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Experimental Selection of a Brown Planthopper Population on Mixtures of Resistant Rice Lines

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Summary

The adaptation of the brown planthopper (BPH) to mixture of resistant rice lines was studied. The BPH population which had no ability to infest any resistant rice varieties in Japan was reared on the three plots of single resistant lines and the four mixtures of two to three resistant lines. BPH adapted to the single resistant lines by the fifth generation of selection and to the mixtures by the ninth generation. The BPH colonies selected on the mixtures were more virulent than those on the single resistant lines, which enabled to infest all the component lines. These results revealed that the mixtures of two to three BPH resistant lines were only effective to delay the development of the varietal resistance-breaking populations for some genearations but ineffective to prevent it.

Key Words : *Oryza sativa*, insect resistance, multiline, rice brown planthopper, biotype, break-down.

Introduction

Brown planthopper, *Nilaparvata lugens* Stål. (abbr. BPH) is the important pest of rice in temperate and tropical Asian countries. BPH sucks the sap of rice plants and causes the damage symptoms, which are commonly referred to as hopper-burn. BPH also transmits rice grassy stunt virus and rice ragged stunt virus. Both the direct damage and the transmitted viral diseases bring the serious yield losses of rice production. Among various methods for controlling BPH, resistant varieties are the most economical in tropical Asian countries (Heinrich 1979).

Varietal resistance in rice to BPH has been extensively studied at the International Rice Research Institute (IRRI) and other national research institutions. The first BPH resistant variety IR26 with *Bph1* gene was released in 1974 from IRRI, but its genetic resistance was lost within three years after release due to the occurrence of IR26-defeating BPH population in the Philippines (Feuer 1976). IR36 with the *bph2* gene was also succumbed to more virulent BPH population (IRRI 1982).

The multiline variety, a mixture of isogenic lines with different resistance genes, is a possible countermeasure to overcome the vulnerability of single resistance genes to plant diseases (Allan *et al*. 1983, Frey *et al*. 1988). In this paper, we tried to examine experimentally the effectiveness of mixtures of resistant rice lines with different genes for resistance on the delay of development of varietal resistance-breaking BPH populations.

Materials and Methods

Three rice lines with single resistance genes registered by the Ministry of Agriculture, Forestry and Fisheries were used as materials for BPH selection test. They were Rice Norin-PL 3 (abbr. PL3), Rice Norin-PL 4 (PL4) and Rice Norin-PL 7 (PL7) with the resistance genes *Bph1, bph2* and *bph4*, respectively (Kaneda *et al*. 1985, Kaneda *et al*. 1986, Nemoto *et al*. 1988). Seven selection plots were prepared: one was composed of three lines, three were of the combination of two resistant lines and three were of each of single resistant lines (Table 1). In the mixture plots, seeds of each component line were mixed at the equal rate. Eight hundred seeds in each plot were sown in a seedling tray of 26.0 × 30.0 × 3.0-cm. The seedling tray with seedlings at two-leaf stage was introduced into the insect cage of 30.0 × 30.0 × 43.0-cm. The BPH population which was collected in the Central Agricultural Experiment Station field at Koonosu, Saitama in 1975 and maintained on the susceptible varieties by Ito and Kishimoto (1981) was employed as the original population. This BPH population had no ability to infest any resistant rice varieties in Japan. Four hundred nymphs at second instar of the original population were released in each cage. The cages were placed for 16 hours at 27 °C under fluorescent lamps and for remaining eight hours at 23 °C in the darkness. Newly-emerged BPH adults in

Table 1. Host lines used for BPH biotype selection

	Plot combination	Component line	Resistance gene
1	Single	PL3	*Bph 1*
2	Single	PL4	*bph 2*
3	Single	PL7	*bph 4*
4	Two-line mixture	PL3+PL4	*Bph 1+bph 2*
5	Two-line mixture	PL3+PL7	*Bph 1+bph 4*
6	Two-line mixture	PL4+PL7	*bph 2+bph 4*
7	Three-line mixture	PL3+PL4+PL7	*Bph 1+bph 2+bph 4*
8	Check	Musashikogane	None

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each test plot were transferred onto the same new seedlings for ovipositon. Four hundred second- instar nymphs of the next generation were then released onto the test plot again. This process was repeated for 12 generations and the emergence rate was recorded in every other generation.

