



Improving techniques for assessing rice grain quality

John Oliver¹, Arun Aryan² & Russell Reinke³

¹NSW DPI, Wagga Wagga Agricultural Institute, ²Charles Sturt University, Wagga Wagga, ³NSW DPI, Yanco Agricultural Institute

in a rice hull

- The Rice Grain Quality Project has investigated and developed methods for making the screening of grain quality characteristics of lines in the rice breeding program more precise and efficient
- The project has also carried out research to understand the genetic characteristics that are linked to the cooking quality of different types of rice

The Rice Grain Quality Project has continued to evaluate the quality parameters of rice breeding lines and to improve screening techniques for more precision and efficiency. A greater emphasis is being placed on the use of molecular markers to assist selection.

The background work in improving screening techniques has proceeded on a number of fronts over the past year, and has also provided the opportunity for students to be involved in research projects.

Measurement for amylose type

Amylose is a component of starch and an important determinant of rice cooking quality. Unlike amylopectin, the other component of starch, which is controlled by several genes, amylose synthesis is mainly controlled by a single gene (*Waxy* gene).

The genetic code of any organism is made up of four bases (nucleotides) commonly abbreviated as A, T, C or G. In the upstream regulatory part of the *Waxy* gene, the genetic code contains a sequence of C and T bases in succession. The length of this repeat sequence varies between varieties and has been reported to be associated with the amylose content and hence the cooking quality of a rice line. More importantly, analysis of this CT repeat number is a good way to track the pedigree of a crossbred. The NSW DPI rice improvement program at Yanco measures and categorises all crossbreds according to the length of the CT repeat.

Measurement of CT repeats is a relatively straight forward molecular analysis, but somewhat expensive. Through the course of this project, Dr Arun Aryan was able to modify the methodology of the test so that less expensive materials could be used, the result being that the modified test is easier and potentially cheaper to conduct than the existing method.

Validation & implementation of the new method

The *Waxy* gene exists in rice in one of two forms (or alleles) designated Wx^a or Wx^b . Firm-cooking *indica* rices are the Wx^a form, while soft-cooking *japonica* rices are the Wx^b form.

The difference between the two forms is due to a difference of one base (or nucleotide) in the genetic code. At one part of the *Waxy* gene, a G base is altered to a T base, affecting the expression of *Waxy* gene, and in turn the amount of amylose synthesised in the rice.

Current methodology requires two separate analyses to distinguish this variation in the *Waxy* gene. Dr Aryan developed a method to test for either the G or the T variation in a single analysis.

Effect of starch components on cooking

Ms Annabel Boret, a French university student, spent six months in the Yanco cereal laboratory as part of her requirement for industrial experience. Her project was to understand how cooking properties are affected by starch components of rice grain and which combinations of different forms of starch enzyme genes are responsible for specific cooking quality traits.

In the initial phase, Annabel analysed different rice lines for their genetic make-up and later studied their cooking properties, to draw a correlation between the two aspects.

A range of cultivars (18) of diverse origin and different cooking properties was grown under controlled conditions of light and temperature, until maturity. The genetic analysis was conducted on leaf samples from which DNA was extracted and the type of alleles/genes present in these lines were identified. The key alleles looked at were the *Waxy* allele (Wx^a or Wx^b), the starch branching enzyme gene *SBE1*, and the CT repeat sequence in the *Waxy* gene.



Physicochemical analysis required harvesting of mature grain samples and measurements of a pasting curve including gelatinisation temperature, texture and grain elongation after cooking. These cooking results were then correlated with the genetic analysis.

From the analyses of results, Annabel established the following simple correlations between the genetic make-up and the cooking properties of these elite lines.

- Waxy gene with Wx^a allele gives positive setback on RVA (an analysis of viscosity)
- Waxy gene with Wx^b allele gives negative setback on RVA
- All the *japonica* type rice lines studied contained an insertion element (*Tourist Os6*) in the regulatory region of their *SBE1* gene, while *indica* lines did not. However, no clear correlation could be established between the cooking properties of these rice lines and their *SBE1* gene types (alleles).

Starch branching enzyme in Australian rice


This work was conducted by Ms Gayle Warnock, CSU student, as her BSc summer scholarship research project undertaken in the Yanco laboratories.

A recent report indicated that the promoter region of *SBE1* gene in *japonica* rice harbours an insertion element or transposon (*Tourist Os6*) that is absent in the *SBE1* gene of *indica* rice.

Transposon insertions are known to modulate gene expression, however no such expression studies have been reported for the *SBE1* alleles of rice. The aim of Gayle's project was to check the distribution of two *SBE1* alleles in Australian rice cultivars.

Australian cultivars and their parental lines from the germplasm collection at Yanco were grown to seedling stage and DNA was extracted. The presence of the *indica* or *japonica* type *SBE1* allele was detected and analysis indicated that most Australian rice cultivars have the *japonica* type *SBE1* allele.

Of interest is that some of the parental lines harbour the *indica* type *SBE1* allele, indicating that somehow crossbreds with *japonica* type *SBE1* allele were preferentially selected by breeders. On this basis, cultivar 'Baru' warrants further investigation. One of the key Australian varieties 'Doongara', contained the *indica* type *SBE1* allele, which could be linked to its unique Basmati type (firm-cooking) starch properties.

Genetic profiling of parental material in this manner, assists the breeders in selecting parents with desired attributes for further variety development. 

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John Oliver

Research Leader, Cereal Genetics & Improvement

NSW DPI, Wagga Wagga Agricultural Institute

T: 02 6938 1816

E: john.oliver@dpi.nsw.gov.au