

For a resistant planting material Hevea-Microcyclus ulei interactions

he Rubber Programme is carrying out research in Brazil and French Guiana on the genetic control of *M. ulei*. The main purpose of this work, which is being carried out in partnership with the Michelin estates in Brazil, is the characterization and genetic selection of durable resistance to this parasite, which is responsible for South American leaf blight (SALB) on *Hevea*.

To that end, two main lines of research are being investigated. The first aims to acquire as clear an understanding as possible of *M. ulei* diversity, as an initial evaluation of its ability to overcome new resistances. The second seeks to exploit known sources of resistance within a programme of variety creation and genetic marking of genes involved in such resistance.

Pathogenicity diversity

The study of diversity in the pathogencity of *M. ulei* involved a sample of 88 strains from the Brazilian states of Bahia and Mato Grosso, and from the coastal strip of French Guiana. The virulence and aggressiveness of the strains were assessed under controlled conditions on a range of differential hosts comprising seven *H. brasiliensis* and *H. benthamiana* clones.

Figure 1 summarizes all the results obtained. Of the 88 strains studied, 73 distinct genotypes were observed and classed into 5 groups and 2 sub-groups. This classification did not correspond to the geographical origin of the strains. The table specifies the main characteristics of these groups. The considerable susceptibility of clone FX 3864, which is not very discriminant in the sample tested, can be seen. Group 1 is characterized by strains whose

virulence seems to be directed against *H. benthamiana*, according to the differential range used. Group 3 stands out from the others as the only one containing strains that were virulent on FX 985. Groups 4 and 5 did do not have any strong specificity and group 2 shows low pathogenicity on the range of hosts in the study.

Work prior to this study revealed the existence of several races of *M. ulei*, from 4 to 10 depending on the authors. Our work, which was conducted on a larger scale, revealed the considerable diversity of the pathogen, 73/88 in the sample tested. No geographical specialization of the parasite was detected, but it is reasonable to suspect a certain degree of parasite specialization depending on the species, *H. brasiliensis* or *H. benthamiana*.

These results pinpoint the nature of the *Hevea-Microcyclus ulei* pathosystem: the substantial diversity observed largely explains the speed with which the parasite has overcome resistances set up against it to date. The current work is intended to confirm that result. The sexuality of *M. ulei* is probably the main driving force behind this diversity.

Variety creation and resistance marking

Work on the creation of Hevea clones resistant to M. ulei has made considerable progress in recent years. The definition of a clear breeding strategy along three lines with short, medium and long-term objectives enabled a start to be made to the work described below.

A large collection of original clones is available at the Michelin estate in Bahia. These clones are all derived from crosses

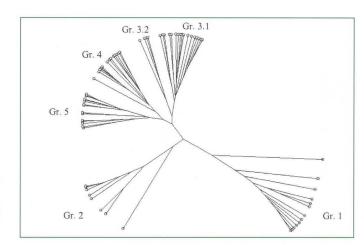


Figure 1.
Hierarchical classification
of the pathogenicity
of a sample of 88 strains
of M. ulei on 7 H. brasiliensis
and H. benthamiana clones

Strains		Clones						
Group	Number	FX 3864	FX 985	FX 2261	FX 2804	IAN 3087	FX 3899	MDF 180
		H. bras.	H. bras.	H. bras.	H. benth.	H. benth.	H. benth.	H. bras.
Group 1	15	+/-			+	+	+	+/-
Group 2	20	+/-				-	-	-
Group 3.1	17	+	+	+/-		+	+/-	+
Group 3.2	5	+	+	-		-	+	+/-
Group 4	15	+	-	+/-			-	+
Group 5	16	+		+	+/-	+	+/-	+

with material from the Madre de Dios region, in the Peruvian Amazon, on which few studies have been carried to date, but which appears to be highly promising. Following a large-scale mass selection phase involving more than 800 clones, the best of them in terms of resistance to M. ulei and rubber production, are currently undergoing a large-scale test phase. In the short term, these tests should produce material that can be used to replant zones severely infested by M. ulei.

