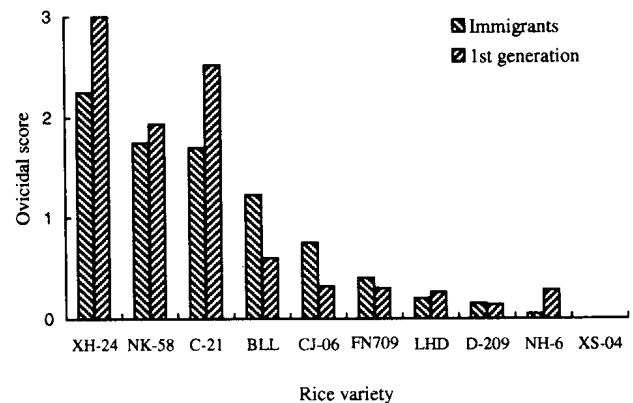
### 4.2 Origin of sucking inhibitory resistance

Sucking inhibitory resistance in CJ-06 was derived restrictedly from XS-04 through XS-620. The sucking inhibitory resistance of XS-04 could be retraced to NH-6 via FN-709 or C-209. However, NK-58 and LHD, the parents of

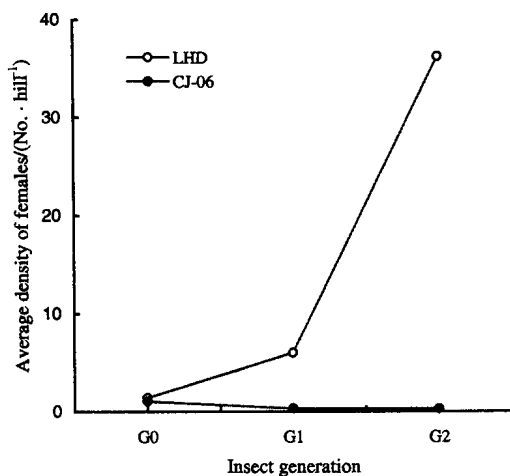
**Table 2. Relative nymphal densities of WBPH in rice varieties related to CJ-06.**

Rice variety	Relative nymphal density <sup>1)</sup> (no. per hill)	
	1st generation	2nd generation
LHD	24.5 a	486.3 a
BLL	10.9 ab	66.4 ab
NK-58	8.1 abc	54.7 ab
XS-04	4.1 bed	5.6 bed
C-21	3.0 bed	32.3 abc
CJ-06	2.3 bed	2.0 cd
XH-24	2.3 cd	5.8 bed
D-209	1.9 d	0.9 d
NH-6	0.6 d	0.6 d
FN-709	0.2 d	0.1 d

<sup>1)</sup>In a column, means followed by the same lowercase letters are not significantly different by Kruskal-Wallis test,  $P = 0.05$ .



**Fig. 5. Intensity of ovicidal symptoms in rice varieties related to CJ-06 infested with immigrant and the 1st generation females of WBPH in the fields.**

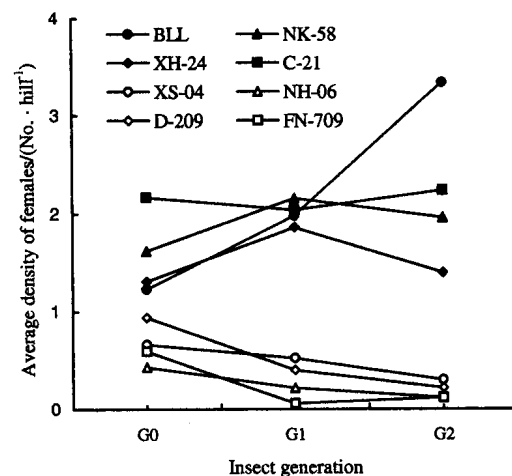


**Fig. 6. Population trends of WBPH in rice varieties related to CJ-06.**

G0(Immigrants); Average density of macropterous females in 100 hills on July 3;

G1(1st generation); Average density of brachypterous females in 50 hills in July 24–31;

G2(2nd generation); Average density of macropterous females in 50 hills in August 14–21.



