United States Department of Agriculture

Animal and Plant Health Inspection Service

Plant Protection and Quarantine

New Pest Response Guidelines

Scots Pine Blister Rust (*Cronartium flaccidum* and *Peridermium pini*)

First Edition Issued 2012

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Cover Image

Symptoms of *Cronartium flaccidum* on Scots pine (*Pinus sylvestris*). Source: (Risto Jalkanen).

Chapter 1

Introduction

Contents

Introduction

Use *New Pest Response Guidelines: Scots Pine Blister Rust (*Cronartium flaccidum *and* Peridermium pini*)* when designing a program to detect, monitor, control, contain, or eradicate, an outbreak of Scots pine blister rust in the United States and collaborating territories.

The United States Department of Agriculture, Animal and Plant Health Inspection Service, Plant Protection and Quarantine (USDA–APHIS–PPQ) developed the guidelines through discussion, meeting, or agreement with staff members at the USDA-Agricultural Research Service and advisors at universities.

Any new detection may require the establishment of an Incident Command System to facilitate emergency management. This document is meant to provide the necessary information to launch a response to a detection of *Cronartium flaccidum* and *Peridermium pini*.

If *Cronartium flaccidum* and *Peridermium pini* are detected, PPQ personnel will produce a site-specific action plan based on the guidelines. As the program develops and new information becomes available, the guidelines will be updated.

Users

The guidelines is intended as a reference for the following users who have been assigned responsibilities for a plant health emergency for any of the selected Scots pine blister rust:

- PPQ personnel
- Emergency response coordinators
- State agriculture department personnel
- Others concerned with developing local survey or control programs

Contacts

When an emergency pest response program for *Cronartium flaccidum* and *Peridermium pini* has been implemented, the success of the program depends on the cooperation, assistance, and understanding of other involved groups. The appropriate liaisons and information officers should distribute news of the program's progress and developments to interested groups, including the following:

- Academic entities with agricultural interests
- Agricultural interests in other countries
- Commercial interests
- Grower groups such as specific commodity or industry groups
- Land-grant universities and Cooperative Extension Services

- National, State and local news media
- Other Federal, State, county, and municipal agricultural officials
- Public health agencies
- The public
- State and local law enforcement officials
- Tribal governments

Initiating an Emergency Pest Response Program

An emergency pest response program consists of detection and delimitation, and may be followed by programs in regulation, containment, eradication and control. The New Pest Advisory Group (NPAG) will evaluate the pest. After assessing the risk to U.S. plant health, and consulting with experts and regulatory personnel, NPAG will recommend a course of action to PPQ management.

Follow this sequence when initiating an emergency pest response program:

1. A new or reintroduced pest is discovered and reported
2. The pest is examined and pre-identified by regional or area identifier
3. The pest's identity is confirmed by a national taxonomic authority recognized by USDA–APHIS–PPQ-National Identification System
4. Published New Pest Response Guidelines are consulted or a new NPAG is assembled in order to evaluate the pest
5. Depending on the urgency, official notifications are made to the National Plant Board, cooperators, and trading partners
6. A delimiting survey is conducted at the site of detection
7. An Incident Assessment Team may be sent to evaluate the site
8. A recommendation is made, based on the assessment of surveys, other data, and recommendation of the Incident Assessment Team or the NPAG, as follows:
   - **A.** Take no action
   - **B.** Regulate the pest
   - **C.** Contain the pest
   - **D.** Suppress the pest
   - **E.** Eradicate the pest
9. State Departments of Agriculture are consulted

10. If appropriate, a control strategy is selected
11. A PPQ Deputy Administrator authorizes a response
12. A command post is selected and the Incident Command System is implemented
13. State departments of agriculture cooperate with parallel actions using a Unified Command structure
14. Traceback and trace-forward investigations are conducted
15. Field identification procedures are standardized
16. Data reporting is standardized
17. Regulatory actions are taken
18. Environmental Assessments are completed as necessary
19. Treatment is applied for required pest generational time
20. Environmental monitoring is conducted, if appropriate
21. Pest monitoring surveys are conducted to evaluate program success
22. Programs are designed for eradication, containment, or long-term use

Preventing an Infestation

Federal and State regulatory officials must conduct inspections and apply prescribed measures to ensure that pests do not spread within or between properties. Federal and State regulatory officials conducting inspections should follow the sanitation guidelines in the section *Survey Procedures* on page 4-1 before entering and upon leaving each property to prevent contamination.

