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Identification of pre- and post-infection genes potentially implied in quantitative banana response to crown rot disease

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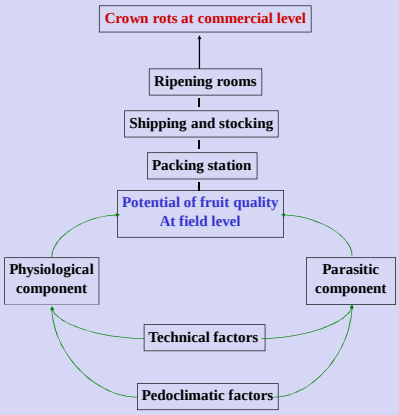
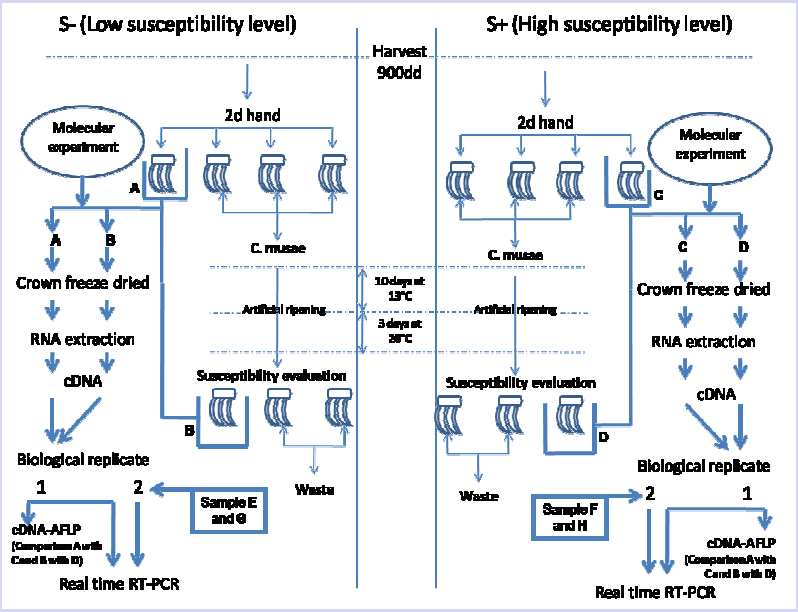
INTRODUCTION



Crown rot, caused by a parasitic complex, is one of the main post-harvest diseases affecting bananas. *Colletotrichum musae* is often regarded as one of the most prevalent fungi contributing to the crown rot complex. Geographical and seasonal variations in symptom expression have been observed. It has been suggested that this phenomenon results partially from variations at the level of fruit quality potential. This potential is controlled by a physiological component, but also by a parasitic component, which appears as the level of banana contamination. The physiological component of the fruit quality potential determines fruit susceptibility, i.e. the response of the fruit to inoculum pressure. The biological responses of the fruit, including disease susceptibility are controlled and regulated by gene expression. One way to understanding the reactions involved in variation of banana susceptibility to the disease is to study the expression of genes involved in these processes. The aim of this study was to identifiy the pre- and post-infection genes potentially implied in fruit to the disease susceptibility variation by cDNA-AFLP approach.

MATERIAL AND METHOD



For each biological replicate, 2 banana trees (Musa sp [AAA group, Cavendish subgroup] cv Grand Nain) with high different susceptibility level were identified (S^+ and S^-). Only the second hands of the bunch were collected and each one was separated into 4 clusters of 4 fingers. For each susceptibility level, one of the four clusters served to collected crown tissues at the harvest stage, 1 hour before the inoculation (1hbi) of *C. musae* at 10^3 conidia/ml. The 3 others served to evaluate a posteriori (13days after inoculation (13dpi)) the susceptibility of the banana to crown rot disease cause by *C. musae*. The crown of one of these 3 clusters was recuperated to serve as sample for molecular analyses at this time. Crowns collected were immediately frozen in liquid nitrogen, freeze dried at -80 C for 24 h and storing it at room temperature before RNA extraction and reverse transcription. The cDNA-AFLP was applied on A to D cDNA pools corresponding at the first biological replicate. cDNA population were compared two by two (S^+ and S^-) in accord with their collected time: 1hbi or 13dpi. The results obtained by cDNA-AFLP were confirmed by real time RT-PCR with 2 independent biological replicate. The susceptibility level was determined by measuring the internal necrotic surface (mm^2) after artificial inoculation.

RESULTS AND DISCUSSION

TDF's	Annotation	Isolation Stage	Regulation level in S-	
			1hbi	13dpi
48B.1	Putative protein kinase	1hbi	+2	+4
44B.2	Dual specificity phosphatase family protein	1hbi	-4	+4
31.1	Zinc finger (C3HC4 RING) family protein	1hbi	-2	+3
33.2	Dopamine β-monooxygenase	1hbi	+1	+4
44.1	Hypothetical protein	1hbi	-4	-4
47.1	Hypothetical protein	1hbi	-1	+4
294.2	Cellulose synthase	13dpi	-2	-2
283.1	Putative glycolipid transfer protein	13dpi	-1	+3
232.2	Serine carboxypeptidase	13dpi	+2	+3
317.1	Putative ubiquitin carboxyl terminal hydrolase	13dpi	+2	-1
145.2	Putative ribose-5-phosphate isomerase	13dpi	-2	-4
284.1	Putative lactoylglutathione lyase	13dpi	-1	-3
190.2	CCR4 associated factor 1-related protein	13dpi	-1	-2

Regulation level obtained by rela time RT-PCR. Mean of the 2 independent biological replicates: 1 = <2; 2 = 2-5; 3 = 5-10; 4 = >10

Some genes appear to be involved in signal transduction and others in the development of specific defense mechanisms. However, the precise function of these proteins has yet to be studied more closely.

Du gène à la fonction: groupe de contact FNRS, Biologie végétale , Gembloux 2 avril 2009