

# Sustainable cocoa Phytophthora resistance

## Integrated genetic, genomic and marker assisted selection approaches to provide a sustainable T. cacao resistance towards Phytophthora species

### ABSTRACT

The objectives of this project were to carry out an integrated genetic and genomic approach in order to improve our knowledge on the genetic and molecular bases of cocoa resistance to Phytophthora, a cocoa disease responsible of important yield losses in all producing countries. For this purpose, several studies were conducted:

A first synthesis of all already existing genetic studies related to cocoa resistance to Phytophthora was first carried out. This meta-analysis has highlighted the polygenic nature of this quantitative resistance and the existence of numerous sources of resistance. Complementary analyses (QTL analyses) were conducted on progenies from Côte d'Ivoire and allowed to identify other QTLs due to different progenitors.

A search for candidate genes was first made from cDNA libraries constructed from T. cacao/Phytophthora interactions and corresponding EST. Genetic markers were defined in these EST and mapped. Cocoa genes, homologous to defense or resistance genes already known in other species could be mapped thanks to 45 EST\_SNP and 6 EST\_SSR.

During this project we have initiated the whole cocoa genome sequencing. An assembly of 76% of the total genome was realised, gathering most of the cocoa expressed genes (98%), and with 82% of them anchored in the 10 cocoa Chromosomes. The immediate availability of this sequence has allowed to analyse in detail 3 gene families known to be potentially involved in defense and resistance mechanisms in other species : genes orthologous to NBS (297 genes), LRR LRK (253 genes) and NPR (4 genes) could be identified and localised in the cocoa sequence. Links between the genome sequence and the genetic map were established, and several candidate genes have been identified in the QTL regions.

Functional genomic studies were simultaneously carried out. A microarray was constituted from EST homologous to genes known as involved in resistance mechanisms and highlighted 193 genes differentially expressed between infected and non infected cocoa tissues, including 48 protein kinases, 35 transcription factors, 7 cell wall biosynthesis genes, 80 PR (pathogenesis related protein), and 23 other receptors. A complementary approach based on complete sequencing of transcriptomes, made by Illumina RNAseq, was initiated at the end of the project and must complete the first results obtained with microarrays.

These results will support the analyses of candidate genes located in the QTL regions and will trace the metabolic pathways used by T. cacao to install its resistance to Phytophthora.

First attempts of marker assisted selection (MAS), based on the accumulation of favorable alleles at the level of 3 QTLs have demonstrated the efficiency of MAS to increase the resistance level of genotypes inside a same large progeny.

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**Project leader :** Claire Lanaud

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## PERSPECTIVES

This project has provided numerous molecular resources, publically available, and original results which have opened new fields of research, and which will continue to be valorised. We can mention particularly : the candidate gene validation, the deciphering of metabolic pathways involved in the phytophthora resistance mechanism using gene network approaches, and the marker assisted selection based on a high marker density, along all the genome, to predict and improve more efficiently the resistance level of new cocoa varieties.

Liste des publications liées au projet RTRA :

Publications dans des revues de rang A :

Mathilde Allegret†, Xavier Argout†, Michel Boccara, Olivier Fouet, Yolande Roguet, Aurelie Berard, Jean Marc Thevenin, Aurelie Chauveau, Ronan Rivallan, Didier Clement, Brigitte Courtois, Karina Gramacho, Anne Boland-Auge, Mathias Tahi, Pathmanathan Umaharan, Dominique Brunel, and Claire Lanaud. 2011. Discovery and mapping of a new expressed sequence tag-single nucleotide polymorphism and simple sequence repeat panel for large-scale genetic studies and breeding of *Theobroma cacao* L. *Dna Research* pp. 1-13, (2011). doi:10.1093/dnares/dsr039

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Communications à des congrès

## Communications orales à des congrès

Xavier Argout et al. 2011. Deciphering the genome structure and paleohistory of the cocoa genome). Oral. Com. Plant and Animal Genome congress. San Diego (USA) 14-19 jan. 2011.

Claire Lanaud. 2010. Advances in cocoa molecular genetics and prospects. Proceedings of the 16th International Cocoa Research Conference, Bali 2009. ( ISBN 978-065-959-5). pp. 105- 117.

Akaza Moroh Joseph, N'GoranJeanne Andi KOHI, Sangare Abdourahaman, Fouet Olivier, Rivalan Ronan, Billot Claire, Lanaud Claire. 2010. QTL analysis in cocoa progenies located in Côte d'Ivoire for some traits of interest using SSR markers. Proceedings of the 16th International Cocoa Research Conference, Bali 2009 ( ISBN 978-065-959-5). P195-201

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