NH-6, had no sucking inhibitory resistance. Therefore, its origin remains obscure. It must be confirmed whether or not progeny with the sucking inhibitory trait could be bred from the parents. NH-6 is one of the commonly adopted intermediate parents in the pedigree of CJ-06. However, its sucking inhibitory resistance is not necessarily inherited to the descendants. C-21 and XH-24 are the descendants of NH-6, but do not express sucking inhibitory resistance. Two indica IR varieties, IR26 and IR28, have been used to introduce *Bph 1* gene for the brown planthopper resistance<sup>[1,11]</sup>. However, these IR varieties were found to be highly susceptible to WBPH as has been evaluated at IRRI<sup>[10]</sup>, having neither ovicidal- nor sucking inhibitory resistance. AJ-23 is derived from indica/japonica hybridization between dwarf indica Aizizhan and japonica landrace Laolaiqing. AJ-23 did not show a definite resistance to WBPH. Thus, no indica varieties were the donors of sucking inhibitory resistance to WBPH in CJ-06.

#### 4.3 Field evaluation of WBPH resistance in intermediate parents of CJ-06

Sucking inhibitory- and/or ovicidal resistance in CJ-06 and its family varieties were evaluated by exposing them to the natural infestations of WBPH under the field conditions. WBPH could not establish populations on CJ-06 that had the dual resistance traits. On the contrary, WBPH reproduced exponentially for two consecutive generations, and caused serious damages on LHD that has no resistance traits (Fig. 6-A). The rest of the varieties were divided into two groups depending upon the resistance trait that they had (Fig. 6-B). NH-06, D-209, FN-709 and XS-04, in which WBPH populations were not developed because of non-preference response of immigrant adults, have the sucking inhibitory resistance, but not significant ovicidal resistance. Adult density declined as generations progressed on those varieties. The population trend on these varieties was basically the same as that on CJ-06.

On the other hand, NK-58, C-21, XH-24 and BLL have ovicidal activity but no sucking inhibitory resistance. Therefore, laying eggs by WBPHs was not inhibited on these varieties. As the results, conspicuous necrotic ovicidal symptoms appeared on their leaf-sheaths, when the immigrants invaded and the first-generation adults emerged. Vegetative growth of these varieties was slightly deterred by intensive ovicidal response. Instead, population upsurge of WBPH nymphs and consequent destructive damage to the host plants was effectively avoided by high egg mortality. However, these ovicidal varieties allowed survival and development of a small number of nymphs that escaped the ovicidal response.

The field evaluation confirmed that WBPH resistance in NH-6, D-209, FN-709 and XS-04, which have sucking inhibitory trait, was as strong as that in CJ-06. On the basis of the genealogical relationship, it is apparent that the sucking inhibitory trait was inherited from NH-6 to CJ-06 through FN-

709 or D-209, XS-04 and XS-620. However, the sucking inhibitory trait could not be traced back to the parents of NH-6. The parents, NK-58 and LHD, were not sucking inhibitory. This conflict must be clarified by further investigations. C-21 and XH-24 were also derived from NH-6, but they did not carry the sucking inhibitory trait. However, WBPH populations were effectively suppressed on C-21 and XH-24 by ovicidal response that was inherited from NK-58, BLL and possibly NK-57 (Kinmaze, a Japanese japonica).

It is noteworthy that the japonica varieties that express field resistance to WBPH by either sucking inhibitory- and/or ovicidal traits are included in the pedigree of CJ-06. However, it has not been noticed that these japonica varieties were resistant to WBPH. In particular, NH-6 was a leading variety released in 1965, and widely planted covering the plains along the middle and lower reaches of Changjiang River in 1970–1985 without awareness of its potential resistance to WBPH<sup>[5]</sup>, probably because WBPH had not yet emerged as an economic insect pest. Nevertheless, the sucking inhibitory resistance in NH-6 was inherited unnoticed to XS-04 and XS-620, which were bred in 1980 and 1986, respectively, through intermediate breeding lines FN-709 and D-209. Besides, combining sucking inhibitory- and ovicidal traits consequently strengthened WBPH resistance in XS-620 by multiple crosses among different intermediate parents. XS-620 had originally been bred as japonica rice with the brown planthopper resistance<sup>[1]</sup>. After WBPH infestations became serious, XS-620 was found moderately resistant to WBPH<sup>[11]</sup>. Compound resistance to WBPH in XS-620 was inherited to CJ-06 by chance without intentional selection.

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