Genomic-based development of a real-time PCR protocol for improving the diagnosis of *Xanthomonas arboricola* pv. *pruni*, the causal agent of bacterial spot disease of *Prunus* spp.

Jerson Garita-Cambroner[1,2], Ana Palacio-Bielsa[3] and **Jaime Cubero**[1]

[1] *Instituto Nacional de Investigación y Tecnología Agraria y Alimentaria (INIA), Madrid, Spain. cubero@inia.es*

[2] *Instituto Tecnológico Agrario de Castilla y León (ITACyL), Valladolid, Spain*

[3] *Centro de Investigación y Tecnología Agroalimentaria de Aragón/Instituto Agroalimentario de Aragón, IA2 (CITA-Universidad de Zaragoza), Zaragoza, Spain*

Keywords: *Xap*, bacterial spot of stone fruits, quarantine

Comparative genomics of *Xanthomonas arboricola* species has unveiled a detailed view of the instraspecific evolutionary history of this plant-pathogen as well as a characterization of its pathogenic arsenal associated with the disease process. In addition to these analyses, phenotypic and pathogenic assays have revealed the presence of non-pathogenic strains which are phylogenetically different to those pathogenic strains of the pathovars *pruni*. Comparative analyses among these two groups showed up a wide range of genomic regions that could be useful to differentiate these two intraspecific groups that cohabit on *Prunus*. Taking advantage of the variation in this genomic feature, a real-time PCR protocol, based on a partial sequences of the *xopE3* gene, has been developed to differentiate *Prunus*-pathogenic and non-pathogenic strains of *X. arboricola* and to refine the diagnosis of this quarantine pathogen in the EU. The use of this new protocol in conjunction with a previous protocol based in the amplification of the gen *ftsX* showed a high specificity to differentiate pathovar *pruni* from the other instraspecific groups of *X. arboricola* as well as a high sensitivity (100 cfu/ml or 100 pg/µl of DNA) and efficiency (1.8-2.0, being 2.0 a 100% of efficiency). This new protocol is a valuable molecular tool for the improvement of the diagnosis of the causal agent of bacterial spot of stone fruits and almond.