Resistance factors of pecky rice incidence caused by the rice stink bugs (Leptocorisa chinensis, Nezara viridula) in rice line CRR-99-95W

Kazuhiko Sugiura\textsuperscript{a}, Takao Ōi\textsuperscript{b}, Toshiharu Tanaka\textsuperscript{b}, Aoi Hamagishira\textsuperscript{a}, Rachana Ouk\textsuperscript{c}, Mitsuru Nakamura\textsuperscript{a}, Yasuto Ide\textsuperscript{a}, Kengo Tsuda\textsuperscript{a}, Akira Ito\textsuperscript{a} and Akira Yamauchi\textsuperscript{b}

\textsuperscript{a}Dissemination Strategy Department, Aichi Agricultural Research Center, Nagakute, Japan; \textsuperscript{b}Graduate School of Bioagricultural Sciences, Nagoya University, Nagoya, Japan; \textsuperscript{c}Agricultural Extension Center, Ama Agriculture, Forestry, and Fisheries Office, Tsushima, Japan;

ABSTRACT

Pecky rice incidence caused by rice stink bugs greatly decreases the quality of rice grain and has come a big problem not only in Japan but also many other rice producing countries recently. In this study, antixenosis and tolerance were examined as the factors of resistance to rice stink bugs attack of rice line CRR-99-95W. Two experiments were conducted to determine a possibility of antixenosis involvement as a factor of the resistance in CRR-99-95W. The results showed no correlation between the number of Leptocorisa chinensis parasites and pecky rice incidence. Furthermore, no significant difference was found between the sucking frequency of Nezara viridula on mature husks of CRR-99-95W and that on the check genotypes. These results suggest that CRR-99-95W does not exhibit antixenosis effect against rice stink bugs. Then, to examine the tolerance factor involved in the resistance mechanism, the husks structures were analyzed. The results showed that the packing ratio of cell wall of the sclerenchyma fibers in the palea tended to be higher in CRR-99-95W compared to the check genotype. In addition, the width of the hook openness of the palea of CRR-99-95W was narrower than that of the check genotype. Such morphological characteristic of CRR-99-95W may play an important role in its resistance against stink bug attack.

KEYWORDS

Hooking portion; husk; oryza sativa L; sclerenchymatous fiber; tolerance; antixenosis

CONTACT Kazuhiko Sugiura kazuhiko_sugiura@pref.aichi.lg.jp

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Introduction

Pecky rice incidence caused by rice stink bugs has been on the raise in Japan since 1990 (Ito, 2004; Yamashita, 2008). The presence of pecks on rice kernels reduces their commercial value due to the deterioration of its appearance and quality, thereby decreasing farmer’s income severely. In addition, rice stink bug infestation induces the discoloration by several fungi being introduced at the time of the feeding (Lee et al., 1993), and then causes severe yield losses, which is a major problem worldwide (Jahn et al., 2004; Swanson & Newsom, 1962; Tindall et al., 2005). Although a few genotypes have been reported as resistant to stink bug of pecky rice such as PSBRc20, IR64 (Jahn et al., 2004), these genotypes are difficult to utilize for breeding because they have only moderate resistance. Therefore, we additionally screened rice genotypes that can be used for breeding new resistant cultivars against stink bugs, and then we selected one cultivar Milyang 44 and one breeding line CRR-99-95 W (Sugiura et al., 2017). The two genotypes exhibited the resistance to rice stink bug infestation by showing suppressed pecky rice incidence caused by Leptocoris chinensis with main infestation on the hooking portion of spikelet (hooking-attacking stink bugs; Takeuchi et al., 2004b) and Nezara viridula with infestation on any parts of the spikelet (indiscriminately-attacking stink bugs; Kawamura, 2007). It was suggested that the resistance in Milyang 44 would be related to their husk structures (Nakamura et al., 2017, 2020) while the responsible factors for resistance to pecky rice caused by stink bug in CRR-99-95 W have not been clearly identified.

