Control strategy for rice stripe virus transmitted by small brown plant hopper (*Laodelphax striatellus*)

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**ABSTRACT**

Rice stripe virus (RSV) causes chlorotic stripes, mottling, and necrotic streaks on rice leaves (*Oryza sativa*), resulting in a loss of production. RSV disease mainly occurs in Asian countries, including Taiwan, Japan, Korea, and China. In Japan, the disease devastated rice production from 1960 to 1985, and subsided thereafter owing to the introduction of new varieties resistant to RSV. However, its incidence has begun to increase again since 2004. Its vector, small brown plant hopper (*Laodelphax striatellus*, SBPH) persistently transmit RSV, and transmitted it from female adults to their progeny via egg. In this presentation, the molecular characteristics of transovarial transmission of RSV in SBPH are introduced. Countermeasures including adjusted timing of pesticide application and use of resistant varieties are discussed.

**Keywords**: rice stripe virus, small brown plant hopper, resistant cultivars, insecticides, control measures

**INTRODUCTION**

Rice stripe virus (RSV) causes chlorotic stripes, mottling, and necrotic streaks on rice leaves (*Oryza sativa*). Rice plants severely infected with RSV often show panicle sterility, resulting in lower yield. RSV mainly occurs in Asian countries, including Taiwan, Japan, Korea, and China. Although many Gramineae crops, such as wheat, barley, and corn can be infected with RSV, the damages are usually less severe in the fields. The rice disease caused by RSV was first identified around 1897 in Japan. Its damage on rice production was disastrous between 1960 and 1985. Owing to the introduction of varieties resistant to RSV, the disease gradually subsided and became less significant until 2004. However, the incidence has increased again since 2005.
According to the statistics reported by Japan Plant Protection Association, RSV affected more than 137,000 hectares of rice fields, which account for approximately 9% of the total cultivated area in Japan in 2015. Therefore, effective control measures are urgently needed.

According to the current taxonomy of International Committee on Taxonomy of Viruses, Rice stripe virus belongs to the Tenuivirus genus, the Phenuiviridae family. RSV genome consists of four single-stranded RNA molecules (RNA1 through RNA4), and seven genes are encoded in negative or ambisense orientation. RSV is persistently transmitted by the small brown plant hopper (Laodelphax striatellus, SBPH) and other planthoppers. The virus propagates in the insect vectors and is transmitted from female adults to their progeny at the egg stage (transovarial transmission). Transovarial transmission of RSV is considered the main reason for the high viruliferous rate in nature, which makes the control of rice stripe disease more difficult. Understanding how RSV is maintained in SBPH and how RSV is transmitted to rice plants is crucial to developing effective control measures against this disease.

In our presentation, we would like to introduce the characteristics of transovarial transmission of RSV in SBPH, and control measures of RSV using resistant cultivars and the appropriate timing for the application of insecticides.

**Molecular characteristics of transovarial transmission of RSV in SBPH**

Under experimental conditions, the percentage of eggs exhibiting transovarial transmission was estimated to be greater than 90%. The amount of RSV maintained through transovarial transmission was analyzed during the course of development and reproduction of SBPH. The expression of the RSV coat protein (CP) gene was measured by RT-qPCR as an estimate of RSV content in nymphs and adults of SBPH at various developmental stages. The 18S ribosome RNA gene of SBPH was chosen as the reference for relative quantification. Based on the relative CP expression levels, the amount of RSV did not differ significantly during the nymphal stage or between adult females of different ages although the amount of RSV tended to increase slightly as males became older. The average RSV content in males was 1.30 to 2.49 times that in females. These results indicated that RSV multiplies in synchrony with the multiplication of host cells during the nymphal stage, and the increase of RSV stopped or slowed after the insects reached the adult stage. Then, the amount of RSV in SBPH
female adults was compared between generations. The amount of RSV in three viruliferous females and their progenies was analyzed by RT-qPCR. The result indicated that RSV content did not differ significantly between parents and their progenies. (For full article of this study, see Okuda et al. 2017)

The viruliferous population of SBPH were grown on a susceptible rice cultivar Koshihikari and its near isogenic lines harboring RSV resistant gene(s), and examined whether these resistance traits affect the retainability of RSV in SBPH. The result showed the percentage of viruliferous insects was not significantly different among susceptible and resistant rice plants. Then, the transovarial transmission efficiency of SBPH grown on different hosts was examined for four generations. The percentages of viruliferous insects in the first generation grown on rice cv. Koshihikari, Asahino-yume (resistant to RSV), and wheat cv. Norin 60 were 50.0%, 46.8%, and 51.3%, respectively. The percentages decreased as generations progressed; however, the transovarial transmission efficiency was not significantly different between susceptible and resistant rice cultivars or between rice and wheat. These results indicated that the resistant properties of rice cultivars have no direct effect on the retainability of RSV in SBPH.