Scope

The guidelines is divided into the following chapters:

8. *Pathways on page 8-1*

The guidelines also includes appendixes, a references section, a glossary, and an index.

The Introduction contains basic information about the guidelines. This chapter includes the guideline's purpose, scope, users, and application; a list of related documents that provide the authority for the guidelines content; directions about how to use the guidelines; and the conventions (unfamiliar or unique symbols and highlighting) that appear throughout the guidelines.

Authorities

The regulatory authority for taking the actions listed in the guidelines is contained in the following authorities:

- Plant Protection Act of 2000 (Statute 7 USC 7701-7758)
- Executive Order 13175, Consultation and Coordination with Indian and Tribal Governments
- Fish and Wildlife Coordination Act
- National Historic Preservation Act of 1966
- Endangered Species Act
- Endangered and Threatened Plants (50 CFR 17.12)
- National Environmental Policy Act

Program Safety

Safety of the public and program personnel is a priority in pre-program planning and training and throughout program operations. Safety officers and supervisors must enforce on-the-job safety procedures.

Support for Program Decisionmaking

USDA–APHIS–PPQ-Center for Plant Health, Science and Technology (CPHST) provides technical support to emergency pest response program directors about risk assessments, survey methods, control strategies, regulatory treatments, and other aspects of pest response programs. PPQ managers meet with State departments of agriculture in developing guidelines and policies for pest response programs.

How to Use the Guidelines

The guidelines is a portable electronic document that is updated periodically. Download the current version from its source, and then use Adobe Reader® to view it on your computer screen. You can print the guidelines for convenience. However, links and navigational tools are only functional when the document is viewed in Adobe Reader®. Remember that printed copies of the guidelines are obsolete once a new version has been issued.

Conventions

Conventions are established by custom and are widely recognized and accepted. Conventions used in the guidelines are listed in this section.

Advisories

Advisories are used throughout the guidelines to bring important information to your attention. Please carefully review each advisory. The definitions have been updated so that they coincide with the America National Standards Institute (ANSI) and are in the format shown below.

EXAMPLE Example provides an example of the topic.

Important Important indicates information that is helpful.

CAUTION

CAUTION indicates that people could possibly be endangered and slightly hurt.

DANGER

DANGEROUS indicates that people could easily be hurt or killed.

NOTICE

NOTICE indicates a possibly dangerous situation where goods might be damaged.

WARNING

WARNING indicates that people could possibly be hurt or killed.

Boldfacing

Boldfaced type is used to highlight negative or important words. These words are: never, not, do not, other than, prohibited.

Lists

Bulleted lists indicate that there is no order to the information being listed. Numbered lists indicate that information will be used in a particular order.

Disclaimers

All disclaimers are located on the unnumbered page that follows the cover.

Table of Contents

Every chapter has a table of contents that lists the heading titles at the beginning to help facilitate finding information.

Control Data

Information placed at the top and bottom of each page helps users keep track of where they are in the guidelines. At the top of the page is the chapter and first-level heading. At the bottom of the page is the month, year, title, and page number. PPQ–EDP-Emergency Programs is the unit responsible for the content of the guidelines.

Change Bar

A vertical black change bar in the left margin is used to indicate a change in the guidelines. Change bars from the previous update are deleted when the chapter or appendix is revised.

