# Peer Community In , Genomics

## Scoring symptoms of a plant viral disease

### Olivier Panaud based on peer reviews by Grégoire Aubert and Valérie Geffroy

Alexander Silva, Maria Elker Montoya, Constanza Quintero, Juan Cuasquer, Joe Tohme, Eduardo Graterol, Maribel Cruz, Mathias Lorieux (2023) Genetic bases of resistance to the rice hoja blanca disease deciphered by a QTL approach. bioRxiv, ver. 2, peer-reviewed and recommended by Peer Community in Genomics.

https://doi.org/10.1101/2022.11.07.515427

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The paper from Silva et al. (2023) provides new insights into the genetic bases of natural resistance of rice to the Rice Hoja Blanca (RHB) disease, one of its most serious diseases in tropical countries of the American continent and the Caribbean. This disease is caused by the Rice Hoja Blanca Virus, or RHBV, the vector of which is the planthopper insect *Tagosodes orizicolus* Müir. It is responsible for serious damage to the rice crop (Morales and Jennings 2010). The authors take a Quantitative Trait Loci (QTL) detection approach to find genomic regions statistically associated with the resistant phenotype. To this aim, they use four resistant x susceptible crosses (the susceptible parent being the same in all four crosses) to maximize the chances to find new QTLs. The F2 populations derived from the crosses are genotyped using Single Nucleotide Polymorphisms (SNPs) extracted from whole-genome sequencing (WGS) data of the resistant parents, and the F3 families derived from the F2 individuals are scored for disease symptoms. For this, they use a computer-aided image analysis protocol that they designed so they can estimate the severity of the damages in the plant. They find several new QTLs, some being apparently more associated with disease severity, others with disease incidence. They also find that a previously identified QTL of *Oryza sativa* ssp. japonica origin is also present in the indica cluster (Romero et al. 2014). Finally, they discuss the candidate genes that could underlie the QTLs and provide a simple model for resistance.

It has to be noted that scoring symptoms of a viral disease such as RHB is very challenging. It requires maintaining populations of viruliferous insect vectors, mastering times and conditions for infestation by nymphs, and precise symptom scoring. It also requires the preparation of segregating populations, their genotyping with enough genetic markers, and mastering QTL detection methods. All these aspects are present in this work. In particular, the phenotyping of symptom severity implemented using computer-aided image processing represents an impressive, enormous amount of work.

From the genomics side, the fine-scale genotyping is based on the WGS of the parental lines (resistant and susceptible), followed by the application of suitable bioinformatic tools for SNP extraction and primers prediction that can be used on their Fluidigm platform. It also required implementing data correction algorithms to achieve precise genetic maps in the four crosses. The QTL detection itself required careful statistical pre-processing of phenotypic data. The authors then used a combination of several QTL detection methods, including an original meta-QTL method they developed in the software MapDisto.

The authors then perform a very complete and convincing analysis of candidate genes, which includes genes already identified for a similar disease (RSV) on chromosome 11 of rice. What remains to elucidate is whether the candidate genes are actually involved or not in the disease resistance process. The team has already started implementing gene knockout strategies to study some of them in more detail. It will be interesting to see whether those genes act against the virus itself, or against the insect vector.

Overall the work is of high quality and represents an important advance in the knowledge of disease resistance. In addition, it has many implications for crop breeding, allowing the setup of large-scale, marker-assisted strategies, for new resistant elite varieties of rice.

