

## **PHYLLOSTICTA MUSARUM INDUCED DEFENCE RESPONSES IN BANANA CV. EMBUL AND THEIR PRACTICAL IMPLICATIONS**

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*Phyllosticta musarum*, the causal organism of freckle disease, has been shown to induce several defence responses in the immature banana fruit cv. Embul, including accumulation of phytoalexins, pathogenesis-related proteins and cell wall fortifications in the fruit peel. Furthermore, it has been shown that *P. musarum*-infected bananas are resistant to anthracnose disease caused by *Colletotrichum musae*. *Phyllosticta musarum* infection leads to limited necrosis in the peel that does not expand into rotting during ripening. *Colletotrichum musae* on the other hand remains quiescent and causes complete tissue maceration during fruit ripening. It has been documented that the defence responses induced by *C. musae* are very much weaker than those of *P. musarum*. The objectives of the present study were to further investigate *P. musarum*-elicited defence responses and to build a model that describes various host pathogen interactions.

*Phyllosticta musarum*-infected and *C. musae*-inoculated fruit peel was subjected to a gel diffusion assay to assess chitinase activity. In order to determine the molecular weight of chitinase, extracts were electrophoresed in a SDS slab gel of 12% acrylamide, containing glycol chitin. Subsequent to electrophoresis the gel was re-natured and stained in order to observe chitinase activity bands.  $\beta$ -1, 3-glucanase activity of *P. musarum*-infected or healthy fruit peel was determined by a colorimetric method. Cell wall derived elicitors of *P. musarum* and *C. musae* were extracted and purified using gel filtration column chromatography.

*P. musarum*-infected fruit peel showed a significantly higher chitinase activity ( $p=0.05$ ) compared to healthy or *C. musae* inoculated fruit peel. The major chitinase band was at a low molecular weight region 18,000 Daltons. *Phyllosticta musarum*-infected fruit peel was found to contain a remarkably higher  $\beta$ -1, 3-glucanase activity ( $p=0.05$ ) compared to the uninfected fruit peel. Partial purification of elicitor extracts revealed that *P. musarum* elicitor consisted mainly of polysaccharides while that of *C. musae* contained polysaccharides/glycoproteins or both.

Based on the above results a model was built to explain the host-pathogen interactions. Initially the pathogen-derived elicitors; particularly *P. musarum* elicitor consisting of carbohydrates elicit defence responses in the host. These include hypersensitive reactions at the infection site and structural reinforcements of surrounding cells that prevent further colonization of the pathogens. The accumulation of phytoalexins and induction of chitinases and  $\beta$ -1, 3-glucanases further prevent the growth of invading pathogens such as *C. musae*. These lytic enzymes can digest the invading fungal hyphae and as a result chitin and glucan monomers may get released. These may in turn be able to act as further elicitation of phytoalexins and other defence related enzymes. All these defences in *P. musarum* infected fruit peel may in combination act against the development of *C. musae*. These defence responses have also been induced by treatment of fruit with defence inducers such as Bion® (Acibenzolar-S-methyl ester) which enhanced fruit resistance to anthracnose disease. *Financial assistance by ACIAR is acknowledged.*