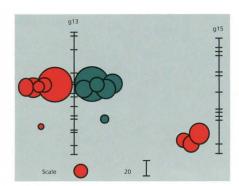
A programme of variety creation and selection was launched in 1993 at the Edouard Michelin estate, in Mato Grosso, Brazil. By recombining parents carrying high partial resistance and parents with a high production potential, we are seeking to assemble within the same varieties characteristics that are favourable for their cultivation in Latin America. The choice of genotypes produced under this programme was based on family selection involving quantitative genetics. Until now, selected resistant clones had only undergone mass selection. It is thus hoped to gain greater precision in the choice of best genotypes, and reduce assessment costs. The first clonal trials, planted since 1998, reveal a good correlation between the family values of resistance to *M. ulei* and those obtained in the previous phase. At that time, the trials involved seedlings, which did not offer the possibility of genotype replication. The soundness of the method used has been confirmed. It is thus possible to envisage supplying planting material in the medium term that is both high-yielding and resistant.

New prospects for the creation of *Hevea* varieties resistant to M. ulei have been opened up with the major work carried out on genetic mapping of Hevea. It culminated in the defence of a thesis in 1999. This genetic mapping work was carried out on progeny PB 260 x RO 38, whose RO 38 parent has a genetic background of resistance to M. ulei through its H. benthamiana origin. After creating a saturated consensus map using more than 700 markers—mostly AFLP and RFLP, but also isozymes and microsatellites—, a search for QTL markers of resistance was launched. Inoculation of the fungus under controlled conditions on 200 individuals of the progeny revealed 6 regions of the genome involved to varying degrees in such resistance. One of these regions, located on chromosome g13, seemed to be particularly interesting since it explained a large proportion of phenotypic variance, from 16 to 36% depending on the traits considered and the strains inoculated (figure 2). In addition, it seemed to be independent from the type of strain, since the 5 strains tested revealed a QTL of resistance in this region. This result was confirmed under natural infestation conditions: observations carried out in French Guiana on the same progeny revealed the presence of a major resistance gene in the same region of chromosome g13, for a trait that had not been studied under controlled conditions. All the resistance QTLs detected came from the H. benthamiana grandparent.

Conclusions and prospects

These results with genetic marking of major genes of resistance to *M. ulei* are promising, since they suggest that it will be possible to use marker assisted selection in the future, aimed at resistance to *M. ulei*. Three operations need to be carried out beforehand.

- New sources of resistance will have to be identified within the species *H. brasiliensis*, which do not have the drawbacks of RO 38 for agronomic and production traits. Some parents of Madre de Dios origin are very good candidates for this.
- Further mapping work will be carried out, and QTLs of resistance will be located on progenies derived from these parents. This phase should be simplified by the experience already acquired on PB 260 x RO 38.
- A variety creation strategy will be implemented based on early screening in



the nursery of individuals combining the largest number of favourable alleles at loci marking major genes of resistance. Those individuals will then be integrated into a conventional breeding scheme and assessed according to agronomic and production criteria.

Figure 2. Synthetic map indicating the location of the main QTLs for sporulation intensity (in red) and lesion diameter (in green) on chromosomes g13 and g15. The vertical dotted lines represent the 5 strains of *M. ulei* used under controlled conditions. (From the thesis by D. Lespinasse, 1999)

List of publications —

Lespinasse d., 1999. Cartographie génétique de l'hévéa (*Hevea spp.*) et déterminisme de la résistance au champignon pathogène *Microcyclus ulei*. Thesis, Université des Sciences et Techniques du Languedoc, Montpellier, France, 120 p.

Lespinasse D., Grivet L., Troispoux V., Rodier-Goud M., Pinard F., Seguin M., 2000. Identification of QTLs involved in the resistance to South American leaf blight (*Microcyclus ulei*) in the rubber tree. Theor. Appl. Genetics 100 (6): 975-984.

LESPINASSE D., RODIER-GOUD M., GRIVET L., LECONTE A., LEGNATÉ H., SEGUIN M., 2000. A saturated genetic linkage map of rubber tree (*Hevea spp.*) based on RFLP, AFLP, microsatellite, and isozyme markers. Theor. Appl. Genetics 100 (1): 127-138.