Decision Tables

Decision tables are used throughout the guidelines. The first and middle columns in each table represent conditions, and the last column represents the action to take after all conditions listed for that row are considered. Begin with the column headings and move left-to-right, and if the condition does not apply, then continue one row at a time until you find the condition that does apply.

Table 1-1 How to Use Decision Tables

If you:	And if the condition applies:	Then:
Read this column cell and row first	Continue in this cell	TAKE the action listed in this cell
Find the previous condition did not apply, then read this column cell	Continue in this cell	TAKE the action listed in this cell

Footnotes

Footnotes comment on or cite a reference to text and are referenced by number. The footnotes used in the guidelines include general text footnotes, figure footnotes, and table footnotes. General text footnotes are located at the bottom of the page.

When space allows, figure and table footnotes are located directly below the associated figure or table. However, for multi-page tables or tables that cover the length of a page, footnote numbers and footnote text cannot be listed on the same page. If a table or figure continues beyond one page, the associated footnotes will appear on the page following the end of the figure or table.

Heading Levels

Within each chapter and section there can be four heading levels; each heading is green and is located within the middle and right side of the page. The first-level heading is indicated by a horizontal line across the page, and the heading follows directly below. The second-, third-, and fourth-level headings each have a font size smaller than the preceding heading level. The fourth-level heading runs in with the text that follows.

Hypertext Links

Figures, headings, and tables are cross-referenced in the body of the guidelines and are highlighted in boldface type. These appear in blue hypertext in the online guidelines.

Italics

The following items are italicized throughout the guidelines:

- Cross-references to headings and titles
- Names of publications
- Scientific names

Numbering Scheme

A two-level numbering scheme is used in the guidelines for pages, tables, and figures. The first number represents the chapter. The second number represented the page, table, or figure. This numbering scheme allows for identifying and updating. Dashes are used in page numbering to differentiate page numbers from decimal points.

Transmittal Number

The transmittal number contains the month, year, and a consecutively-issued number (beginning with -01 for the first edition and increasing consecutively for each update to the edition). The transmittal number is only changed when the specific chapter sections, appendixes, or glossary, tables, or index is updated. If no changes are made, then the transmittal number remains the unchanged. The transmittal number only changes for the entire guidelines when a new edition is issued or changes are made to the entire guidelines.

Acknowledgements

Writers, editors, reviewers, creators of cover images, and other contributors to the guidelines, are acknowledged in the acknowledgements section. Names, affiliations, and Web site addresses of the creators of photographic images, illustrations, and diagrams, are acknowledged in the caption accompanying the figure.

How to Cite the Guidelines

Cite the guidelines as follows: U.S. Department of Agriculture, Animal Plant Health Inspection Service, Plant Protection and Quarantine. 2011. *New Pest Response Guidelines: Scots Pine Blister Rust (*Cronartium flaccidum *and* Peridermium pini*).* Washington, D.C. http://www.aphis.usda.gov/import_export/plants/manuals/online_manuals.shtml

How to Find More Information

Contact USDA–APHIS–PPQ–EDP-Emergency Management for more information about the guidelines. Refer to *Resources* on page A-1 for contact information.

Chapter

2

Pest Information

Contents

Introduction

Use *Chapter 2 Pest Information* to learn more about the classification, history, host range, and biology of the host alternating rust fungus *Cronartium flaccidum* and the related pine-to-pine form *Peridermium pini*, etiologic agents of Scots pine blister rust disease on two-needle pines (*Pinus* spp.). This disease is reported to occur in Europe and Asia and it is absent from the United States and collaborating territories.

Classification

The rust fungi are Basidiomycetes of the order Uredinales that are destructive plant pathogens and generally characterized by complex life-cycles. Molecular and morphological evidence indicate that *Cronartium flaccidum* and *Peridermium pini* are taxonomically very closely related despite differences in their life cycles. *Table 2-1* on page 2-2 presents the current classification only for the heteroecious (host alternating) form *C. flaccidum*. The autoecious form *P. pini*, whose life cycle is limited to coniferous hosts, will be considered throughout the guidelines, as a host-specialized adaptation originally derived from *C. flaccidum*.