Painter (1951) listed three factors that are involved in the mechanism of insect resistance in crops: (1) nonpreference, (2) antibiosis, and (3) tolerance. Later, Kogan and Ortman (1978) proposed that ‘nonpreference’ should be termed ‘antixenosis’, because ‘nonpreference’ refers to the characteristics of the insect to the crop, whereas ‘antixenosis’ refers to the property of not being preferred as a host plant to insects. Antibiosis refers to a plant’s ability to decrease survival rate of insects, retard growth of them, and inhibit egg laying. Tolerance refers to the ability of a host plant to withstand and minimize insect damage.

We previously examined the antibiosis of CRR-99-95 W, of which four panicles grown in pots were covered with a net (0.5 m wide and 0.6 m long), and five adult L. chinensis were released inside for 7 days. As a result, none of the stink bugs died, in other words they could survive on the plants for that period (Sugiura et al., 2018), therefore, resistance of CRR-99-95 W would not be due to antibiosis but may be attributed to antixenosis, tolerance, or a combination of both.

Regarding antixenosis, Ando and Kishino (1981) reported a resistance assay on the antixenosis of green leafhopper (Nephotettix cincticeps), which evaluated insect resistance basing on the number of the parasites on plants. Sogawa et al. (1999) reported that rice genotype Chenjian06 showed low number of parasites of white-backed planthopper (Sogatella furcifera). Sogatella furcifera was inhibited from sucking Chenjian06 plant due to less excrete of honeydew as compared with other genotypes. Therefore, they concluded that Chenjian06 has antixenosis and antibiosis against Sogatella furcifera. If antixenosis or antibiosis is involved in the factors for the resistance to pecky rice incidence, the number of stink bug parasites that infect the host plant is expected to be fewer in resistant than susceptible genotypes.

Tolerance was reported to be closely related with various plant structural traits such as spines, trichomes, waxy cuticles, silica accumulation, which can act as a physical barrier to insect attachment, feeding, and oviposition (Figueiredo et al., 2013; White & Eigenbrode, 2000). For example, Sasamoto (1958) reported that the application of silica to rice plants increased the rigidity of shoots and reduced the damage caused by the rice stem borer (Chilo suppressalis). Nakamura et al. (2020) focused on the structure of husk of Milyang 44 as a factor of tolerance because the resistance to pecky rice incident increased with the maturity of husk. As a result, they found that the cell wall of the sclerenchymatous fibers of the husk were thicker than that of the check genotype, which made the husk harder and thus might act as a tolerance factor for the resistance mechanisms to the stink bugs (Nakamura et al., 2020). Regarding the hardness of the husks, Seo & Ota (1983) reported that the accumulations of silica and lignin in the sclerenchymatous fibers were the main factors that contributed to the physical hardness of the husks. Since both silica and lignin accumulate as rice plant matures (Seo & Ota, 1983; Yoshida, 1965), it is highly likely that these factors are related to the resistance of N. viridula that attacks any portions on the husk surface. L. chinensis that inserts its stylet into the gap of the interlocking of the edges of palea and lemma (Furuike & Kiyota, 1993) injures the hooking portion of the husk (Figure 1a). Therefore, the structural strength of the hook-shaped edges (Figure 1b) are considered to be one of the factors reducing pecky rice incidence.

Another tolerance factor in resistance is the structure of panicle. It is known that the order of flowering and thus ripening among spikelets in a panicle is fixed, and that secondary and basal rachis-branch spikelets flower and ripen slower than those on the primary rachis-branch (Hoshikawa, 1989). The structure of panicle would be involved in spikelet maturity and hardness,
secondary and basal rachis-branches spikelets that flower later are expected to have less mechanical tolerance than susceptible genotypes.

