GENOMICS-ASSISTED ALLELE MINING AND ITS INTEGRATION TO BREEDING IN RICE

Masahiro Yano, Yusaku Uga, Shuichi Fukuoka, Kaoru Ebana, Jun-ichi Yonemaru, Toshio Yamamoto
AgroGenomics Research Center, National Institute of Agrobiological Sciences, Tsukuba, Japan

ABSTRACT
Since whole genome sequence of rice has been decoded, it has been more than 6 years. Rice sequence information has contributed on innovation of selection strategy in rice breeding (marker assisted selection) as well as functional analysis of rice genes. Elucidation of the association between nucleotide and phenotypic changes is inevitable to this end and has been a big challenge in molecular genetics and breeding of rice. Toward this goal, we have been involved in the genetic dissection of natural phenotypic variations in rice and have identified several genes involved in complex traits, including heading date, pre-harvest sprouting, root morphology, disease resistance and eating quality. To enhance the power of genetic dissection of complex phenotypes, we are developing several mapping populations, such as recombinant inbred lines and chromosome segment substitution lines, which will allow us to extract the useful alleles from natural variants. Recently, QTL for durable resistance to rice blast has been cloned from Japanese upland rice. This finding has opened new opportunity to introduction of the unique blast resistance gene without a linkage drag of low eating quality. We have also detected a major QTL for deeper rooting on chromosome 9. Based on the MAS, near-isogenic lines of IR64 with deeper root has been developed and is now under evaluation of its potential of drought avoidance. To facilitate allele mining using novel plant materials, we have also embarked on the genome-wide discovery of single nucleotide polymorphisms (SNPs). These SNP explore new strategy of genetic and breeding study. We have already revealed genomic sequences of several cultivars to detect SNP and definition of haplotype block of Japanese rice cultivars and high-yielding cultivars for forage use. Currently association between these particular haplotypes and phenotypic performance is investigated. These progresses have gradually acted as a force to change a landscape of selection in rice breeding.

KEYWORDS
Single nucleotide polymorphism (SNP), marker assisted selection, quantitative trait locus (QTL), mapping population

REFERENCES
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Genes identified in Japanese research activities

Genes identified in Japanese research activities

Genes contribute on improvement of rice cultivars

Natural variations in rice

Topics
➢ Discovery of useful allele and its introduction to elite cultivars (root morphology)
➢ Plant materials development for the allele mining
➢ Establishment of a platform of genome wide SNP typing for genetic analysis and marker assisted selection

Drought avoidance using root traits QTL in rice

Global warming and spreading desertification are causing serious drought damage in rice-growing areas in many developing countries that rely solely on rain to grow rice. Therefore, breeding of drought-resistant lowland rice is becoming an important research topic.

➢ A deeper and thicker root system is an important component associated with avoiding drought stress in rice.
➢ QTL cloning and its utilization may be useful to improve drought avoidance in rice.
Phenotypic difference between IR64 and Kinandang Patong

IR64
Lowland indica
High yield
Shallow root

Kinandang Patong (KP)
Upland
Tropical japonica
Deep root

Evaluation system of deeper rooting

Phenotypic difference between IR64 and Kinandang Patong

Uga et al. JXB (2011)

Ratio of deeper roots (%)

IR64
KP
Hetero

Uga et al. JXB (2011)

Positional cloning of Dro1 (Deeper rooting 1)

QTL mapping in RIL

LoD = 26.9
R² = 66.6%
AE = 16.1%

KP allele at the Dro1 increase deep roots

Ratio of deeper rooting (%)

IR64
Dro1-NIL

KP allele at the Dro1 increase deep roots

Y. Uga. unpublished data

Survey of diversity in root morphology in rice

Trench method is an appropriate way to directly measure root distribution in the field.

Dro1 can reach in deep soil layer without root elongation

Y. Uga. unpublished data
Ongoing research activities on evaluation of performance of drought avoidance by Dro1

The International Center for Tropical Agriculture (CIAT) in Colombia

Dro1-NIL has been tested under different levels of drought stresses using experimental field with rain-out shelter.

