



## New variants of *AvrPiz-t* identified in *Pyricularia oryzae* from Malaysia

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### ABSTRACT

Rice blast disease (causal agent: *Pyricularia oryzae*) is an important rice disease worldwide as it can cause significant yield loss. Resistance genes in rice can recognise the corresponding avirulence genes (*Avr*-gene) of *P. oryzae* and suppress the attack of the pathogen as proposed in 'gene-for-gene' interaction. This interaction is highly specific. The resistance will be rapidly breakdown owing to the plasticity of *Avr*-gene. The breaking down of disease resistance can cause problem in managing the disease as well as sustaining a resistant rice variety. This paper aimed to study and analyse the sequence variation of *AvrPiz-t* gene from *P. oryzae* isolates in Malaysia (Sarawak) together with *AvrPiz-t* sequences available from National Centre for Biotechnology Information (NCBI). This study compared the *AvrPiz-t* haplotype found with previously reported haplotypes. A total of 20 Malaysian *P. oryzae* isolates were obtained from six divisions of Sarawak and were verified using internal transcribed spacer. Together with seven Malaysian *P. oryzae* isolates from a previous study, a total of 27 *AvrPiz-t* gene sequences from Malaysian isolates were successfully amplified and verified. The *AvrPiz-t* open reading frame (ORF) of 27 Malaysian isolates were aligned with 123 *AvrPiz-t* ORF from other countries that were available in NCBI database and 100 *AvrPiz-t* ORF from a previous study (Total = 250 sequences). Unique sequences were identified and translated into amino sequence. The amino acid sequences were aligned with haplotype sequences from previous studies. In total, there were 19 *AvrPiz-t* haplotypes, with eight newly identified haplotypes. The *AvrPiz-t* haplotypes were distributed across 14 countries with haplotype H9 as the dominant haplotype. Neutrality tests were performed for the 250 *AvrPiz-t* ORF sequences. The 250 isolates of *P. oryzae* were undergoing directional natural selection. Detached leaf inoculation using filter paper method of selected Malaysia (Sarawak) *P. oryzae* isolates carrying one of the new *AvrPiz-t* haplotypes showed different level of pathogenicity towards Sarawak rice landraces. The data obtained serves as baseline information for disease management in Malaysia, especially for Sarawak state. Additionally, the information is also a reference for future rice breeding program in Sarawak.

### Introduction

Rice blast is caused by *Pyricularia oryzae* and known as one of the most destructive rice diseases. Annual yield loss for rice production caused by rice blast can go up to 10 – 30 % globally (Skamnioti and Gurr, 2009; Zhou, 2016). Incorporating resistance (R) gene into susceptible rice cultivars is a way to manage rice blast disease. Following gene-for-gene interaction proposed by Flor (1971), the resistance gene (R-gene) in plant recognises the avirulent gene (*Avr*-gene) in the

pathogen and result in resistance. Instability of *Avr*-genes due to mutation, genetic recombination and sexual mating leads to high diversity of *Avr*-genes and causes rapid resistance breakdown of rice cultivars (Huang et al., 2014; Wang et al., 2020).

To date, there are 40 *Avr* genes identified (Ning et al., 2020) and 12 of them have been cloned and characterised; *Avr-Pita* (Orbach et al., 2000), *Avr-CO39* (Farman and Leong, 1998), *PWL1* (Kang et al., 1995), *PWL2* (Sweigard et al., 1995), *ACE1* (Fudal et al., 2005), *AvrPiz-t* (Li et al., 2009), *AvrPia*, *AvrPii*, *Avr-Pik/km/kp* (Yoshida et al., 2009),

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