In the present study, we examined the antixenosis and tolerance to identify the causal factors of resistance against stink bugs for CRR-99-95 W. For the evaluation of antixenosis, the number and sucking frequency of stink bugs were compared between CRR-99-95 W and the check genotypes. For tolerance, we compared the hardness of the husk between the genotypes; specifically, the sclerenchymatous fibers and silica content, which are the main factors of the husk hardness, and the morphology of the hooking portion of the husk, where L. chinensis mainly injures. For the involvement of panicle structure, we examined the relationship between spikelet ripening and stink bug damage on the grains by counting the number of spikelets and damaged grains on different rachis-branches in a panicle.

**Materials and methods**

**Plant and bugs preparation**

Rice (Oryza sativa L.) genotypes were grown in the experimental field of the Aichi Agricultural Research Center (AARC) (35°16 N, 137°06E) in Japan. Two species of stink bugs were used in this study; N. viridula were obtained from population reared on dried soybean in the environmentally controlled chamber (25°C, 16 h light: 8 h dark) at Nagoya University in Nagoya, Japan. L. chinensis were obtained from the weed areas in the AARC and reared in grass weeds covered with mosquito nets in a glass house at the AARC.

**Antixenosis examination**

The number of stink bug parasites and pecky rice incidence were determined at the AARC in 2011 with a group screening method using eleven genotypes; CRR-99-95 W, Aichinokao SBL, Nipponbare, Yumematsuri, Yan Xuan, Yan Xuan 203, Nakeng 11, Zaiyeqin 8, GP 206, GP 242, and Dian Yu 1.

The group screening method was conducted using the following procedures. First, we grew one plant in a plastic pot (diameter, 113 mm; height, 115 mm, Fujiwara Scientific, Tokyo, Japan), used two pots for each genotype, and cut off the other panicles, leaving four panicles of average size for each plant at approximately 20 days after heading. Then, the pots with plants were placed in mosquito nets and exposed to feeding by stink bugs for seven days. After maturity, the panicles were harvested and visually examined for pecky rice incidence. The number of stink bug parasites per plant...
was counted in the morning of any two days during the period of stink bug attack on the plants, excluding the day of the insect release.

The sucking frequency by stink bugs was also counted. Spikelets of CRR-99-95 W and Aichinokaori SBL (check genotype) grown in 2013 were collected at maturity. Four spikelets were placed in one case in the environmentally controlled chamber (25°C, 16 h light: 8 h dark) at Nagoya University. Ten *N. viridula* adults were randomly selected and released into the chamber, and the number of times that the bugs attempted to suck on the spikelets was counted from video camera recordings, and recorded as the sucking frequency. The recording time of the video camera was 12 h. Four tests were conducted using different plants of spikelets for each test.

**Husk hardness measurement**

As mentioned above the group screening method for *L. chinensis* was conducted at the AARC in 2012. Eight genotypes were tested by using two pots with one plant: CRR-99-95 W, Aichinokaori SBL, Koshihikari, Yan Xiang, Yan Xiang 203, Nakeng 11, Zaiyeqin 8, and GP242. The husks of ten spikelets from the first to third primary rachis-branch of the panicle that were not used in the group screening method were used in this experiment. The hardness of the collected husks was measured using a rheometer (CR-3000; Sun Scientific, Tokyo, Japan). A cotton needle (0.84 mm thick) was used as a plunger, and the measurement was conducted at a speed of 100 mm min⁻¹ and a depth of 2.0 mm.

**Silica content measurement**

The silica content was measured for the husks of CRR-99-95 W and Aichinokaori SBL that were sampled in 2019. The panicles were sampled 20 days after heading and divided into husks and grains after drying. The samples were decomposed in a nitric acid-perchloric acid mixture and then filtered. After the filter paper was dried and moistened with ethanol, it was carbonized in an electric furnace, and the residuals were weighed after cooling (Crops Analysis Committee, 1975).

