学位論文の要旨(論文の内容の要旨) Summary of the Dissertation (Summary of Dissertation Contents)

論 文 題 目 Dissertation title:

Application of Marker- Assisted Selection for Breeding Drought-Tolerant Rice (Oryza sativa L.) in Vietnam

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Rice, (*Oryza sativa* L.) is one of the most important crops providing staple food for a large segment of the world population Climate change is likely to adversely impact on rice production in Tropical Asia. There are two broad areas of environmental stresses: abiotic factors (salinity, heat, drought, cold, submergence, radiation, and heavy metals) and biotic factors (pathogens and herbivore). Especially environmental stresses including heat, cold, drought, and salinity factors extremely affected on average agriculture yield in the world. Among these indicators, drought is one of the most common stresses, causing a remarkable loss of crops.

To be more specific, drought stress is one of the main constraints to rice production and yield stability in rainfed upland ecology. It is estimated that 70% of the yield losses could be due to water scarcity affecting with approximately 27 million ha of rainfed upland. Vietnam has been predicted to become one of the most vulnerable countries and significantly impacted countries due to the influences of climate change. Farmers in these regions usually utilized rainfall to cultivate rice plants. The drought resistance of plants is one of the most complex biological processes, which involves numerous changes at the physiological, cellular, and molecular levels. The effect of drought on rice plants considerably varies with genotypes, developmental stages, and degree and duration of drought stress. Many genes have been identified to be involved in the response to drought stress in plants. However, as yield is a complex trait, there is a necessity for a genetic and physiological analysis of yield contributing traits under drought stress.

Currently, several methods have been used to develop drought-tolerant crops: conventional breeding, conventional breeding utilizing marker-assisted selection (MAS) and genetic engineering. So far, genetic engineering has been successfully discovered many genes which are involved in the plant responses to drought stress. However, due to irregular rain, rice plants were damaged by drought during seedling, and from flowering to maturity stage. The injuries on leaves at seedling stage, sterile spikelet at reproductive stage and stem shoot under drought stress condition are key traits related to drought tolerance. Therefore, selection of drought-tolerant individuals should not only observe morphological traits but also physiological, biochemical and genotype by environment interaction.

The fact is that, the genetic bases of drought tolerance (DT) and drought avoidance (DA) at reproductive stage in rice were analyzed using a combination of inbred line population from a cross between *indica* lowland and a tropical *japonica* upland cultivar. A genetic linkage map consisting of 245 SSR markers was constructed for mapping QTL for these traits. A total of 27 QTLs were resolved for 7 traits of relative performance of fitness and yield, 36 QTL for 5 root traits under control, and 38 for 7 root traits under drought stress conditions. Only a small portion of QTL for fitness- and yield-related traits overlapped with QTLs for root traits indicate that DT and DA had distinct genetic bases.

It is obvious that, the physiology and molecular biology information of rice stress tolerance is beneficial the biotechnological improvement of rice productivity. Of which, DNA biotechnology is becoming more prevalent by its great contribution in this field. Progress of developing rice varieties for the unfavorable areas has proved that the modern breeding tools can address many problems of farmers. The development of the highyielding drought tolerant rice varieties is considered the most economical mode, which enhance and stabilizes the productivity of drought contaminated areas. Recent advances in plant breeding with the development of agricultural technologies have provided numerous tools for breeders to improve phenotypic screening, ranging from marker assisted selection (MAS) of key traits to molecular breeding (MB) and genetic engineering. To be more specific, markers are used for selecting qualitative as well as quantitative traits. The number of empirical researchers applied broadly those tools in improving tolerant rice cultivars to drought stress in China, India, and Thailand. The important prerequisites for successful selection of the early generation with MAS are the population sizes and heritability levels of the selected traits. From conventional breeding techniques, genetic functional markers could shorten rice varietal development. In other words, this study provided the detailed information on the relative importance of marker-assisted selection of drought tolerance.

