Global transcriptome analyses of cassava-*Xam* interaction using a cassava cDNA microarray

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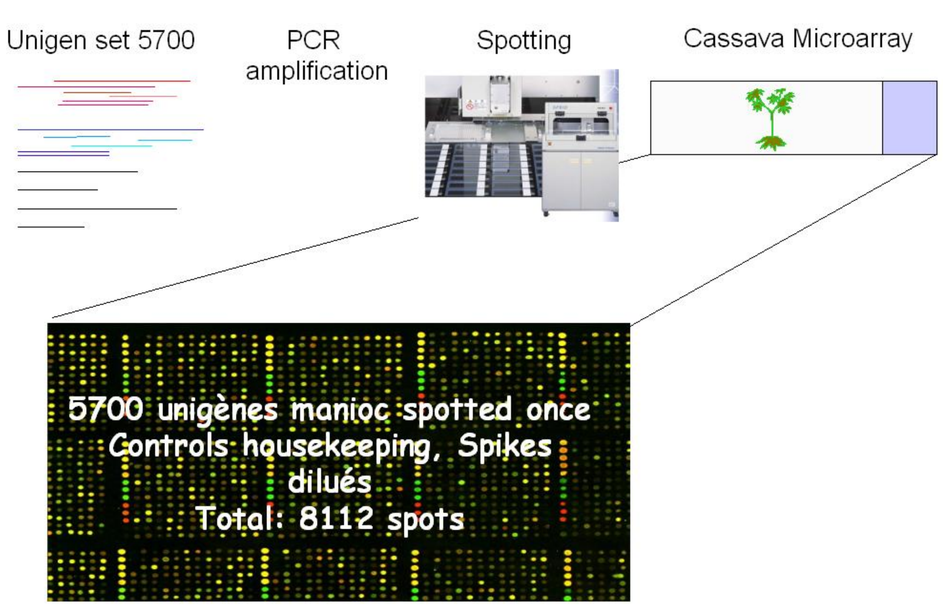
INTRODUCTION

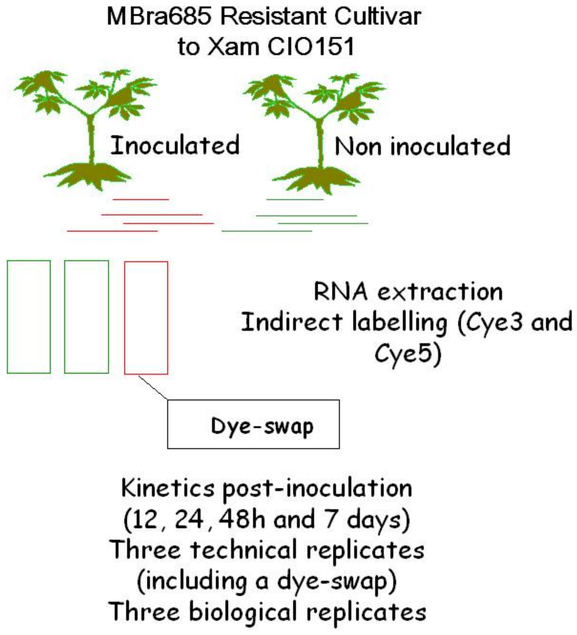
Cassava is a staple crop for millions of people in the tropics. The application of molecular genetic analysis for cassava breeding has been limited compared to others crops. Recently progress have been made in the development of genomic and bioinformatics tools to increase our knowledge of cassava genome structure and cassava gene function. A large cassava EST database has been developed in our laboratory. This represents an important contribution to the genomic resource and permits the beginning of a large-scale analysis of expression profiling in cassava. Microarray analysis is a very informative tool to study the responses of hundreds or thousands of genes simultaneously providing also novel insights into the study of plant-pathogen interactions. A cassava cDNA microarray was constructed and used to study the cassava- *Xanthomonas axonopodis* pv. *manihotis (Xam)* interaction. For the microarray construction, 5700 clones from the cassava unigen set were amplified by PCR and printed once on glass slides. Microarray hybridization was performed using cDNA from cassava plants (resistant variety MBRA685) collected at 12, 24, 48 hours and 7 days post-infection as treatment and cDNA from healthy plants as control. Functional genomic tools such as the cassava microarray give a first comprehensive overview of the molecular basis of defense response to the bacterial blight pathogen and will help in the future in understanding the defense mechanisms to other important pests and diseases.