Table 2-1 Classification of *Cronartium flaccidum* (Teleomorph)[1]

Kingdom	Fungi[1]
Phylum	Basidiomycota
Class	Urediniomycetes
Order	Urediniales
Family	Cronartiaceae
Genus	*Cronartium*
Species	*Cronartium flaccidum* (Alb. & Schwein.) G. Winter

1 CABI, 2011b.

Note: The diverse interactions of this heteroecious fungus with its hosts have resulted in a complicated nomenclature. Several species of *Cronartium* were initially identified for the rust on the various dicots that are primary hosts. An anamorph, *Peridermium cornui*, was described as the aecial form of one of these species (*C. asclepiadeum*) that is now synonymized with *C. flaccidum* (Wilson and Henderson, 1966).

Autoecious Form

Peridermium pini (Willd. : Pers.) Lév. [syn. *Endocronartium pini* (Willd.) Y. Hirats]

Synonym:

Sphaeria flaccida Alb. & Schwein.
Peridermium pini sensu auct. p.p.
Sphaeria flaccida Alb. & Schwein.
Erineum asclepiadeum Willd.
Cronartium asclepiadeum (Willd.) Fr.
Cronartium paeoniae Castagne, Cat. Pl. Mars.
Peridermium pini f. corticola Mussat

Disease Common Names:

Scots pine blister rust

Resin top disease

Scotch pine blister rust

Cronartium rust

Blister rust

Pine-stem rust

Resin canker

Two-needle pine blister rust

Historical Information

The two rust forms were previously considered distinct species based on differences in the morphology and cytology of aeciospores, aeciospore germ tubes and life cycle (Hiratsuka, 1968). Subsequent studies by Moricca et al. (1996) and Hantula et al. (1998) showed that *Cronartium flaccidum* was very closely related to *Peridermium pini* by examining internal transcribed spacer (ITS) sequences of ribosomal DNA and random amplified microsatellite (RAMS) markers. Similarly, Vogler and Bruns (1998) determined that there was a close phylogenetic relationship between C. *flaccidum* and *P. pini*.

The analysis of a broader collection of aeciospores of *Peridermium pini* and *Cronartium flaccidum* by Kasanen (1997) and Kaitera et al. (1999b) revealed that the two rust species could not be distinguished based upon germ tube morphology as previously suggested by Hiratsuka (1969). Based on molecular and morphological data, authors now consider the two fungi to be synonymous. *Peridermium pini* was shown to be clonal and it was believed to have its origin as a haploid life cycle mutant of C. *flaccidum*, which has a sexual life cycle (Kasanen, 2001; Kasanen et al., 2000).

Damage

The disease currently affects several hard or two-needle pine species distributed throughout Europe and Asia but is not known to occur in North America. In the United States, *Cronartium flaccidum* alternate hosts are commonly present together with extensive native and non-native pine species, including Scots pine, which would be significantly impacted should this disease be introduced.

Cronartium flaccidum is included in the List of Regulated Pest by the United States (USDA–APHIS, 2000). This rust can be damaging on native and introduced pines or the alternate plant host. Because the infections on pines develop slowly, the fungus might be easily overlooked, and its accidental introduction could occur through importation of conifer seedlings or trees.

Blister rust caused by *Cronartium flaccidum* has been described as severe, rapidly advancing, and dangerous (Hantula et al., 2002; Ragazzi and Dellavalle Fedi, 1983). Blister rust has been a major factor in reducing forest productivity for centuries (Hantula et al., 2002). In the 1960s and 1970s the heteroecious form (*C. flaccidum*) spread epidemically in Mediterranean countries and decimated forests of two-needle pines. The disease is severe on Scots pine (*Pinus sylvestris*). The high numbers of coniferous hosts and the very widespread distribution of one of the main alternate hosts (*Vincetoxicum hirundinaria*), led to great losses in Italy during the 1960s, where entire plantations were destroyed due to the presence of new plantings and favorable conditions (Hantula et al., 2002; Ragazzi et al., 1989; Smith et al., 1988).

