Expression profiling of the metabolism of ripening/spoilage in harvested tomato fruits

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The aim of our study is to identify (new) genes related to (new) metabolic pathways or regulatory pathways that affect fruit spoilage, nutritional quality and marketability. We are studying the mechanism of fruit spoilage derived from Botrytis cinerea (Botrytis) mould in tomato berry fruit. Botrytis is a plant pathogenic fungus that infects over 200 different plant species and provokes significant crop losses, particularly after harvest. During all stages of infection it produces a spectrum of cell wall degrading enzymes, among which are several polygalacturonases (PGs). Infection also results in volatile compounds release from the tomato berry fruit; ethylene, ethanol, short chain aldehydes (hexenals) and carbon dioxide, that may be useful markers to monitor the physiological status of the infected fruits, as early signals of disease progression.

We are currently using a RT-PCR approach to monitor fruit (tomato) and fungus (Botrytis) gene expression related to the monitored volatile compounds in fruit in standard and infected conditions (ACS and ACO: Ethylene biosynthesis), the biosynthetic pathway of potential natural pesticides (LoxA/B, HPL and AOS for hexenals and Jasmonic acid), and to monitor Pathogen Response to infection (PR1 gene and Jasmonic acid). Preliminary analysis in infected tomato fruits (wild type plants) already provided the expression profile of those gene pools during disease progression. Together with the Botrytis-specific primer set (BoPG1 and BeActin), these molecular tools will be used to describe the gene expression profile in tomato fruit upon fungal infection and spoilage in tomato. Moreover, our analysis based on the monitoring of specific metabolic compounds related to fruit ripening/spoilage/pathogenesis will indicate “key” time-point of physiological changes of the fruit. Those key points will be evaluated for cDNA library construction. Such libraries will be used to study differential gene expression by expression profiling using the DNA-microarray technology. The expression profiling will enable the analysis of thousand of genes, thus enabling the isolation of cDNA clones whose expression is related to spoilage/pathogenesis, maybe unravelling new metabolic pathways or differential regulation of described ones.

Together, our approaches will provide a picture of Botrytis growth in vivo on infected tomato fruits. Several storage parameters such as temperature and humidity, as well as the developmental stage of the harvested fruit, affects its quality

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and determine successful Botrytis infection and therefore fruit spoilage. So far, the traditional solutions to these problems have been storage under controlled or modified atmospheres and the use of synthetic pesticides. The understanding of the mechanism of fungal penetration and of the plant response to the pathogen attack will enable us to identify the components of the natural defence response in plants which can be used not only as early and sensitive indicators for spoilage, but which can also be optimised in order to enhance resistance.