Control using resistant varieties

Rice cultivars resistant to viruses are widely used to minimize the damage due to virus infection. Screening of RSV resistant rice varieties was initiated in the early 1960s, and the results showed a high level of resistance in some Indica type rice varieties and Japanese upland rice varieties. A resistance gene (Stvb-i) was identified in an Indica-type rice cultivar Modan. Through breeding programs, Stvb-i gene was successfully introduced to develop resistant cultivars, which effectively control RSV disease in some areas in Japan. Currently, molecular marker-assisted selective breeding for Stvb-i made it possible to accelerate the development of new varieties. Introduction of Stvb-i into susceptible rice cultivars, Aichinokaori and Hatsushimo (designated as Aichinokaori SBL and Hatsushimo SBL), which replaced the original cultivars contributed to the reduction of pesticide usage and provide stable productions.

However, owing to the preference of Japanese consumers, some old varieties, which are not resistant to RSV, are still in use. Particularly Koshihikari, which was registered in 1956, still accounts for more than 36% of the total rice production for 2016 in Japan. A near-isogenic cultivar of Koshihikari was developed by introducing RSV resistant
genes, Stva and Stvb, originated from Japanese upland rice, Kanto72 and registered as Koshihikari Kinchushi SBL1 (SBL1) in 2012. In 2016 and 2017, SBL1 was grown in a district heavily affected with RSV in Ibaraki Prefecture to examine its practical readiness in the field. The result showed the RSV infection rate of SBL1 was less than 1% while that of Koshihikari grown in an adjacent field reached 100% at the end of the cultivation period. Now we are soliciting farmers who can engage on a large-scale trial of SBL1 cultivation.

**Control using chemical insecticides**

It has been reported that diseases caused by insect-borne viruses can be effectively decreased by the identification of the emergence of their vectors and timely application of insecticides. As for RSV, it would be possible to reduce the damage by elucidating multiple factors based on virus transmission and the occurrence ecology of vectors to control them at the optimal timing. We have studied how the RSV causes damage and spreads within paddy fields. Our investigations revealed that in a rice producing area in Ibaraki Prefecture, in central Japan, diseased plants start to appear in mid-June to early July, and from which the disease spreads from affected plants to adjacent plants throughout the season. This suggests that RSV viruliferous SBPH enters paddy fields, where they infect plants and lay eggs. Subsequently, hatched viruliferous nymphs transmit RSV to surrounding plants, thereby rapidly spreading the disease. Our analysis of the damage caused by RSV showed that the earlier the onset of disease, the more extensive damage cause due to a reduced number of healthy panicles. This suggests that to reduce damage caused by RSV, it is necessary to ensure the growth of a sufficient number of healthy panicles by controlling the vector insect during the crop’s early growth stage. To be most effective, pest control efforts should be timed to target either the first-generation adults that colonize the paddy fields or the second-generation nymphs and adults that cause the rapid increase in the number of diseased plants within a field. (For full article of this study, see Shiba et al. 2017)

The timing of SBPH emergence can be precisely estimated using a prediction program based on the total effective temperature for SBPH, which is provided by Japan Plant Protection Association. Insecticide application such as silafluofen at 100 ppm timed for the emergence of the second-generation nymph resulted in effective suppression of the disease. To make the prediction more precise, we are developing a
new program based on the total effective temperature and the Agro-Meteorological Grid Square Data System to predict the emergence of SBPH in a one square km spatial resolution. We hope this program will help farmers to obtain the optimal control measure of RSV diseases.

LITERATURE CITED

Fig. 1. (A) typical symptoms caused by rice stripe virus on rice; (B) a vector insect, Laodelphax striatellus (female).

Fig. 2. Changes of RSV affected areas from 1975-2015 in Japan.