According to Martinsson and Nilsson (1987), *Cronartium flaccidum* rust reduced radial stem increment by 40 to 70 percent in severe attacks and by 20 to 40 percent in minor attacks on *Pinus sylvestris* in Sweden. Kaitera et al. (1994) estimated that disease caused by *Peridermium pini* in Finland reduced the market value of saw timber trees by 18 percent and pulpwood trees by 3 percent as a result of stem lesions, and by 15 percent and 14 percent, respectively, due to the death of tree tops.

In Britain, the disease rate on Scots pine caused by the autoecious form (*Peridermium pini*) increased from the 1960s to the 1980s (Greig, 1987) causing considerable volume losses on trees with stem lesions and crown symptoms (Gibbs et al., 1987). In Greece, in a six year period *Cronartium flaccidum* had infected or killed over 5000 m^3 in a forest of approximately 1000 ha (Diamandis and Kam, 1986).

Economic Impact

Scots pine (*Pinus sylvestris*), is an introduced species in North America, brought here from Europe probably in colonial days. Scots pine has been widely planted in the United States, especially in the Northeast, Lake States, Central States, and Pacific Northwest. It is now considered naturalized in parts of New England and the Lake States. The species has also been planted across southern Canada. Although it is used for both pulpwood and sawlogs, its principal value in the United States appears to be as a Christmas tree, as an ornamental, and for erosion control.

To evaluate the potential economic impact to the United States caused by the introduction and establishment of *Cronartium flaccidum,* the value of its major (common) host Scots pine was taken into consideration and its use for producing Christmas tree. The U.S. Christmas tree sales data are summarized in *Table 2-2* on page 2-6.

According to USDA's 2009 Census of Horticultural Specialties (USDA–NASS, 2010), nearly 2,700 operations sold 12.9 million Christmas trees valued at $249.8 million in 2009 (*Table 2-3* on page 2-6). Of those, Scots pine sales accounted for $9.8 million.

A more comprehensive evaluation of *Cronartium flaccidum* economic impact would have to consider that Scots pine is only one among more than 15 other pine host species. Susceptible *Pinus* spp. known to be present in the United States are: *Pinus halepensis*, *P. mugo*, *P. nigra*, *P. pinaster*, *P. pinea*, *P. ponderosa* and *P. wallichiana*. In the U.S. there are at least five *Pinus* spp. which are listed as federally threatened or endangered and a potential host, (USFWS, 2011). Moreover, there is some indication that *P. resinosa* (red pine), considered endangered in Connecticut, Illinois, New Jersey and closely related to known hosts, may act as a weak host for this rust (Raddi and Fagnani, 1978).

An estimate of the potential disease eradication/management costs can be made based on a similar disease of pine trees, white pine blister rust, caused by C*ronartium ribicola* that was introduced into western North America in the late 1800s. The attempts to control this devastating disease of pine are estimated to have cost over one billion in current U.S. dollars not including the effects on forest productivity and the ecological impacts (Geils et al., 2009). The introduction of white pine blister rust originated from nursery stocks, which can be important long-distance vectors for these pathogens (Parke and Grünwald, 2012). *Cronartium flaccidum* has alternate hosts in the ornamental plant industry including *Impatiens* spp. and *Paeonia* spp. Refer to *Hosts* on page 2-12. These nursery industries represented over $266 million and $8 million, respectively, in aggregate wholesale market value in 2009 (NASS, 2010). It would be important to consider the impacts of disease control and spread within these nursery industries, should Scots pine blister rust be introduced to the United States.

