

## **Ekspresi Gen CYP6ER1 pada Populasi *Nilaparvata lugens* Stål setelah Paparan Imidakloprid dan Resistensinya Terhadap Nitenpiram, Dinotefuran, dan Fipronil**

### **Abstrak**

Wereng batang coklat (*Nilaparvata lugens*) merupakan salah satu hama pencucuk penghisap penting pada tanaman padi di Indonesia. Gen P450s CYP6ER1 dilaporkan berkontribusi terhadap resistensi imidakloprid pada *N. lugens*. Tujuan penelitian ini untuk mengetahui level ekspresi gen CYP6ER1 pada tahap hidup larva (instar 1, 5), imago jantan dan imago betina, pada dua konsentrasi (350 dan 1.050 ppm) pada empat waktu papar (24, 48, 72, 96 jam setelah perlakuan) imidakloprid. *N. lugens* yang berasal dari populasi Kecamatan Pringsewu, Kabupaten Pringsewu, Provinsi Lampung diseleksi imidakloprid sebanyak dua kali (Psw2) dengan rasio resistensi 3,7 kali dan enam kali (Psw6) sebesar 5,4 kali. Berdasarkan *reverse transcription quantitative PCR* (RT-qPCR) diketahui peningkatan ekspresi gen CYP6ER1 pada Psw2 dan Psw6 instar satu yang dipapar imidakloprid pada instar 3 generasi sebelumnya. Pola ekspresi pada stadia berbeda menunjukkan kebutuhan metabolisme yang berbeda. Peningkatan ekspresi gen CYP6ER1 terjadi secara cepat mulai dari 24 jam pertama dan meningkat hingga 96 jam setelah papar, selaras dengan peningkatan konsentrasi papar. Hal tersebut mengindikasikan bahwa seleksi dengan imidakloprid dapat menyeleksi individu *N. lugens* yang lebih peka, sehingga terjadi peningkatan rasio resistensi dan ekspresi gen CYP6ER1. Informasi terkait pola ekspresi dari gen CYP6ER1 berpotensi digunakan sebagai diagnosa cepat resistensi yang bermanfaat dalam strategi pengelolaan laju resistensi imidakloprid. Sementara itu status resistensi imidakloprid dan ekspresi gen CYP6ER1 pada *N. lugens* di Jawa bervariasi. Pengambilan sampel dilakukan di Jawa Timur (Kab. Malang, Kab. Ngawi, dan Kab. Ponorogo), Jawa Tengah (Kab. Klaten dan Kab. Purworejo) dan Jawa Barat (Kab. Cirebon) pada saat *outbreak* dan tidak *outbreak*. Pengujian menggunakan F1 menghasilkan rasio resistensi (RR) yang beragam 1,0–6,9 kali dibandingkan populasi paling peka. Ekspresi relatif gen CYP6ER1 0,15–6,32 kali, tidak selalu sebanding dengan rasio resistensi imidakloprid. Hal ini mengindikasikan bahwa di lapangan banyak hal yang berpengaruh terhadap perkembangan resistensi imidakloprid. Hasil penelitian ini bermanfaat sebagai upaya pemantauan perkembangan resistensi imidakloprid. Di Indonesia sendiri terbagi menjadi tiga haplotipe, namun tidak spesifik mengelompokkan populasi Jawa dan Sumatra dalam haplotipe yang sama. Populasi Indonesia khususnya Jawa dan Sumatra memiliki keragaman haplotipe (Hd) sebesar 0,573 dan nucleotide diversity ( $\phi$ ) sebesar 0,00174. Hal ini mengindikasikan kemungkinan terjadinya migrasi antara populasi Jawa dan Sumatra, demikian pula populasi resisten antar wilayah. Dua populasi lapangan berasal dari Kabupaten Pringsewu, Provinsi Lampung dan Lendah, Kabupaten Kulon Progo, Provinsi Daerah Istimewa Yogyakarta yang diseleksi dengan imidakloprid sebanyak delapan generasi dan populasi laboratorium sebagai kontrol. Seleksi dengan insektisida imidakloprid menyebabkan pertambahan  $LC_{50}$  secara berkala. Populasi Pringsewu sebelum diseleksi memiliki  $LC_{50}$  361,87 ppm menjadi 1.321,10 ppm setelah delapan kali seleksi. Sementara populasi Lendah sebelum diseleksi memiliki  $LC_{50}$  225,95 ppm menjadi 1.116,94 ppm setelah delapan kali seleksi. Populasi Pringsewu dan Lendah setelah seleksi imidakloprid memiliki *resistance ratio* (RR) sebesar 6,48 dan 5,48 kali dibandingkan dengan populasi laboratorium. Populasi Pringsewu setelah seleksi tidak menunjukkan potensi resistensi silang terhadap nitenpiram (1,35 kali), dinotefuran (0,03 kali) dan fipronil (1,42 kali). Sementara itu populasi Lendah setelah seleksi menunjukkan potensi resistensi silang terhadap nitenpiram (7,76 kali) dan fipronil (4,12 kali) namun tidak terhadap dinotefuran (0,01 kali). Dinotefuran berpotensi digunakan dalam pengendalian *N. lugens* resisten imidakloprid. Penelitian ini penting dalam manajemen resistensi insektisida di Indonesia.