The varietal preference of BPH colonies selected on each plot was evaluated under the free choice condition among the seedlings of resistant lines PL3, PL4, PL7 and the susceptible check variety Musashikogane. Germinated seeds of the lines were sown in a soil-filled tray of 15.5×26.0×3.0-cm, where each line was represented by a row of 13 seeds. The experiment was replicated three times. Each tray with seedlings at two-leaf stage was infested with about 1,000 second- to third-instar nymphs of each selected colonies. Original population was included as a control. The number of BPH nymphs settled on each line was counted at intervals of 12 hours for three consecutive days. The cumulative number of BPH nymphs on a line which was compared to that on Musashikogane was used as a selection index of preference for the respective test lines.

Results

The emergence rate of adult BPHs on the susceptible check variety Musashikogane was approximately 80% through 12 generations. The emergence rates in the first generation on the single resistant lines ranged from 36% to 58%, and they were siginificantly lower than that on Musashikogane (Fig. 1). However the rates increased as the generations progressed, and reached approximately 80% in the fifth generation, comparable to that on Musashikogane. The emergence rates in the first generation on the mixtures of resistant lines ranged from 15% to 51% (Fig. 2). The rates increased more slowly than those on each of the single resistant lines, and the rates in the fifth generation on the mixtures of the resistant lines still ranged from 55% to 68%. The rates, however, reached 86% by the ninth generation.

Table 2. Preference by the selected BPH colonies on single host lines

Line	Resistance gene	Preference[1]			
		PL3-C[2]	PL4-C[2]	PL7-C[2]	Original-C
Norin-PL3	*Bph 1*	110 b	70 a	75 a	69 a
Norin-PL4	*bph 2*	102 b	105 b	92 b	66 a
Norin-PL7	*bph 4*	62 a	88 ab	96 b	63 a
Musashikogane	+	100 a	100 b	100 b	100 b

[1] Preference=(Cumulative number of nymphs on respective lines)/(Total number of nymphs on Musashikogane)×100.
[2] 'C' of PL3-C means a colony which was selected on its host resistant lines after 12 generations. Original-C means that the original population had no ability infest any resistant varieties.
[3] Means with the same letter are not significantly different at the 5 % level of probability by Duncan's multiple range test.

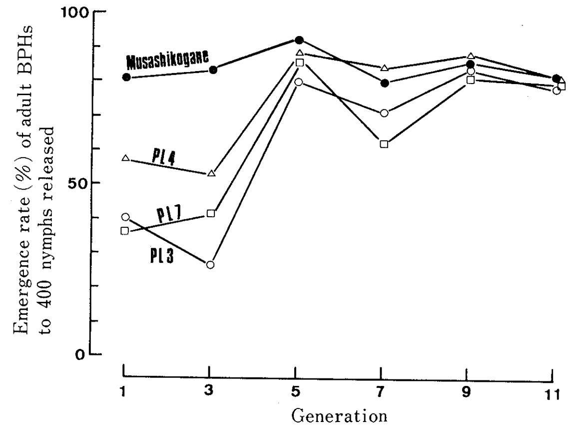

Fig. 1. Change of the emergence rate of adult BPHs selected on single resistant lines

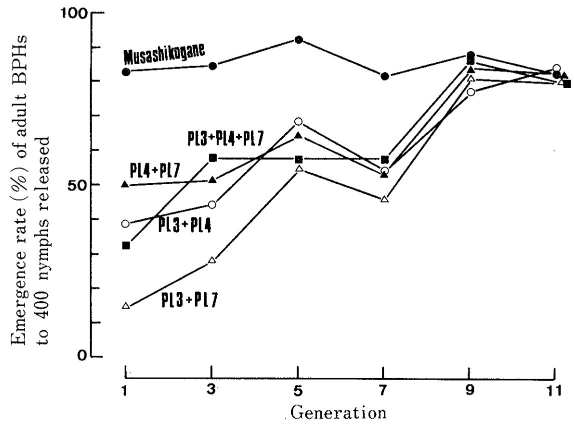

Fig. 2. Change of the emergence rate of adult BPHs selected on the mixtures of resistant lines

Table 2 shows the differential preference of each BPH colony on the test lines with different resistance genes. The numbers of BPHs of original population settled on resistant lines were significantly less than that on the susceptible Musashikogane. The PL3-colony which was selected on PL3 settled with the comparable number on PL3, PL4 and Musashikogane, but significantly less on PL7. The PL4-colony settled more on PL4, PL7 and Musashikogane but less on PL3. The numbers of the PL7-colony settling on PL4 and PL7 were comparable to that on Musashikogane but less on PL3. In most case, the selected colonies settled well on the host lines to which they were selected. However some colonies settled well on the non-host lines to which they were not selected, too. This could be seen by the numbers of BPHs of the PL3- and PL7-colonies on PL4, and of the PL4-colony on PL7.