MATERIALS AND METHODS

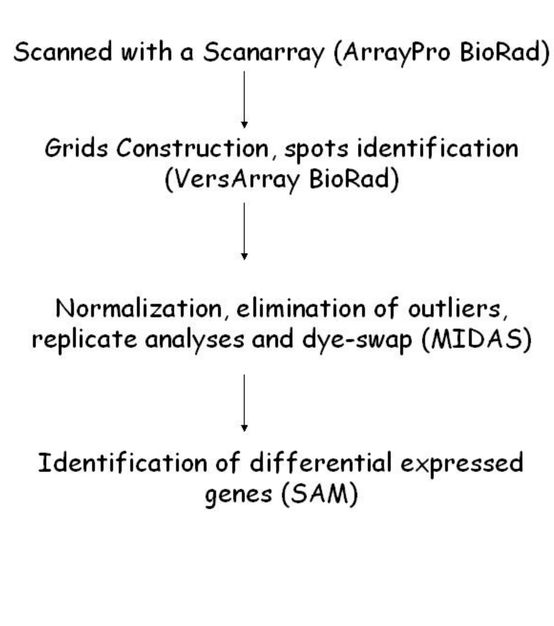
Cassava microarray construction



Experimental design

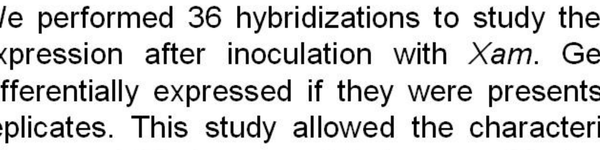


Analyses



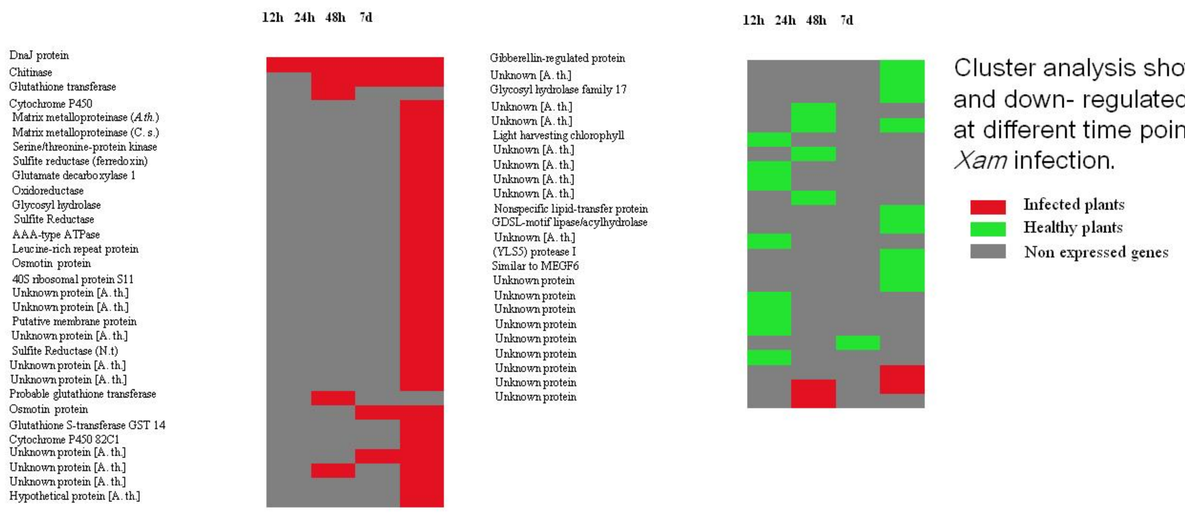
RESULTS

Three technical replicates were conducted, including a dye-swap (Fig 1), and three biological replicates.

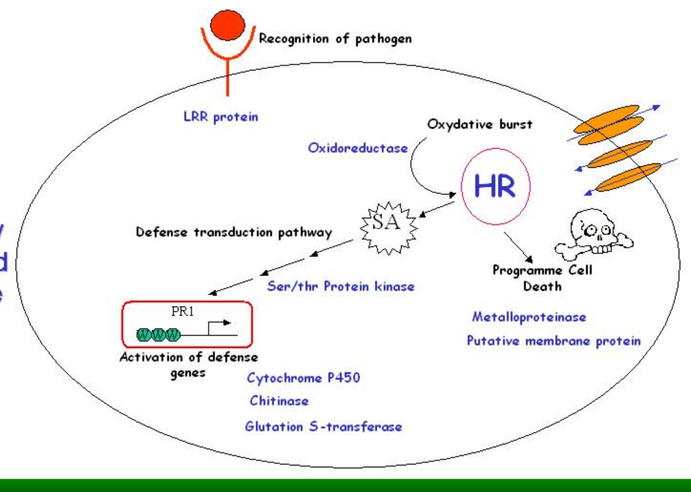


Overlay images showing differential expression in a dye-swap experiment

- We performed 36 hybridizations to study the kinetics of cassava gene expression after inoculation with *Xam*. Genes were considered as differentially expressed if they were presents in at least two biological replicates. This study allowed the characterization of 45 up-regulated genes and 23 down-regulated genes. Most of these were expressed 7 days after inoculation.
- Among those we identified genes involved in the oxidative burst as oxygenase, genes showing similarity to metalloproteinase and membrane protein that are involved in the senescence, necrosis, programmed cell death processes and hypersentive response (HR).
- A ser/threonine kinase and a LRR protein were also differentially expressed, these proteins can be involved in the transduction pathway signal.



Putative function of the cassava genes identified in the microarray analyses in the host plant cell and related to host defense response



CONCLUSIONS AND PERSPECTIVES

• A first generation of cassava microarray was generated containing 5700 unigenes, and was used for a large scale expression analysis of cassava-*Xam* interaction. This microarray represent a valuable tool for conducting further gene expression studies on other traits such as starch content or resistance to other pests and diseases.

• Several genes putatively involved in cassava defense mechanisms were identified and their expression will be confirmed using a QRT-PCR approach.