**Kata kunci:** neonikotinoid, resistensi metabolik, resistensi silang, *up-regulation*, wereng

## **CYP6ER1 Gene Expression in Nilaparvata lugens Stål Populations Exposed to Imidacloprid and Their Resistance to Nitenpyram, Dinotefuran, and Fipronil**

### *Abstract*

The brown planthopper (*Nilaparvata lugens*) is one of the most important phloem-feeding pests of rice in Indonesia. The cytochrome P450 gene CYP6ER1 has been implicated in imidacloprid resistance. This study aimed to determine the expression levels of the CYP6ER1 gene at different developmental stages (1<sup>st</sup> and 5<sup>th</sup> instars, male, and female adults) under two imidacloprid concentrations (350 and 1.050 ppm) and four exposure durations (24, 48, 72, and 96 hours post-treatment). *N. lugens* population from Pringsewu subdistrict, Lampung Province, was selected for imidacloprid resistance twice (Psw2; resistance ratio (RR) = 3,7-fold) and six times (Psw6; RR = 5,4-fold). Reverse transcription quantitative PCR (RT-qPCR) analysis showed increased CYP6ER1 expression in Psw2 and Psw6 1<sup>st</sup> instar that were exposed to imidacloprid in the previous 3<sup>rd</sup> instar generation. Expression patterns varied across developmental stages, suggesting differences in metabolic activity. CYP6ER1 expression increased rapidly within the first 24 hours and continued up-regulated to 96 hours, in line with the increasing exposure concentration. These results indicate that imidacloprid selection enhances resistance levels and CYP6ER1 expression. Monitoring CYP6ER1 expression patterns may serve as a valuable diagnostic tool for the early detection of imidacloprid resistance, thereby supporting more effective resistance management strategies. Meanwhile, the status of imidacloprid resistance and CYP6ER1 gene expression in *N. lugens* in Java was varied. Sampling was conducted in East Java (Malang Regency, Ngawi Regency, and Ponorogo Regency), Central Java (Klaten Regency and Purworejo Regency), and West Java (Cirebon Regency) during both outbreak and non-outbreak periods. Bioassay using F1 yielded RR ranging from 1,0 to 6,9-fold compared to the most sensitive population. The relative expression of the CYP6ER1 gene ranged from 0.15 to 6.32 times and was not always proportional to the imidacloprid resistance ratio. This suggests that numerous factors in the field contribute to the development of imidacloprid resistance. The results of this study support the monitoring of resistance and the development of effective strategies to manage and delay imidacloprid resistance in *N. lugens*. In Indonesia, three haplotypes were identified, but the populations from Java and Sumatra were not grouped into the same haplotype. The Indonesian population, particularly from Java and Sumatra, showed a haplotype diversity ( $H_d$ ) of 0,573 and nucleotide diversity ( $\phi$ ) of 0,00174. These findings suggest possible migration between the Java and Sumatra populations, as well as the presence of resistant populations across regions. Two field populations were collected from Pringsewu District, Lampung Province, and Lendah, Kulon Progo District, Special Region of Yogyakarta Province. These populations were selected with imidacloprid over eight generations, while a laboratory population served as the control. Selection with imidacloprid resulted in a gradual increase in  $LC_{50}$  values. The  $LC_{50}$  of the Pringsewu population increased from 361,87 ppm before selection to 1.321,10 ppm after eight generations. The Lendah population had an  $LC_{50}$  of 225.95 ppm before selection, which increased to 1.116,94 ppm after eight generations of selection. After imidacloprid selection, the Pringsewu and Lendah populations showed resistance ratios (RR) of 6,48 and 5,48 times, respectively, compared with the laboratory population. The Pringsewu population did not exhibit cross-resistance to nitenpyram (RR = 1,35-fold), dinotefuran (RR = 0,03-fold), or fipronil (RR = 1,42-fold). In contrast, the Lendah population demonstrated potential cross-resistance to nitenpyram (RR = 7,76-fold) and fipronil (RR = 4,12-fold), but not to dinotefuran (RR = 0,01-fold). These results indicate that dinotefuran has potential for controlling imidacloprid-resistant *N. lugens*. This study offers valuable insights for managing insecticide resistance in Indonesia.

**Keywords:** cross resistance, metabolic resistance, neonicotinoid, planthopper, up-regulation