**Grain husk histology**

Aichinokaori SBL, CRR-99-95 W, and D15 (Koshihikari/ CRR-99-95 W//Koshihikari) progeny line of CRR-99-95 W that was screened against *L. chinensis*, were grown at the AARC in 2020, and plants that heading date at the same time were used in the experiment. To check the stink bug resistance of each genotype, a group screening method was conducted with three pots by exposing the genotypes to *L. chinensis* as described above.

The husks of the plants that were not used in the group screening method were collected at 21 days after heading and preserved in formalin-acetic acid-alcohol (FAA) fixative solution (1:1:18). The central part of the palea of the prefixed husk was cut, rinsed in 50 mM sodium phosphate buffer (pH 7.2), and then post-fixed with 2% osmium tetroxide in the same buffer. The double-fixed segments were then dehydrated in acetone and embedded in resin (Agar Low Viscosity Resin, Agar Scientific, UK) following the standard methods for transmission electron microscopy (TEM) (Oi et al., 2012). Because the husk was too hard to cut, it was difficult to prepare ultra-thin sections (< 100 nm thickness) for TEM observation. Therefore, in this study, we utilized the semithin section scanning electron microscopy (SEM) (Koga et al., 2018) to observe rice husk structures with high resolution. The segments embedded in resin were cut into serial transversal sections (1.0 µm thick) using a diamond knife with a large water trough (SYM-Jumbo, Syntek, Japan) mounted on an ultramicrotome (EM UC6, Leica, Germany) and placed on glass slides, as described in detail by Ouk et al. (2020). Then the serial sections on glass slides were stained with 2% uranyl acetate for 20 min and lead staining solution (Sigma-Aldrich) for 4 min, and then were coated with a thin layer of gold using a sputter coater (IB-3; Eiko, Japan) for less than 10 seconds to prevent electron charging. Then, the glass slides were placed on a tabletop scanning electron microscope (TM3000; Hitachi, Japan), and stained-section surfaces were observed with the high-intensity mode. The sections were observed serially, and the three in each 10–20 section from one serial section of one palea were image-captured. Then, the digital images were analyzed using the Image-Pro Premier 3D software (Ver. 9.3; Media Cybernetics, USA); the region of sclerenchyma fiber in the center of the palea was binarized to detect the cell wall area (Figure 1c). The hook-shaped curve and the nearest vascular bundle were determined, and then, the width of the hook openness (w) and the minimum distance between the hook and the vascular bundle (d) were also measured to evaluate the thickness of the hooked edges (Figure 1d) using the software. The average of the three images of each palea was used for one repetition value, and four paleas of each plant were observed for a genotype.

**Panicle morphology**

Seedlings of CRR-99-95 W and Aichinokaori SBL, which were seeded in middle of April, were transplanted on each hill in the experimental field in middle of May 2017.
Ten panicles were collected from each plant at maturity for each genotype, and rachis-branches on the panicle were classified as tip rachis-branches (first to third rachis-branch from the tip), middle rachis-branches (fourth to seventh rachis-branch), and basal rachis-branches (eighth to lower rachis-branches). Then the number of spikelets on each of primary or secondary rachis-branch was recorded.

**Pecky rice on spikelet on rachis-branches of different positions**

The panicle in average size of CRR-99-95 W and Koshihikari grown in the field in 2013 were collected at 20 days after heading. The panicles were separated into tip and basal rachis-branches according to the above-stated criteria, and four spikelets of each primary rachis-branch were used for the experiment. In the environmentally control chamber (25°C, 16 h light: 8 h dark), ten of *N. viridula* adults were randomly selected and released into a plastic cylinder case measuring 129 mm in diameter and 97 mm in depth (Risu Co., Ltd., Gifu, Japan) with a lid. Forty-eight hours after release, the husks were removed, and the grains damaged by stink bugs were counted. Five tests were conducted using different plants for each test.

**Statistical analysis**

Statistical analysis was performed using Excel Statistics 2008 (Social Information Service, Japan).

