

## 2014 National Plum Pox Virus (PPV) Survey Plan

### **Background:**

PPV is considered to be the most severe disease affecting commercial *Prunus* species. The disease most commonly affects peach, nectarine, plum, and apricot, and can also infect a number of related ornamental species. While some strains of PPV show an ability to infect cherry, it has not been shown to be a host of the strains of the disease detected in North America. Infected trees produce fruit that is of poor quality and unusable for consumption. Infected trees become unproductive and experience premature mortality. Prior to 1999 PPV had not been known to be present in North America.

In 1999 PPV was detected in Adams County, PA by an observant grower. APHIS, working cooperatively with the Pennsylvania Dept. of Agriculture established a PPV eradication program. The program included survey, regulatory, and control components, and resulted in the eradication of the disease in PA in 2009. The infestation in PA increased awareness of PPV, and as a result of surveys conducted in Canada was found in the Niagara area of Ontario Province in 2000. An eradication program was established by the Canadian government and the Ontario Ministry of Agriculture and is still underway.

In 2000 APHIS began working in cooperation with states with commercial stone fruit production to conduct a national PPV survey. 2010 was the last year that significant national survey activities were carried out. In 2006 PPV was found in a single tree in MI, and in a commercial orchard in NY. The detection in NY was near the infested area in Ontario, leading to speculation that Canada might have been the source of the NY infestation. In 2009 eradication of PPV was declared in MI. APHIS continues to work with the State of New York in conducting an eradication program.

In 2012 it was determined that a national survey for PPV was a high priority for Farm Bill section 10201 funding. The following information should be considered by participating states in developing work plans for conducting current year PPV surveys.

### **General Principles of PPV Testing and Survey Design:**

The way in which PPV infects and becomes systemic in host plants significantly affects the way in which surveys may be conducted. The virus is transmitted by a number of species of aphids commonly present in the U.S., as well as through the use of infected budwood. The strain of PPV present in North America was initially and continues mainly to be the D strain – a strain that has never been shown to be seed borne. While

infected trees may sometimes show symptoms on leaves and fruit, visual inspection of trees is not the basis for a reliable survey method. Many host species and cultivars do not show any obvious symptoms. In addition, there is often considerable latency in the development and spread of the disease in infected trees, often resulting in a period of up to two years between the time a tree becomes infected and the time infection can be detected by the laboratory methods currently in use. For this reason it is not productive or accurate to test samples obtained from nursery stock.

APHIS has determined the most reliable and least expensive way of detecting the presence of PPV in leaf samples collected from infected trees is through the use of an double antibody sandwich ELISA test produced by AgDia Company. This test had been shown to be capable of detecting the presence of PPV in leaves, fruit, and buds collected from infected plants. The test is polyclonal, capable of detecting not only the D strain currently present in the U.S., but also the M, C, W, EA, and Rec strains of PPV. AgDia can provide both in-house testing services as well as ELISA reagent sets for purchase by laboratories wishing to do their own testing. Additional details on the specifics of conducting the ELISA tests will be provided. Laboratories processing their own samples have reported per-sample costs in the range of \$3.50 to \$5.00. We anticipate that the per-sample cost of having samples processed by AgDia to be a little more than \$7.00 per sample.

In the event that participating states choose to process their own samples, APHIS will be able to provide (under permit, of course) limited amounts of lyophilized PPV positive leaves to be used by the laboratories to confirm their ability to detect the presence of PPV in samples. At the present time APHIS does not anticipate conducting a formal accreditation program for participating laboratories.

Detection survey for PPV in orchards is based on the use of a hierarchical sampling method. This involves collecting 8 leaf samples from each of 25 percent of the trees in an orchard. Trees to be sampled are selected in groups of 4, with 32 leaves that are collected from the four trees being ELISA tested as four 8 leaf samples. To determine the total number of samples to be collected in an orchard being surveyed, the following formula can be used:

$$\frac{\text{Number of trees per acre} \times \text{number of acres of in the orchard}}{4}$$

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Thus, in a 9 acre orchard with a planting rate of 140 trees per acre, 315 eight leaf samples would be submitted for ELISA testing. Sampling at lower rates significantly reduces the likelihood of detecting PPV, and sampling at higher rates is only warranted if there are unusual risk factors present.

APHIS recommends that the selection of trees or orchards to be sampled and the rate at which sampling is conducted should be based on the risk associated with the material being sampled. At the present time, outside of NY, there is no reason to think that any orchards or budwood source trees in any state presents a particular risk associated with having been exposed to PPV. Thus, decisions on which trees and orchards are selected for inclusion in the survey are more based on the risk that might be present should PPV be found. Foundation trees used to produce budwood for the production of registered nursery stock trees or certified budwood source trees present the highest risk, and all of these trees should be sampled at the level of 8 leaves per tree.

The second level of risk are individual trees outside of orchard settings, or orchards from which budwood is or recently has been cut – either registered or certified budwood, or common budwood. Individual trees outside of orchards should be sampled at the level of 8 leaves per tree. All of the orchards containing trees from which budwood has been cut should be sampled at the 25 percent level using the hierarchical sampling model. Once orchards containing trees used for budwood production have been sampled, the remaining funds may be used to conduct hierarchical surveys of a representative group of orchards throughout the state.

The best time to collect samples is in the spring - from the time new leaves are fully expanded until it becomes too hot for reliable leaf samples to be collected. Once average daily temperatures exceed 95 degrees F the virus titer in leaves declines significantly and samples from infected trees would be likely produce false negative results. In some southern areas this means that there is a relatively narrow window in which samples can usefully be collected – while in other more northern areas the window in which samples can be collected may be as long as several months.

### **In Summary:**

- All foundation trees should be sampled.
- All other trees used as budwood sources and not in orchards should be sampled.
- Any orchards containing trees from which budwood is cut should be sampled at a 25 percent level, using a hierarchical sampling model.
- Any remaining funds can be used to survey a representative group of orchards, with sampling being conducted at the same 25 percent level.
- States can either process their own samples using ELISA kits purchased from AgDia, or can have samples processed by another state or by AgDia.