In brief, the specific objectives of the study include

- Selection of parental materials for drought tolerance
- Detection of differences in quantitative trait loci (QTL) in their relative effect on components of quantitative drought tolerance
- Development of drought tolerance populations

In the first study, the rice germplasms were screened using a total of 165 SSR markers used for characterization and evaluation of genetic diversity. Among them, 73 SSR markers were found to be polymorphic among the accessions. Maximum PIC (polymorphic information content) values were detected in three markers including RM11125, RM21, and RM5629, which were between 0.78 and 0.79. Cluster analysis of microsatellite markers revealed that by a genetic distance of 0.63, the rice varieties were separated into three clusters. In the integration of drought tolerance, agro-morphological traits, and genetic diversity, four cultivars OM4900, IR78913-B-22-B-B-B, OM6162, and IR75499-73-1-B performed as the most promising parental donors for developing agronomic and drought-tolerant rice.

Secondly, rice germplasms were analyzed both physical and chemical properties. Among 44 rice germplasms, IR79008-B-11-B-B-1 showed overall good physical characteristics (head rice, grain length, grain width, chalkiness), and three varieties (IR75499-73-1-B, OM6162 and OM4900) had good chemical characteristics (amylose content, gel consistency, protein content, gelatinization temperature, and aroma). The important highlight of the study was that progenies of the cross between OM6162/ SwarnaSub1 had a low amylose content, high gel consistency, high protein content, and low chalkiness.

Thirdly, path analysis appears as the best method to evaluate the relationship between yield and relevant traits. Path analysis permits estimation of direct effects of various traits on yield as well as their indirect effects via other components traits. Based on path analysis, traits as a number of filled grains/panicles, the number of filled-grain/panicle, and harvest index had strong and direct positive effect correlation with grain yield. Adaptability of rice to the drought and submergence stresses is the most important objective of the rice breeding program. Additionally, rice yield can be improved with a comprehensive combination of both conventional and molecular breeding techniques. Totally 16 markers were used to screen for parental polymorphism. Polymorphism of parents of 6 pairs of crosses served for backcross. Currently, at least two populations determined the usefulness of this powerful approach to identify associations between traits of interest such asyield potential, drought tolerant characteristics and genetic markers using diverse genotypes (OM6162/swanasub1// OM6162 and OMCS2000/IR75499-73-1-B//OMCS2000).

The final study, development of new lines with tolerance to drought, a multipart trait, is a major challenge and a thorough understanding of the physiological and molecular mechanisms that direct the yield of rice under drought stress condition is necessary. Therefore, the objective of this study was to investigate the effect of drought stress at seedling and reproductive stages in the development of drought tolerance. Totally seven markers (RM219, RM201 RM105, RM23602, RM23877, RM24103 and RM328) were used for an identifying to drought tolerance on chromosome 9 for the BC₂F₂ generation of OMCS2000/IR75499-73-1-B//OMCS2000. New breeding BC-derived lines were screened for drought tolerance using phenotyping and molecular markers. Two advanced breeding lines (BC₂F₂-54 and BC₂F₂-45) were adapted to drought stress by both genotypic analysis. The plant height, number of tillers, and filled grain had the positive correlation with yield/hill under drought stress.

In the current study, the results have been obtained from various experiments, it can be concluded that these markers were earlier identified to be linked to drought resistance and yield traits in rice through conventional QTL mapping efforts. This research also successfully introduced both (OM6162/swanasub1//OM6162) and (OMCS2000/IR75499-73-1-B//OMCS2000) populations. This study focused initially on finding the right combination of lines to give high yields under drought stress. Two lines $(BC_2F_2-45 \text{ and } BC_2F_2-54)$ provide urgent objective for breeders released as new cultivars in providing higher incomes to the Vietnam farmers.

備考

論文の要旨はA4判用紙を使用し、4,000字以内とする。ただし、英文の場合は1,500語以内と する。

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