#### **References:**

Morales F and Jennings P (2010) Rice hoja blanca: a complex plant-virus-vector pathosystem. CAB Reviews. https://doi.org/10.1079/PAVSNNR20105043

Romero LE, Lozano I, Garavito A, et al (2014) Major QTLs control resistance to Rice hoja blanca virus and its vector Tagosodes orizicolus. G3 | Genes, Genomes, Genetics 4:133–142. https://doi.org/10.1534/g3.113.009373

Silva A, Montoya ME, Quintero C, Cuasquer J, Tohme J, Graterol E, Cruz M, Lorieux M (2023) Genetic bases of resistance to the rice hoja blanca disease deciphered by a QTL approach. bioRxiv, 2022.11.07.515427, ver. 2 peer-reviewed and recommended by Peer Community in Genomics https://doi.org/10.1101/2022.11.07.515427

#### Reviews

#### **Evaluation round #1**

DOI or URL of the preprint: https://doi.org/10.1101/2022.11.07.515427 Version of the preprint: 1

#### Authors' reply, 21 March 2023

We posted the final version on the preprint server **Download author's reply** 

#### Decision by Olivier Panaud, posted 16 January 2023, validated 16 January 2023

#### minor revisions

This manuscript has been reviewed by two reviewers who consider that this work is of interest. However, they have both made some remarks in order to improve the quality of the manuscript. The authors should take these remarks into consideration before their article could be recommended.

#### Reviewed by Grégoire Aubert, 06 January 2023

The paper by Alexander Silva et al entitled "A more complete story of the genetic bases of resistance to the rice hoja blanca disease" reports a QTL approach using four biparental populations in order to understand the genetic bases of a virus disease caused by RHBV in rice. To do so, an image analysis protocol is developed to estimate the extent of symptoms. The study confirms one major QTL previously reported and identifies additional loci involved in the virus resistance. The approach allows the authors to pinpoint relevant candidate genes in the QTL confidence intervals, which will allow functional validation experiments to confirm them or not. This paper brings very valuable information for the community. One point to clarify is the plant material that has been used to measure the severity of the symptoms, as it impacts the independence of this trait from incidence. Please also see comments below,

Title: I would suggest: Genetic bases of resistance to the rice hoja blanca disease deciphered by a QTL approach

Line 62: Has the local ancestry analysis mentioned here been performed by the authors or published elsewhere? Please provide a reference.

Line 123: It is not totally clear to me on which plants the ALA is calculated: is it only on plants with symptoms identified in the previous step? Or is it on 10 random infected plants per F3 family? The mention of "plants with symptoms" is ambiguous, please clarify this point as it is important for the interpretation of the results. If done on 10 random plants, your severity measure depends strongly of the incidence rate. If done only on plants with symptoms, it would help to disconnect incidence from severity of the symptoms, and dissect the mechanisms of resistance.

Line 142: A supplemental table with the markers used in the study would be helpful (SNP flanking sequence, genome position, parental alleles) to allow replication by other researchers. That would also be particularly useful for breeders for marker-assisted selection.

Line 204: 'following' instead of 'flowing'?

Line 232: A supplemental figure illustrating the segregation distortion along the genome for the 4 crosses would support the text well.

Line 251:" a significant lower severity": Considering the confidence interval (25.8+-32.7%) given for WAS208 for severity, are the F3 families with low severity marks really significantly different than WAS208? What threshold has been chosen? How many families are in this case?

Line 271: If severity has been measured on all plants, and not only on symptomatic plants, the interdependence between the two traits is clear.

Line 309: I am not sure about the conclusion about qHBV11.1 controlling "primarily" the RHB severity in FD2000, infection being prior to the expansion of symptoms. This QTL explains also a substantial part of incidence in that cross (R<sup>2</sup>=0.29). It would be interesting to find out why no effect of the QTL on incidence was found in WAS208 and Badka (Detection threshold? Interactions with other QTLs? Level of infection lower in the FD2000 cross?)

Line 393: On the potential role of AGO4 in virus resistance, I suggest to cite also Bhattacharjee et al. ("Virus resistance induced by NB–LRR proteins involves Argonaute4-dependent translational control." The Plant Journal 58.6 (2009): 940-951).

Line 462: I didn't find any mention of the QTLs qHBV9 and qHBV10 in the tables or in the results section, they appear only in the discussion, am I right? Can it be added?

Line 493: One application of this study is the possibility for breeders to increase the level of RHBV resistance by pyramiding the different QTL. That can be mentioned here.

#### Reviewed by Valérie Geffroy, 02 January 2023

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