The International Rice Research Institute (IRRI) in Philippines

Dro1-NIL will be tested under lowland condition and multiple locations in IRRI’s drought breeding network. Dro1 will be introduced into other mega varieties to study the expression of drought tolerance in different genetic backgrounds.

Plant materials and their utility in genetic mapping

<table>
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<tr>
<th>Primary mapping populations</th>
<th>Potential application</th>
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<tr>
<td><strong>F1</strong></td>
<td><strong>Detection of major QTLs</strong></td>
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<tr>
<td><strong>BC1F1</strong></td>
<td><strong>Validation of genetic interaction</strong></td>
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<tr>
<td><strong>BC2F1</strong></td>
<td><strong>Determination of minor QTLs</strong></td>
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<tr>
<td><strong>BC3F1</strong></td>
<td><strong>Detection of genetic interaction</strong></td>
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<td><strong>BC4F1</strong></td>
<td><strong>Saturation mapping</strong></td>
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<td><strong>BC5F1</strong></td>
<td><strong>Gene pyramiding</strong></td>
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<td><strong>BC6F1</strong></td>
<td><strong>Fine mapping</strong></td>
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<tr>
<td><strong>BC7F1</strong></td>
<td><strong>Introgression, elimination of linkage drag</strong></td>
</tr>
<tr>
<td><strong>CSSL</strong></td>
<td><strong>Advanced mapping populations</strong></td>
</tr>
<tr>
<td><strong>NIL</strong></td>
<td><strong>RIL</strong></td>
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</tbody>
</table>

Mapping populations available for genetic research

Distributed from Rice Genome Resource Center (RGRC) at NEAS
http://www.rgrc.dna.affrc.go.jp/
Re-sequencing of rice diverse accessions by NGS

<table>
<thead>
<tr>
<th>Accession</th>
<th>Seq(Gb)</th>
<th>Seq. depth</th>
<th>Method</th>
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<tbody>
<tr>
<td>Kasalath</td>
<td>10.4 x 26.9</td>
<td>single &amp; pair end</td>
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<td>Kao Mac Kho</td>
<td>31.8 x 85.1</td>
<td>single &amp; pair end</td>
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<td>Naba</td>
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<td>single &amp; pair end</td>
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<td>Type 331-5</td>
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<td>Khao Nam Jen</td>
<td>4.2 x 19.4</td>
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<td>Moha</td>
<td>6.9 x 23.8</td>
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<tr>
<td>Beih Khe</td>
<td>10.4 x 26.0</td>
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<td>Xu Zee Zhong</td>
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</table>

Asian accessions

Accession | Seq(Gb) | Seq. depth | Method          |
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<td>コシヒカリ</td>
<td>5.8 x 15.6</td>
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<tr>
<td>陸羽</td>
<td>1.8 x 4.8</td>
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</tr>
<tr>
<td>栄光</td>
<td>6.1 x 16.2</td>
<td>single end</td>
<td></td>
</tr>
<tr>
<td>嘉平</td>
<td>6.1 x 16.2</td>
<td>single end</td>
<td></td>
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<tr>
<td>小白谷</td>
<td>16.8 x 40.0</td>
<td>pair end</td>
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</table>

Temp. and Trop. Japonica

For genetically diverse accessions

3334 SNPs

For Japanese cultivars

2551 SNPs

768 SNPs

Development of platform of SNP analysis in genetic analysis and marker assisted breeding

SNP typing array

Identification and utilization of natural variations

Mapping populations (F2, RILs etc.) using a wide range of natural variants

Phenotyping: different methods and conditions

Successive backcrossing

CSSL of interests

Nearly isogenic lines

Selection marker

QTL pyramiding

Contributors


Funding

Ministry of Agriculture, Forestry & Fisheries (MAFF), Japan