Table 2-2 Value of U.S. Christmas Tree Production Potentially Affected by *Cronartium flaccidum*[1]

Crop	Value of US production (1,000 $)
Cut Christmas trees, total	249,821
Scots pine (*Pinus sylvestris*)	9,786

1 USDA—NASS, 2010.

Ecological Range

Cronartium flaccidum is known from Europe and parts of northern and eastern Asia in the Northern Hemisphere (Smith et al., 1988). The autoecious form *Peridermium pini* only occurs in Europe, according to Hiratsuka (1969), but Tai (1979) and Chen (2002) report it from China. Given the difficulty of distinguishing the two forms on *Pinus* without molecular examination or inoculation of dicot hosts (Hantula et al., 2002), some of the Asian reports might be erroneous.

Asia: China, Japan, and Korea.

Europe: Armenia, Austria, Azerbaijan, Belgium, Bosnia and Herzegovina, Bulgaria, Croatia, Czech Republic, Denmark, Estonia, Finland, France, Georgia, Germany, Greece, Hungary, Italy, Kosovo, Lithuania, Macedonia, Montenegro, Netherlands, Norway, Poland, Portugal, Romania, Russia, Scotland, Serbia, Slovakia, Slovenia, Spain, Sweden, Switzerland, Ukraine, and United Kingdom (CABI, 2010; Diamandis and Kam, 1986; Geils et al., 2009; Gibbs et al., 1988).

A report from India is considered invalid (CABI, 2010). According to Farr and Rossman (2011) there is a record of a synonym of this pathogen (*Cronartium asclepiadeum*) from Vermont in 1898. The validity of this record is not known; however, all other sources indicate that *C. flaccidum* is exotic to the United States.

Table 2-3 *Cronartium flaccidum* Reported World Distribution and References

Geographic Region	Country	References
Asia		
Armenia	Present, no further details	CMI, 1989

Table 2-3 ***Cronartium flaccidum*** **Reported World Distribution and References**

Geographic Region	Country	References
Azerbaijan	Present, no further details	CMI, 1989
China	Restricted distribution	Chen, 2002
-Anhui	Present, no further details	Cheng et al., 1998
-Guizhou	Present, no further details	Chen, 2002; Jing and Wang, 1989
-Heilongjiang	Present, no further details	Cheng et al., 1995; CMI, 1989
-Henan	Present, no further details	Chen, 2002
-Hubei	Present, no further details	Chen, 2002; Jing and Wang, 1989
-Jiangsu	Present, no further details	CMI, 1989
-Jilin	Present, no further details	CMI, 1989
-Liaoning	Present, no further details	CMI, 1989
-Nei Menggu	Present, no further details	Chen, 2002
-Shaanxi	Present, no further details	Cao et al., 2000; Jing et al., 1995; Jing and Wang, 1989; Zhuang and X., 2005
-Shanxi	Present, no further details	Cheng et al., 1998
-Sichuan	Present, no further details	CMI, 1989; Jing and Wang, 1989
-Tibet	Present, no further details	Chen, 2002
-Yunnan	Present, no further details	CMI, 1989
-Zhejiang	Present, no further details	CMI, 1989
Georgia (Republic of)	Present, no further details	CMI, 1989
India	Absent, invalid record	-
Japan	Present, no further details	CMI, 1989
-Hokkaido	Present, no further details	Hiratsuka, 1932
-Honshu	Widespread	Kobayashi, 2007
-Kyushu	Present, no further details	Kobayashi, 2007
Kazakhstan	Present, no further details	Churakov, 1989
Korea, DPR	Present, no further details	-
Korea, Republic of	Present, no further details	Cho and Shin, 2004; CMI, 1989; Yi et al., 1985
Taiwan	Present, no further details	Hiratsuka and Chen, 1991
Europe		
Austria	Present, no further details	CMI, 1989; Widder, 1941
Belgium	Present, no further details	CMI, 1989
Bulgaria	Widespread	CMI, 1989; Denchev, 1995; Widder, 1941
Czechoslovakia (former)	Widespread	CMI, 1989; Klebahn, 1938
Denmark	Present, no further details	CMI, 1989
Estonia	Present, no further details	CMI, 1989

