# Presence of phytoplasma infections in tomato plants in Mauritius

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#### **Abstract**

Phytoplasmas were detected and identified in some tomato cultivation areas in Mauritius. Symptoms most frequently observed were abnormal shoot proliferation, stunting, reduced leaf and fruit size and shortened internodes. In field-grown tomatoes the incidence of abnormalities rarely exceeded 10%, but under hydroponics up to 100% incidence has been recorded. Two different phytoplasmas were identified by PCR/RFLP analyses. Field-grown tomatoes were infected with a phytoplasma belonging to ribosomal subgroup 16SrI-C and the hydroponically-grown tomatoes were infected with a phytoplasma belonging to ribosomal group 16SrV. Further studies need to be done in order to determine their occurrence, incidence, characterization, host range and mode of transmission, so that eventually the most effective method to control phytoplasma diseases will be determined.

Key words: Phytoplasmas, tomato, PCR/RFLP, plant disease.

#### Introduction

Over the past years in Mauritius, there have been several occasions when tomato growers have reported abnormalities on tomato (*Lycopersicum esculentum*), grown in the open field or under hydroponics conditions across Mauritius. Symptoms most frequently observed were abnormal shoot proliferation, stunting, reduced leaf and fruit size and shortened internodes. In field-grown tomatoes the incidence of abnormalities rarely exceeded 10%, but under hydroponics up to 100% incidence has been recorded.

To date, there has been only one official report of the presence of phytoplasmas on tomato in Mauritius (Dookun *et al.*, 1999). The investigators detected the presence of phytoplasmas from tomato plants using polymerase chain reaction (PCR) with universal primers that amplify the 16S rDNA sequences. Before this first report, tomato plants exhibiting stunted growth or bunchy top symptoms were attributed to abiotic factors or phytotoxicity when other common pathogenic organisms (viruses, bacteria, fungi) could not be detected from the symptomatic leaf samples.

# Materials and methods

Fresh samples of tomato showing phytoplasma-like symptoms were employed for nucleic acid extraction to verify phytoplasma presence. Material was from different kinds of tomato cultivation and from different varieties growing in Mauritius collected over several years. Total nucleic acids were extracted from a 1 g mixture of leaf midribs and phloem tissues from tomato fruits (Prince *et al.*, 1993), dissolved in Tris-EDTA pH 8 buffer, and maintained at 4 °C. Twenty ng/μl of nucleic acid were used for amplification.

After direct PCR with primer pair P1/P7, nested PCR with R16F2n/R2 primers (Gundersen and Lee, 1996) or F1/B6 (Duduk *et al.*, 2004) was performed. Further

nested PCR were performed with R16(I)F1/R1 and R16(V)F1/R1 (Lee *et al.*, 1995) primer pairs. PCR and nested PCR reactions were carried out following published protocol (Schaff *et al.*, 1992). Identification of detected phytoplasmas was done using RFLP analyses on amplified ribosomal DNA fragments with *TruI*, *RsaI*, *HhaI*, *Tsp509I*, *TaqI*, *AluI* (Fermentas, MBI, Vilnius, Lithuania) restriction enzymes. Polyacrylamide 5% gels stained with ethidium bromide were employed to compare profiles to reference phytoplasmas (Bertaccini *et al.*, 2000).

## Results and discussion

In year 2000 and 2003, the presence of aster yellows phytoplasmas belonging to the clover phyllody ribosomal subgroup (16SrI-C) were identified from tomato plants of variety Sirius, grown in the open field after nested PCR and RFLP analyses. Symptoms observed under field conditions were bushy growth, stunting, leaf curling and purple colouration of the leaves (figure 1).



**Figure 1.** Bushiness and purple colouration on symptomatic tomato shoot.

(In colour at www.bulletinofinsectology.org).

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In 2004 and 2005, phytoplasmas were detected from tomato plants of variety Efrat, grown under hydroponics conditions. Identification by RFLP analyses following nested PCR assays, identified a 16SrV ribosomal group phytoplasma. In the latter case, 100% infection was observed and this resulted in heavy loss to the planter.

From the above findings, it is clear that phytoplasma infections are gaining importance in tomato plantations in Mauritius. It is also clear that phytoplasmas infecting tomatoes in Mauritius are not those typically detected in solanaceous crops in other parts of the world. Therefore, further studies need to be done, in order to determine their occurrence, incidence, their characterization, host range and mode of transmission so that eventually the most effective method to control phytoplasma diseases in tomatoes under Mauritius conditions will be determined.

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