# Mapping QTLs for Resistance to Root Rots Caused by *Phytophthora tropicalis* in Cassava

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## Introduction

Several species of *Phytophthora* (Loke 2004) attack cassava (*Manihot esculenta* Crantz), causing severe root rot and wilting (Figure 1). The development of *Phytophthora* is favored by inadequate agronomic practices, transport of material from affected areas to those free of pathogens, and by planting in compact or very clayey soils. Varietal resistance is a major tool for managing root rot in cassava. We are now selecting for resistance to *Phytophthora* spp. under greenhouse conditions, inoculating shoots and roots with species that were previously identified by sequencing the internal transcribed spacer region in rDNA.

**Figure 1.** Wilting and root rot caused by *Phytophthora* spp., microorganisms that are classified as water molds, not fungi.

Molecular techniques are increasingly being used to decipher the genetic base of complex agronomic traits. Genetic improvement for disease resistance can also be achieved more quickly and effectively by using molecular markers. In this study, the resistance of parents and progeny of cassava K families (M Nga 2 × CM 2177-2) and CM 9582 (M Bra 1045 × M CR 81) to *Phytophthora tropicalis* was evaluated.

## Materials and Methods

**Plant materials.** We harvested and evaluated 1-year-old roots belonging to the cassava K family (92 individuals, years 2000 and 2001, Santander de Quilichao, Cauca, Colombia) and CM 9582 (43 genotypes, year 2001, Florida, Valle, Colombia) in 2000 and 2001. One resistant (M Bra 1045) and one susceptible (M Col 2066) variety to root rot were also included as checks.

**The pathogen.** As inoculum, isolate 44, identified as *P. tropicalis*, was used. This isolate was obtained from cassava infected with root rot in Barcelona (Quindío, Colombia). The inoculum was cultured in medium prepared with oat agar, antibiotics, and fungicides. Incubation was carried out at 20°C to 26°C for 7 days (Loke 2004).

**Inoculation.** Within an isolation chamber, roots were perforated and inoculated with a fragment of the pathogen. Each genotype was also inoculated with a negative control, that is, culture medium with no *P. tropicalis.* Once inoculated, the roots were deposited in plastic bags containing moisture, and left at 22°C in darkness for 6 days.

**Evaluation and data analysis.** From each root, the height and width of both the wound and the entire cross section were measured.

These data were recorded and processed through *Excel*'s calculation program. The experimental unit was the root. At least six roots roots per genotype were inoculated.

**QTL analysis.** The female-derived cassava map (K family) was based on the segregation of female alleles, corresponding to 192 markers that comprised RFLP, random amplified polymorphic DNA (RAPD), isoenzymes, microsatellites, expressed sequence tags (ESTs) and known genes (Fregene, unpublished). To minimize the detection of false positives, a significant association between a DNA marker and *Phytophthora* resistance was declared if the probability was more than 0.005. The degree of phenotypic variance explained by each marker was obtained from the regression coefficient ($r^2$). All data were analyzed with Q-gene software on McIntosh.

## Results and Discussion

Genotypes of the cassava K family, evaluated during 2000 and 2001, showed infected areas covering between 22% and 80% of roots (Figure 2). The distribution of frequencies of root area affected by *P. tropicalis* corresponded to a normal distribution.

Some genotypes that had, in 2000, intermediate resistance to *P. tropicalis* tended to become susceptible in 2001 and vice versa (Figure 3). The correlation between the evaluations of 2000 and 2001 was -0.15.

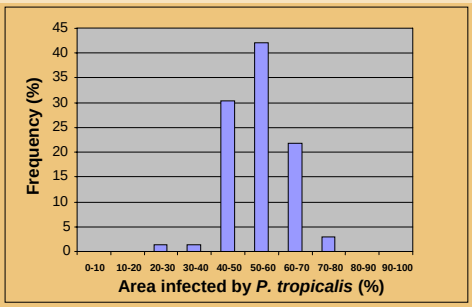


**Figure 2.** Distribution of frequencies of genotypes from the cassava K family according to the percentage of area infected by root-rot pathogen *Phytophthora tropicalis*.

Variability in the expression of resistance between years indicates that the environment affects the phenotypic expression, generating variation and affecting QTL expression. Variability in resistance across years may also indicate the K family to be polygenic.

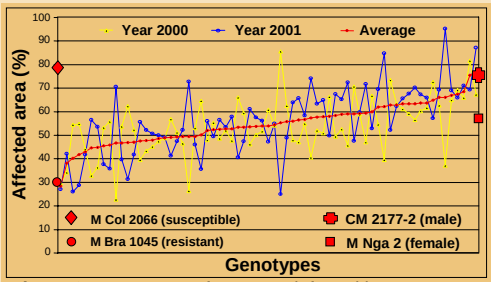


**Figure 3.** Percentage of root area infected by *Phytophthora tropicalis* across genotypes of the cassava K family for years 2000 and 2001.

Genotypes of the cassava family CM 9582 (M Bra 1045 × M CR 81) showed 70% to 90% of areas infected. The distribution of frequency of CM 9582 genotypes for area infected by *P. tropicalis* presented a rising curve. As shown in previous studies, M Bra 1045 is tolerant of *P. tropicalis*. The genetic base of M Bra 1045 can be assumed to be polygenic, and to have epistasis in this cross.

**Table 1.** QTLs explaining the highest values of phenotypic variance for resistance to Phytophthora root rot in cassava, according to the percentage of root area affected.

| Linkage group[a] | Markers | F[b] | V[c] |
|---|---|---|---|
| C (3) | RGY172 | 0.029 | 5.4 |
| H (8) | SSRY178 | 0.315 | 1.3 |
| J (10) | CDY76 | 0.163 | 4.0 |
| | K2a | 0.040 | 8.6 |
| N (14) | SSRY13 | 0.078 | 4.2 |
| Q (17) | SSRY911 | 0.047 | 5.7 |
| V (22) | NS911 | 0.007 | 9.0 |
| | GY153 | 0.049 | 4.5 |

[a] Female map
[b] *F* of statistical analysis
[c] $r^2$

Table 1 shows the results of the single-marker regression analysis of percentage of infected area in roots inoculated in the laboratory. Eight QTLs were defined by analyzing 92 individuals of the cassava K family, two of which explained 9.0% and 8.6% of phenotypic variance. The eight QTLs are located on linkage groups C, H, J, N, Q, and V of the female-derived framework map.

## Conclusions

Results show that resistance to *Phytophthora* root rot is polygenic in the cassava K family. The occurrence of individuals more resistant than the two parents and the detection of QTLs associated with molecular markers from the female-derived map show that resistance alleles coming from both parents contribute to resistance in the progenies (transgressive segregation). Such characteristics are well known in heterozygous species and are useful for combining resistance genetic factors in the same cultivar (Jorge et al. 2001).

Although the populations differed in their genetic base of resistance to *Phytophthora*, the levels of resistance observed were not sufficiently high to warrant use in genetic improvement programs. Hence, identifying new parents and developing new populations are desirable.

## References

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