



Perfect molecular markers for fragrance & gelatinisation temperature

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IN A RICE HULL

- ▶ Molecular markers have been identified that will help Australian rice breeders identify 'difficult to measure' traits, early in the rice breeding program
- ▶ Markers for fragrance and gelatinisation temperature have been identified, meaning that quick DNA analysis methods on tissue from young rice plants can replace labour intensive, time consuming analysis on harvested rice grain

Molecular markers are pieces of DNA which flag the presence, or absence, of both desirable and undesirable traits. Plant breeders can use markers to identify traits which are difficult to measure by physical assessment.

The marker can be near the gene which determines a particular trait, or within the gene itself. If the marker is within the gene it is said to be perfect because it can never separate from the gene during the course of plant breeding.

This project has been investigating a number of molecular markers for traits that are important to the Australian rice industry.

Markers for fragrance

Consumers are willing to pay a premium price for fragrant rice, which provides an incentive to develop fragrant rice varieties suited to the environmental conditions of the Australian rice growing areas. Although the flavour and fragrance of both Basmati and Jasmine style rice is associated with high levels of a chemical (2-acetyl-1-pyrroline or 2AP for short) which we can all smell when it is cooked, it is not an easy task to work out in the field if any one rice plant is fragrant or not - especially in the early stages of the breeding program when there is a mixture of fragrant and non-fragrant plants in the field.

Tasting individual grains was one of the original methods for the selection of fragrant rice varieties within the Australian breeding program and it is still the principal means of identifying fragrance in many breeding programs worldwide. However, objective evaluation of fragrance using this method is labour intensive since a panel of analysts is required due to the variability in the ability of individuals to

detect fragrance. In addition to this, the evaluation is difficult, unreliable and has real limitations when processing large numbers of samples because for any one analyst the ability to distinguish between fragrant and non-fragrant samples diminishes with each successive analysis as the senses become saturated and abrasions to the tongue can occur from chewing the hard grain.

Other evaluation methods which involve smelling leaf tissue or grains after heating in water or reacting with chemical solutions are available but these can cause damage to the nasal passages of the analysts.

An objective method of 2AP measurement has been developed but it is time consuming and requires large tissue samples meaning it cannot be used in the very early stages of the breeding program.

The genetic cause of fragrance in Jasmine and Basmati style rice has been recently identified and is due to a deletion in a gene which codes for an enzyme that is involved in the manufacture of 2AP. Non-fragrant rice varieties have what appears to be a fully functional copy of this gene while fragrant varieties have a copy of the gene which has the deletion and this probably disables the enzyme. The disabled enzyme creates a blockage in the system and this ultimately results in the synthesis of more 2AP in fragrant rice than non-fragrant rice.

Knowing the genetic cause of fragrance has allowed the construction of a marker that predicts with 100% accuracy the fragrance status of all cultivars and individual plants. Rice breeders can now take a sample from a seedling, long before it produces seed, and work out if it will be fragrant using the most simple and inexpensive laboratory tools (Figure 1). Because we can now distinguish between fragrant




and non-fragrant rice early in the breeding program, lines that will not be useful can be discarded early in the program, making it easier and quicker for the rice breeders to produce fragrant rice varieties.

Markers for gelatinisation temperature

Rice starch is the energy source for many millions of people around the world. Starch is a polymer of glucose which is another way of saying a long chain of glucose molecules joined together. Starch presents as a mixture of two forms, amylose which is principally a linear polymer and amylopectin which is a more complex mixture of both linear and branched chains of glucose. Rice starch has a semi-crystalline structure which is disrupted by cooking, transforming the starch into a softer edible gel like material. Because it is associated with the cooking time and texture of cooked rice and cool cooked rice, the temperature at which rice starch forms this softer edible gel like material, or gelatinises, is an important component of rice eating quality.

Starch production is undertaken by several enzymes; enzymes that activate the glucose so the chain can be formed, others that extend the chains of amylose and amylopectin, others which place branches in the amylopectin and still others which break these branches in what is believed to be a necessary part of the process of remodelling the growing amylopectin, allowing it to crystallise. A group of researchers working in Japan found

that the gene that has the greatest impact on rice starch gelatinisation temperature is one that is needed for the extension of the linear chains of amylopectin.

Although it was known which gene affected gelatinisation temperature (GT), it was not known precisely which part of the gene was important or what versions of the gene resulted in low GT starch or high GT starch. The Japanese group went some way to solving this puzzle but there was a piece missing. The last piece of the puzzle was put in place when we identified the key differences in the gene which determines if rice starch has a high gelatinisation temperature or a low gelatinisation temperature. Knowing the difference, we can now predict which of these two discrete gelatinisation temperature classes, which differ by 8°C, any one rice plant or variety belongs too. As with fragrance, knowing the genetic cause of gelatinisation temperature differences has allowed the construction of a marker that predicts the gelatinisation temperature status of all cultivars and individual plants, which will in turn allow the rice breeders to distinguish between high and low gelatinisation temperature rice more easily, early in the breeding program. 

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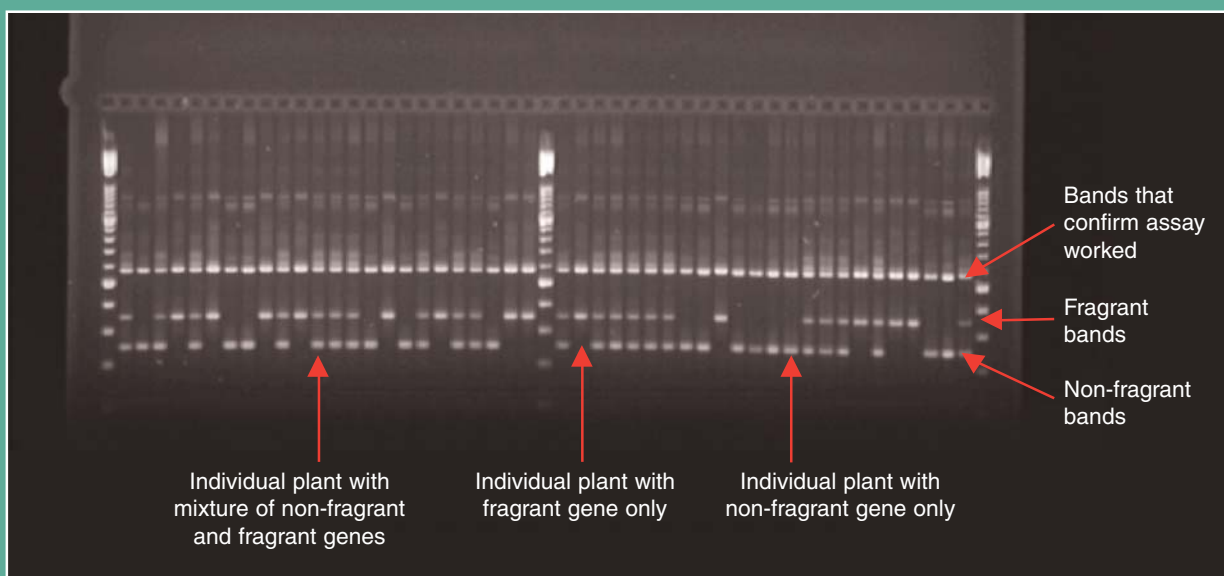


Figure 1 Photograph of DNA from the offspring of a cross of non-fragrant and fragrant rice. The different banding patterns distinguish between plants that have just the fragrant version of the gene, just the non-fragrant version of the gene or a mixture of the fragrant and non-fragrant versions of the gene.