**Results**

**Antixenosis examination**

The group screening method using eleven genotypes showed that the number of *L. chinensis* parasites ranged from 2.3 – 18.5 per plant, and the pecky rice incidence varied greatly from 8% to 75%; there was no correlation between the number of parasites and the pecky rice incidence (**Figure 2**). The number of stink bug parasites on CRR-99-95 W tended to be higher than that of the other nine genotypes, including Aichinokaori SBL, however the pecky rice incidence tended to be lower in CRR-99-95 W. There was no significant difference in the sucking frequency by *N. viridula* on mature grains between CRR-99-95 W and Aichinokaori SBL (**Figure 3**).

**Husk hardness and silica contents**

There was a negative correlation significantly between the husk hardness and the percentage of pecky rice incidence at 20 days after heading (*r* = −0.79, *p* < 0.05; **Figure 4**). CRR-99-95 W had the highest husk hardness and the lowest incidence of pecky rice among the eight genotypes tested, and the incidence of pecky rice tended to decrease as the husk hardness increased.
Husk histology

In addition to CRR-99-95 W and Aichinokaori SBL, as another check genotype, D15 that is the progeny line of CRR-99-95 W was also added for this investigation. D15 showed similar resistance as CRR-99-95 W in the group screening method using *L. chinensis* (Figure 6). The three genotypes were investigated for histological examinations as follows (Figure 7). Figure 7a-c shows SEM images of the middle section of the palea at 21 days after heading. We measured the ratio of cell wall to cytoplasm in area and calculated the mean of cross-sectional area of the cell wall in the sclerenchymatous fiber, and the results showed that the areas of CRR-99-95 W and D15 were larger than that of Aichinokaori SBL, although the difference was not significant between CRR-99-95 W and Aichinokaori SBL (Figure 7g).

Figure 7d-f shows SEM images of cross-sections of the hooking portion of the palea at 21 days after heading. The width of the hook openness in both CRR-99-95 W and D15 were significantly narrower than that of Aichinokaori SBL (Figure 7h). The distance between the hook-shaped curve and the vascular bundle in the edge of palea (Figure 1d) were significantly longer in CRR-99-95 W than in Aichinokaori SBL (Figure 7i).
Figure 7. Semi-thin transversal sections of the palea sampled at 21 days after heading. Sections (1.0 μm thick) embedded in resin were observed by SEM. (a-c) The middle part of the palea, (d-f) the hooking portion of the palea. (a, d) CRR-99-95 W, (b, e) D15, (c, f) Aichinokaori SBL. Scale bars = 50 μm. (g) packing ratio of cell wall of the sclerenchyma fibers; percentage of the cell wall area to the total cells area, (h) opening width of the curve in the hooking portion, (l) the distance between the hook-shaped curve and vascular bundle in the hooking portion. Bars indicate standard deviation (n = 4). Different letters indicate significant differences at a 5% probability level in Tukey’s multiple comparison test.

Panicle morphology and differences in pecky rice incidence on rachis-branch of different positions

CRR-99-95 W had more secondary rachis-branches than Aichinokaori SBL (check genotype). There was no difference in the percentage of tip rachis-branches between the genotypes examined, but the percentage of lower rachis-branch spikelet was significantly higher in CRR-99-95 W than in Aichinokaori SBL (Table 1). Pecky rice incidence by *N. viridula* spikelets at 20 days after heading is shown in Figure 8. In both genotypes, the damage to the basal rachis-branch spikelets was significantly higher than that to the tip spikelets. However, CRR-99-95 W had significantly fewer pecky rice incidence compared to Koshihikari, as a check genotype, on the same rachis-branch spikelet.

