

Detection of *Xanthomonas campestris* pv. *campestris* in Crucifer Seeds by a PCR-based Method

Frida Kleitman,¹ N. Ajtkhozina,⁴ A. Dzaimurzina,⁵ Rivka Hadas,² Tanya Gefen,² I. Barash³ and Shulamit Manulis^{1,*}

¹Dept. of Plant Pathology and ²Official Seed Testing Laboratory, ARO, The Volcani Center, Bet Dagan 50250, Israel [*e-mail: shulam@volcani.agri.gov.il]; ³Dept. of Plant Sciences, Tel-Aviv University, Tel Aviv 69978, Israel; ⁴National Academy of Sciences, Institute of Microbiology and ⁵Kazakh Agricultural Institute, Almaty, Kazakhstan

Black rot caused by *Xanthomonas campestris* pv. *campestris* (*Xcc*) is a serious disease of crucifers which occurs worldwide and causes economic losses in Kazakhstan and Israel. The bacterium is seedborne, and seeds are considered the most important source of primary inoculum. A joint project between Israeli and Kazak scientists was aimed at developing a specific and sensitive PCR-based method for detection of *Xcc* in cabbage and other crucifer seeds. A collection of *Xcc* strains isolated from infected crucifer plants or seeds from Israel and different regions of Kazakhstan was established. Isolations were made on four semi-selective media and the isolates were subjected to biochemical and pathogenicity tests on cabbage seedlings. Random amplified polymorphic DNA (RAPD) analysis was carried out with DNA isolated from 106 strains using 77 different arbitrary primers. Two primers were found to reveal diagnostic polymorphisms between *Xcc* and other *X. campestris* strains. An amplified fragment of 0.46 kb, obtained with one of the primers, was cloned, sequenced and provided a source for generation of two specific primers. PCR carried out with these primers amplified the same fragment only with DNA of *Xcc* and not with other strains of *X. campestris* or with non-pathogenic strains. No homology to known genes could be found in the GenBank. Recently, the whole genome of *Xcc* was sequenced and the cloned sequence was found to be homologous to a hypothetical protein from *Xcc*. The sensitivity threshold of the PCR procedure, determined by adding different concentrations of *Xcc* to an extract of cabbage seeds, was between 2 and 20 bacterial cells in the reaction mixture. The threshold level of the detection procedure was determined with artificially inoculated cabbage seeds. The minimum threshold of infested seeds that can be detected by Bio-PCR is 6–7 cfu ml⁻¹ or 90–110 cfu per 1000 seeds, and by direct PCR it is 9–10 cfu per 1000 seeds. (*P*)

Seedborne Diseases in Imported and Domestic Potato Seed Tubers (1999–2002)

Leah Tsrer (Lahkim)*, Orly Erlich, M. Aharon, Marina Hazanovsky and Sara Lebiush-Mordechai

Dept. of Plant Pathology, ARO, Gilat Experiment Station, M.P. Negev 85280, Israel [*e-mail: tsrer@volcani.agri.gov.il]

Monitoring of seedborne diseases in potato seed tubers continued during 1999–2002. Brown rot caused by *Ralstonia solanacearum* was not observed in any of the imported lots. Common scab was detected in most of the imported lots; 63% of the imported lots were contaminated at moderate and high levels, whereas only 5% of the domestic seed lots were contaminated at these levels. Black scurf was detected in most of the imported lots; on average, 44%, 38% and 1% of the lots were contaminated at low, moderate and high levels, respectively, and 18% were disease-free. In contrast, most of the domestic lots were either disease-free (69%) or had a low disease incidence (13%). Only 16% and 1% of the lots were moderately or highly contaminated, respectively. Silver scurf was observed in most of the imported lots during all years of the survey, with no difference between the producing countries. On average, 10%, 48% and 35% of the lots were contaminated at low, moderate and high levels, respectively. Half of the domestic lots were disease-free, and an average of 11%, 36%