Agdia Launches Rapid Molecular Diagnostic for Detection of Emerging Pathogen Causing Zebra Chip in Potatoes

Agdia, Inc., a leading provider of plant diagnostic products and services, is happy to announce the introduction of an assay for detection of *Candidatus* Liberibacter solanacearum, the causal agent of zebra chip disease on potatoes, on their popular AmplifyRP® XRT platform.

The symptomology of a disease in potato known as zebra chip was first described in 1994 in Mexico. Thereafter, similar outbreaks occurred in Central America and the Southwest U.S. in the early 2000's. Disease incidence was as high as 80% in many fields and caused significant yield reduction. Initially, the causal agent was believed to be a phytoplasma, a pleiomorphic, phloem-inhabiting bacterium, which lacks a cell wall and cannot be cultured. Nevertheless, it was 2008 before the precise etiology of zebra chip disease was elucidated. At that time, two independent research groups from the U.S. and New Zealand determined the source of the disease to be a new species of bacterium, ultimately designated as *Candidatus* Liberibacter solanacearum (*Ca*. Lso).

Figure 1. The potato-tomato psyllid adult
Whitney Cranshaw, Colorado State University, Bugwood.org

Candidatus Liberibacter solanacearum is a species of bacterium that survives exclusively within plant phloem and insect tissue. Consequently, it has resisted laboratory cultivation to this point, and an official exemplar of culture does not exist, hence the designation "*Candidatus*." The Lso species is divided into groups known as haplotypes (haploid genotypes) based on genetic variability, plants affected, insect vectors and geographic distribution. Haplotypes A and B are widespread throughout the U.S., Mexico, Central America and New Zealand. These variants cause zebra chip disease in potato and yellows disease in several additional members of Solanaceae and are spread by the potato-tomato psyllid, *Bactericera cockerelli* (Hemiptera) (**Figure 1.**).

Haplotypes C, D and E are present throughout Europe and North Africa, causing severe yellows disease in members of Apiaceae, including carrot and celery. These are vectored by two species of carrot psyllids: *Trioza apicalis* and *Bactericera trigonica*. Research shows that *T*. *apicalis* and *B*. *trigonica* cannot feed efficiently on solanaceous crops, thereby limiting the spread of Lso in Europe. Moreover, *Bactericera cockerelli* and its

vectored haplotypes have not been detected in Europe. Nevertheless, transmission pathways are dynamic, and the potential for the introduction of zebra chip disease to the European potato market represents a significant phytosanitary risk.

Figure 2. Symptoms of zebra chip disease on potato foliage. Whitney Cranshaw, Colorado State University, Bugwood.org

Aerial symptoms of Lso infections in solanaceous plants include stunting, severe chlorosis and purpling of new foliage, leaf deformation, rosetting, aerial tuber production (potato), scorch and disruption of fruit set and quality (**Figure 2.**). In potato, subterranean symptoms include tuber deformation, stolon collapse and vascular ring browning. When fried for chipping, vascular browning becomes darker in infected tubers, making chips bitter tasting and unmarketable.

Candidatus Liberibacter solanacearum is transmitted locally by the potato-tomato psyllid in a manner similar to that of persistent-propagative plant viruses. The psyllid acquires the bacterium by feeding on the phloem of infected plants. If swallowed, the bacterium passes through the alimentary canal wall into the hemolymph and surrounding tissues. Here, the bacterium reproduces for several days, ultimately accumulating in the insect's salivary glands. Thereafter, it is transmitted to a healthy plant via salivary secretions during feeding activities. Long-distance spread of Lso occurs via the movement of infected plant parts and plant parts harboring infective stages of the psyllid. Infected seed tubers can produce infected plants, but this occurs at a low rate, and plants are short-lived. True seed of solanaceous plants is not known to transmit Lso; however, Lso is reported to be seed-born in carrots.

Management of zebra chip disease is multi-faceted and includes early-season monitoring of psyllid populations, insecticide treatments and identification of infected plant materials. Symptoms of zebra chip disease on aerial plant parts are similar to those caused by phytoplasmas, viruses and nutrient imbalances. Therefore, the use of a molecular diagnostic specific for Lso following visual inspection is essential.

Accurate and timely diagnosis of plants infected with Lso is paramount to successful management throughout the growing season, including the establishment and transplanting phases. And Agdia's new AmplifyRP® XRT assay for detection of *Candidatus* Liberibacter solanacearum provides growers from multiple cropping systems with varying levels of expertise with a powerful diagnostic tool. AmplifyRP® XRT technology promotes the rapid amplification and detection of nucleic acid targets, DNA or RNA, while maintaining a single operating temperature of 42°C. The AmplifyRP® XRT products achieve target sensitivity and specificity comparable to qPCR while having clear

advantages over the lab-based technology. AmplifyRP® XRT products do not require a nucleic acid purification step, as crude sample extracts are prepared using a simple extraction buffer and tested directly. When paired with Agdia's AmpliFire® isothermal fluorometer, the XRT system is a rapid, user-friendly tool that can be implemented in the field or the lab by personnel with limited experience in molecular diagnostics (**Figure 3.**).

Agdia states their new assay is specific for Lso and will detect haplotypes A-F. Moreover, the assay was validated against a comprehensive panel of potential cross reactors, including phytoplasmas and viruses known to infect host plants and cause similar symptomology as Lso. Agdia recommends using leaf, petiole, stem or tuber core tissue with this assay. Inhibition was observed when using pepper leaf, stem and petiole tissue. Please see Agdia's product page to access a comprehensive validation report. The introduction of Agdia's new AmplifyRP® XRT assay expands their catalog to 29 products on this platform.

Figure 3. Field-deployable AmpliFire® isothermal fluorometer

As a leading provider of diagnostic solutions for agriculture, Agdia, Inc. has been serving plant breeders, propagators, growers, universities, regulatory organizations and private testing laboratories since 1981. The company offers a comprehensive portfolio of validated, easy-to-use diagnostics for identifying plant pathogens, plant hormones, and transgenic traits and comprehensive customer support on all products. In addition, Agdia operates an ISO accredited, in-house, testing services laboratory. For more information on Agdia's full line of products and services, visit the company's website at www.agdia.com, e-mail info@agdia.com, phone 1-574-264-2615 (toll-free 800-622-4342) or fax 1-574-264-2153.