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Genomics & Biotechnology



Genomics now involve combinations of different yet complementary methods, and information obtained on one species can be used for other species (research is thus being mutually enriched and efforts are gradually converging). The method implemented in the Genmap project combined genetic mapping with a field analysis of the *Hevea* family studied (screening for QTLs associated with agronomic traits measured in field trials in Thailand).

The Genmap project included the following successive phases:

- **Performing a 'model' cross** (year 2000) by manually pollinating the two RRIM600 and PB217 parents, i.e. two cultivated clones that represent two different metabolic types. The aim was to obtain a broad range of variability in 334 progeny of this cross in order to fulfil the scientific objectives, and also to find clones combining complementary qualities in this progeny.

- **Genotyping 334 progeny** for 267 available and relevant microsatellite markers. For each progeny, the two alleles occurring at the same locus were identified (PCR and electrophoresis).

- **Genetic mapping** of the cross (*cf. Genmap genetic map*) via software processing of genotyped progeny data (*JoinMap3* software), focused on the joint or separate presence of alleles of different markers. Loci corresponding to the different markers were thus found to be located on each of the 18 *Hevea* chromosomes, with the distances between two markers corresponding to the crossover percentages noted between them.

- **Field analysis** (from 2002 to 2010) of the main agronomic traits of 196 progeny, especially growth and latex production traits, biochemical traits associated with metabolic types of production and rubber quality traits.

- **Establishment, by software processing**, of progeny classes associated with each allele per marker, calculation of the values of the agronomic traits of these classes, and identification of loci that determine most of the variation in certain traits (QTLs).

The Agropolis advanced research platform supported phases 2 and 3 of this project. The *Hevea* genetic mapping took 17 months of research time. The use of microsatellite markers represented substantial progress relative to the past use of RFLP markers in terms of research time (PCR efficiency), number of genotyped individuals, distribution of mapped loci on the genome and the polymorphism of assessed alleles.

Molecular genetic markers and genetic mapping

► *Kanlaya Prapan preparing a polyacrylamide gel to separate PCR-derived microsatellite markers*



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So-called 'molecular' genetic markers directly concern the DNA molecule in which they correspond to noncoding zones (loci). They are said to be genetically 'neutral' because they do not influence the evolution of the species, and identification of their alleles in an individual does not depend on the tissue studied or on the medium conditions. The so-called linkage disequilibrium genetic mapping technique has developed considerably since a high number of these markers have become available. This technique involves investigating the extent of character recombination in progeny due to crossover (intra-chromosomal rearrangements that occur during meiosis). Because of these rearrangements, alleles from two loci that were initially present on the same chromosome have a certain degree of probability of being separated in two different gametes (i.e. when a crossover occurs in the space between them).

There are DNA zones (called linkats) where very little crossover occurs and where the same combinations of alleles for neighbouring loci are preserved. Genetic mapping involves analysing the frequency of association of respective alleles from two loci, for all available marker pairs. The markers can thus be located in relative positions to each other, while identifying the linkats and detecting 'super-linkats' corresponding to different chromosomes from the studied species (*Hevea brasiliensis* is a diploid species with 18 chromosome pairs). Each distance between two markers is expressed as a crossover percentage (1 centimorgan = probability of 1% crossover during meiosis), and genetic maps are drawn up on this basis.

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