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THE SEGREGATION PATTERN OF INSECT RESISTANCE GENES IN THE PROGENIES AND CROSSES OF TRANSGENIC ROJOLELE RICE

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ABSTRACT

Successful application of genetic transformation technique, especially in developing rice variety resistant to brown plant hopper and stem borer, will depend on transgene being expressed and the gene inherited in a stable and predictable manner. This study aimed to analyse transgene segregation pattern of the progenies and the crosses of transgenic rice cv. Rojolele harboring *cry*1Ab and *gna* genes. The third generation (T_2) of five transgenic Rojolele events containing *gna* and/or *cry*1Ab were evaluated for two generations to identify the homozygous lines and to study their inheritance. The homozygous lines were selected based on the result of PCR technique. The segregation patterns of *gna* and *cry*1Ab were studied in eight F_2 populations derived from Rojolele x transgenic Rojolele homozygous for *cry*1Ab and or *gna* and their reciprocal crosses. Data resulted from PCR of F_2 population were analysed using a Chi Square test. The study obtained six homozygous lines for *gna*, namely A22-1-32, A22-1-37, C72-1-9, F11-1-48, K21-1-39, K21-1-48, and two homozygous lines for *cry*1Ab, namely K21-1-39 and K21-1-48. Both *cry*1Ab and *gna* transgenes had been inherited through selfing and crossing with their wild type as indicated from the F_1 containing *gna* and *cry*1Ab as many as 48.4% and 47.4%, respectively. In six of the eight crosses, *gna* was inherited in a 3:1 ratio consistent with Mendelian inheritance of a single dominant locus, while in the remaining two crosses, *gna* was segregated in a 1:1 ratio. The presence of *cry*1Ab in F_2 populations also showed a 3:1 segregation ratio in all crosses. In the F_2 population derived from F_1 plant containing *cry*1Ab and *gna*, both transgenes segregated in a 9:3:3:1 dihybrid segregation ratio. This study will add to the diversity of genetic sources for insect resistance and allow further use of these transgenic lines for pyramiding resistance to brown plant hopper and stem borer or separately in rice breeding programs whenever the efficacy tests and biosafety requirements have been completed.

[*Keywords: Oryza sativa*, transgenic plant, genetic resistance, segregation, genetic inheritance]

INTRODUCTION

Brown plant hopper and stem borer are the major insects causing rice production loss in Indonesia. The progress of classical rice breeding for brown plant hopper and stem borer resistance is hampered due to the quick formation of the new biotype of brown plant hopper causing break of resistance even in the elite breeding lines, while for stem borer the major problem is the lack of resistant genes available in the respective species and their wild relatives (Rao and Padhi 1988). The International Rice Research Institute (IRRI) has screened more than 1500 entries for yellow rice stem borer and 6000 entries for stripe rice stem borer resistance (Jackson 1995), resulted some lines which have low to moderate resistance..

Plant genetic engineering is one of the complimentary tools to the conventional breeding for the development of new varieties. This technique has some advantages such as the wider germplasm options, reduction of the number of backcrossing needed to eliminate one of the parental genetic backgrounds and the precision of transferring only the target gene without any other unknown genes introgressed (Conner 1997). A number of transgenic food crops have been released globally using this techniques such as maize, soybean, and rice (www.agbios.com). In this reported work, the parental material harbors *gna* gene encoding for snowdrop (*Galanthus nivalis*) agglutinin and synthetic *cry*1Ab gene encoding crystal protein from *Bacillus thuringiensis* (Sardana *et al.* 1996). Genetic transformation using *gna* gene in tobacco for resistance to aphid was first reported by Hilder *et al.* (1995), followed by the work on rice targeted to brown plant hopper (Rao *et al.* 1998; Sudhakar *et al.* 1998), while *cry* gene has been extensively used in maize for resistance to stem borer (Amstrong *et al.* 1995; Jansens *et al.* 1997) and also in rice (Ghareyazie *et al.* 1997; Breitler *et al.* 2000). Other biotic stress resistance genes introduced to rice were among others the *wasabi defensin* gene for resistance to blast diseases (Kanzaki *et al.* 2002), *potato proteinase inhibitor* II gene (Duan *et al.* 1996), and *CpTi* gene (Xu *et al.* 1996).

Plants have mechanisms for defending themselves from microorganisms and insects. Plant produces

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