Table 1. Number of spikelets on rachis-branches on different positions in a panicle.

| Tip rachis-branch | Middle rachis-branch | Basal rachis-branch |
|-------------------|----------------------|---------------------|
| Genotype          | Primary rachis-branch spikelet number | Secondary rachis-branch spikelet number | Tip rachis-branch spikelet number | Secondary rachis-branch spikelet number | Primary rachis-branch spikelet number | Secondary rachis-branch spikelet number | Primary rachis-branch spikelet number | Secondary rachis-branch spikelet number |
| CRR-99-95 W       | 18**                 | 23**                | 31**                | 25**                | 31**                | 43**                | 22**                | 12**                | 26**                |
| Aichinokaori SBL  | 17                   | 5                   | 31                   | 23                   | 14                   | 53                   | 9                   | 3                   | 16                   |

Tip rachis-branch, first to third rachis-branch; middle rachis-branch, forth to seventh rachis-branch; basal rachis-branch, eighth or lower rachis-branch. **, *, and n.s. indicate a 5% probability level, 1% probability level, and no significance, respectively. The percentage of rachis-branch spikelet was inverse sine transformed, and then a t-test was conducted (n = 10).
Figure 8. Pecky rice incidence caused by *Nezara viridula* at 20 days after heading on rachis-branches of different positions in a panicle. **, and n.s. indicate a 1% probability level, and no significance, respectively. According to the two-way ANOVA (n = 5).

**Discussion**

**Antixenosis of CRR-99-95 W against the pecky rice stink bug**

In relation to antixenosis, no correlation was found between the number of *L. chinensis* parasites per plant and the pecky rice incidence for the eleven genotypes (Figure 2). Furthermore, the number of stink bug parasites was second highest in CRR-99-95 W (9.5 plant⁻¹) among the genotypes ranging from 2.3 plant⁻¹ to 18.5 plant⁻¹, while pecky rice incidence was lowest in CRR-99-95 W. These results suggest that CRR-99-95 W shows higher resistance but less antixenosis against stink bug. In the group screening method (Figure 2), *L. chinensis* were reared in the closed net; therefore, the number of parasites varied depending on the number of plants tested in the net, and the number of spikelets per panicle also depended on the genotypes. Although the number of stink bug parasites was not equal among the genotypes used in the group screening method (Figure 2), the number of stink bugs per spikelet was kept constant in the laboratory for the sucking-frequency test, which showed no significant difference between the genotypes (Figure 3). This result also supports that CRR-99-95 W shows less antixenosis against stink bug. In addition, this study investigated two types of stink bugs; *L. chinensis* is hooking-attacking stink bugs (Takeuchi et al., 2004a); *N. viridula* is indiscriminately-attacking stink bugs (Kawamura, 2007). CRR-99-95 W did not show antixenosis against both types of stink bugs, therefore the resistance of CRR-99-95 W to pecky rice stink bug would not be attributed to antixenosis. In addition, previous experiments showed that CRR-99-95 W had no antibacterial properties (Sugiura et al., 2018), which was supported by this study.

**Tolerance of CRR-99-95 W against the pecky rice stink bug**

In the tolerance experiment, there was a significant and negative correlation between the hardness of the husk and the incidence of pecky rice (Figure 4). As the hardness increased, the incidence of pecky rice decreased. Therefore, we investigated the silica content and the sclerenchymatous fibers, which were reported to be involved in the hardness of husk (Seo & Ota, 1983, Yoshida, 1965), and found that the silica content of CRR-99-95 W was not higher than that of the check genotype (Figure 5). On the other hand, the packing ratio of cell wall of the sclerenchymatous fiber in the palea was higher in CRR-99-95 W and D15 than in Aichinokaori SBL (Figure 7g). The variations (standard deviation) of packing ratio of cell wall of the sclerenchymatous
fiber were found to be smaller in both CRR-99-95 W and D15 than in Aichinokaori SBL. Indiscriminately-attacking stink bugs look for areas on the surface of husk that are easy to perforate and suck (Takeuchi et al., 2004a). CRR-99-95 W and D15, which showed less variations in packing ratio of cell wall of sclerenchymatous fibers, are less likely to be injured by indiscriminately-attacking stink bugs because the hardness of the husk is uniform and there are less areas where are easy to suck. In the case of Milyang 44, the thickness of sclerenchymatous fibers is also considered as one of the factors of resistance mechanism (Nakamura et al., 2020). Although a significant and negative correlation was recognized between the husk hardness and the pecky rice incidence among the eight genotypes (Figure 4), the difference in packing ratio of sclerenchymatous fibers between the CRR-99-95 W and Aichinokaori SBL was smaller (Figure 7g) as compared with the large difference in the incidence of pecky rice between them (Figure 4). Therefore, the factors of resistance mechanisms against indiscriminately-attacking stink bugs cannot be fully explained only by the thickness of sclerenchymatous fibers and thus need to be further investigated.

