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主論文の要旨

Gene mapping of bruchid resistance in moth bean (*Vigna aconitifolia*)

論文題目 (モスビーン (*Vigna aconitifolia*) におけるマメゾウムシ抵抗性の遺伝子マッピング)

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論文内容の要旨

Bruchids are pests that infest stored grain and cause significant seed loss in legume crops. Previous research using an F₂ population (F₂OA) resulting from the cross between wild moth-bean (*Vigna aconitifolia* [Jacq.] Mar'echal) accession TN67 (resistant) and cultivated moth-bean accession ICPMO056 (susceptible) showed that resistance to the azuki bean weevil (*Callosobruchus chinensis* L.) in TN67 was due to a single gene located in the major quantitative trait locus called *qVacBrc2.1*. In this study, we finely mapped *qVacBrc2.1* and identified candidate genes in this locus using both F₂OA and a large F₂ population (F₂NB) derived from the same cross. In contrast to the previous study, we found that resistance to the pest was controlled by two genes in the F₂NB population. Additionally, we added new markers to *qVacBrc2.1* and reanalyzed the QTL in the F₂OA population, which revealed that *qVacBrc2.1* was composed of two linked QTLs, *qVacBrc2.1-A* and *qVacBrc2.1-B*. We verified the presence of *qVacBrc2.1-B* using the F₂NB population. Comparative genomics using three *Vigna* spp. strongly suggested the presence of two tandemly duplicated genes, VacPGIP1 and VacPGIP2, which

encoded polygalacturonase inhibitors (polygalacturonase-inhibiting proteins) and were candidates for conferring resistance. However, only VacPGIP1 was successfully cloned and sequenced. The comparison of VacPGIP1 coding sequences of TN67 and ICPMO056 showed eight single nucleotide polymorphisms, three of which altered the amino-acid sequence of the predicted domains of polygalacturonase inhibitors in ICPMO056. These findings indicate that VacPGIP1 and VacPGIP2 regulated *C. chinensis* resistance in TN67. Furthermore, we used comparative RNA-seq-based transcriptomic analysis to identify candidate genes by performing RNA-seq of immature and semi-mature seeds from both cultivars. The study conducted RNA sequencing on immature and semi-mature seeds of two different cultivars. The results showed that the genes responsible for producing polygalacturonate inhibitors and lectins had varying levels of expression. Both stages showed an increase in expression, with a greater increase observed in the mature stage. This suggests that mature seeds contain a higher concentration of these proteins, which are important for bruchid resistance in moth bean. The findings were confirmed by RT-qPCR analysis. In conclusion, the genes responsible for polygalacturonate inhibitors and lectins are crucial for bruchid resistance in moth bean.