The BPH colonies which were raised on the mixtures of the resistant lines settled more on their component lines equally to the susceptible check variety Musashikogane but less on the non-host lines (Table 3).

Table 3. Preference by the selected BPH colonies on the mixtures of resistantlines

Line	Resistance gene	Preference[1]				
		PL3+PL4-C[2]	PL3+PL7-C	PL4+PL7-C	PL3+PL4+PL7-C	Original-C
Norin-PL3	*Bph 1*	94 ab	113 b	70 a	112 a	69 a
Norin-PL4	*bph 2*	92 ab	80 a	80 ab	110 a	66 a
Norin-PL7	*bph 4*	78 a	119 b	82 ab	94 a	63 a
Musashikogane	+	100 b	100 b	100 b	100 a	100 b

1) Preference=(Cumulative number of nymphs on respective lines)/(Total number of nymphs on Musashikogane)×100.
2) 'C' of PL3+PL4-C means a colony which was selected on its component resistant lines after 12 generations. Original-C means that the original population had no ability infest any resistant varieties.
3) Means with the same letter are not significantly different at the 5 % level of probability by Duncan's multiple range test.

Discussion

The fluctuation on virulence of BPH population was proved by the experimental selection tests. A BPH colony which survived on the resistant variety Mudgo with the *Bph1* gene was established after ten generations in the experimental selection (IRRI 1970). Ito and Kishimoto (1981) selected the BPH colonies attacking Mudgo or ASD7 with the *bph2* gene after the consecutive selection for six to eight generations and found that these BPH colonies preferred their respective host varieties similarly to a susceptible variety. In the present study in which adult emergence rate from second instar nymphs was used as an index of adaptation by BPH, the single resistant lines kept their resistance only for four generations. The emergence rate in our experiment ignored the death rate of nymphs from hatching before second instar. It is possible that the adaptation at every generation was evaluated too high. However it is sure that the varieties with single BPH resistance genes easily lost their resistance by the occurrence of the varietal resistance-breaking BPH populations for a short term.

After the selection for 12 generations, the PL3- and the PL7-colonies were virulent to the non-host line PL4 with the *bph2* gene, while the PL3+PL7-colony was not virulent to PL4. Cheng (1977) and Lee and Choi (1981) obtained the selected colonies for *Bph1* which could attack the varieties with both *Bph1* and *bph2*. On the contrary, IRRI (1970) and Ito and Kishimoto (1981) reported that their selected colonies adapted only to the host varieties used. This contradiction possibly arises from the differences of host varieties or original BPH populations used in the respective selection experiments. Studies on the genetical and physiological nature of resistance genes are further needed.

Multiline varieties are considered to be able to control disease and pest progress and stabilize crop production by its genetic diversity (Browning and Frey 1969). The genetic diversity, however, possibly causes the development of super-races or super-biotypes attacking all the genes present in a multiline variety (Yokoo 1974). On the other hand, Borlaug (1959) stated that an appearance of super-races was impossible because wheat and rust had been kept balance and coexisted. Our results showed that the suppression of adult emergence lasted for four additional generations on the mixtures of two to three BPH resistant lines. However the selected BPH colonies on the mixtures had the ability to attack all the genes of the component lines used. Therefore the mixed cultivation of resistant lines can prolong the adaptation period by another some generations. However, the mixture is not effective to stabilize varietal resistance and has a high possibility to create the BPH colonies with wide virulence.

The cultivation of BPH resistant varieties included multiline should be one part of the integrated pest management in the tropics where rice is grown and BPH survives through the whole year. On the other hand, under the natural conditions in Japan where BPH cannot overwinter, the continuous selection for adaptation is not realistic for BPH and the single-variety cultivation keeps the effectiveness of resistance to BPH except when new virulent BPH colonies fly into this country. The resistant varieties with *Bph1* gene have never been cultivated at large area in Japan. However, the wild type BPH population got virulence to the resistant varieties with resistance gene *Bph1* from 1987 to 1990. These BPH colonies was suspected to migrate from south China and north Vietnam (Sogawa 1992). The mixture cultivation of resistant varieties is rather useful for the prevention of direct BPH damage in such areas as Japan where unknown biotypes come flying from overseas at every crop season.

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