We also found out the structural difference of the hooking portion of the palea (Figure 7d-f). L. chinensis sucks mainly from the gap of the hooking portion (Furuike & Kiyota, 1993; Takeuchi et al., 2004b), where the edges of lemma and palea are interlocked. Furthermore, the outer surfaces of the palea edge lack the siliceous protuberances (Mezaki, 2018). Therefore, the hook-attacking stink bugs suck by extending their stylets, being bent with the hook, more than the gap (Nakamura et al., 2020), or penetrating the structurally weak area of the palea edge (Takeuchi et al., 2004b). Both of CRR-99-95 W and D15 had significantly smaller width of the hook openness than that of the check genotype Aichinokaori SBL (Figure 7h), suggesting that a narrow gaps in the hooking would hamper the insertion of L. chinensis stylet. In addition, the thickness of the hard part just under the hooked-shaped curve (the nearest distance between the curve to the vascular tissue in the edge) in CRR-99-95 W was significantly longer compared to that in Aichinokaori SBL and that in D15 ranged between the two (Figure 7i); suggesting that more robust hooking structures would prevent the penetration of L. chinensis stylet.

These results suggest that CRR-99-95 W resistance to pecky rice stink bugs is due to the tolerance supported by the characteristics of sclerenchymatous fibers and hooking portion of husk, which prevents sucking.

**Breeding of pecky rice stink bug resistant cultivars**

We previously reported that the resistance in CRR-99-95 W increased with maturity (Sugiura et al., 2018). In the present study, the survey of stink bug damages on spikelets on rachis-branches with different positions revealed that the grains on lower rachis-branches tended to show higher incidence of pecky rice (Figure 8). Generally, the order of flowering of various spikelets in a panicle depends on the position of rachis-branches that bear the spikelet in the panicle, and the secondary and lower rachis-branch spikelets flower at a later time in the panicle (Hoshikawa, 1989). The panicle of CRR-99-95 W had a greater number of basal and secondary rachis-branch spikelets (Table 1), which would be a disadvantage for resistance due to late flowering and maturing. Therefore, there is a possibility that selective breeding of the panicle structure with fewer basal and secondary rachis-branch spikelets would increase resistance in plant level. In addition, it is necessary to verify if the resistance pyramiding with that of Milyang 44, which has a similar level of tolerance against L. chinensis, would further strengthen the resistance.

In this study, we showed that the resistance of CRR-99-95 W against stink bugs was attributed to tolerance due to unique structure of its husk. However, the tolerance is the property of withstanding insect attack and reducing damage, and therefore it is difficult to completely suppress the damage of pecky rice incidence only by introducing the tolerance of CRR-99-95 W. Further studies for introducing the antibiosis or antixenosis against stink bugs are necessary to improve the resistance for reducing the damage caused by the stink bugs.

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**Disclosure statement**

No potential conflict of interest was reported by the author(s).

**ORCID**

Toshiharu Tanaka http://orcid.org/0000-0001-8145-5573
Rachana Ouk http://orcid.org/0000-0002-0109-7276
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