Table 2-3 *Cronartium flaccidum* Reported World Distribution and References

Geographic Region	Country	References
Finland	Present, no further details	CMI, 1989; Hantula et al., 1998; Kaitera and Hantula, 1998
Former USSR	Present, no further details	Leont'eva and Stenina, 1990; Rozhkov, 1975; Storo-zhenko, 1987
France	Present, no further details	CMI, 1989
Germany	Present, no further details	CMI, 1989; Klebahn, 1938
Greece	Present, no further details	CMI, 1989; Diamandis and Kam, 1986
Hungary	Widespread	CMI, 1989; Szabo, 1998
Ireland	Absent, invalid record	-
Italy	Present, no further details	CMI, 1989; Moricca and Ragazzi, 1996; Moriondo, 1975; Raddi et al., 1979; Ragazzi and Moriondo, 1980
Latvia	Present, no further details	Kuprevich and Transchel, 1957
Lithuania	Present, no further details	Kuprevich and Transchel, 1957
Montenegro	Present, no further details	Karadzic´ and Vujanovic´, 2009
Netherlands	Present, no further details	CMI, 1989; Hiratsuka, 1968
Norway	Widespread	CMI, 1989; Hiratsuka, 1968; Roll-Hansen, 1973
Poland	Present, no further details	CMI, 1989; Mulenko et al., 2004; Siwecki and Cho-jnacki, 1989
Portugal	Present, few occurrences	-
-Azores	Present, no further details	-
-Portugal (mainland)	Present, no further details	CMI, 1989; Gonçalves, 1936
Romania	Present, no further details	CMI, 1989; Klebahn, 1938
Russian Federation	Restricted distribution	Kuprevich and Transchel, 1957
-Central Russia	Present, no further details	Kuprevich and Transchel, 1957
-Eastern Siberia	Present, no further details	Kuprevich and Transchel, 1957
-Northern Russia	Present, no further details	CMI, 1989
-Russian Far East	Present, no further details	Azbukina, 1995; CMI, 1989; Kakishima et al., 1995
-Siberia	Present, no further details	-
-Southern Russia	Present, no further details	Kuprevich and Transchel, 1957

Table 2-3 ***Cronartium flaccidum*** **Reported World Distribution and References**

Geographic Region	Country	References
-Western Siberia	Present, no further details	Kuzmina and Kuz'Min, 2008
Serbia	Present, no further details	-
Slovenia	Present, no further details	Jurc, 2007
Spain	Present, no further details	CMI, 1989
Sweden	Widespread	CMI, 1989; Hiratsuka, 1968; Klingstrom, 1973; Martinsson and Nilsson, 1987
Switzerland	Present, no further details	CMI, 1989; Widder, 1941
Ukraine	Present, no further details	CMI, 1989; Dudka et al., 2004
United Kingdom	Restricted distribution	CMI, 1989; Wilson and Henderson, 1966
-England and Wales	Restricted distribution	-
-Scotland	Present, no further details	Greig, 1987; Greig and Sharpe, 1991; Pei and Gibbs, 1991
Yugoslavia (former)	Present, no further details	CMI, 1989
Yugoslavia (Serbia and Montenegro)	Present, no further details	Widder, 1941
-Siberia	Present, no further details	-
-Southern Russia	Present, no further details	Kuprevich and Transchel, 1957
-Western Siberia	Present, no further details	Kuzmina and Kuz'Min, 2008
Serbia	Present, no further details	-
Slovenia	Present, no further details	Jurc, 2007
Spain	Present, no further details	CMI, 1989
Sweden	Widespread	CMI, 1989; Hiratsuka, 1968; Klingstrom, 1973; Martinsson and Nilsson, 1987
Switzerland	Present, no further details	CMI, 1989; Widder, 1941
Ukraine	Present, no further details	CMI, 1989; Dudka et al., 2004
United Kingdom	Restricted distribution	CMI, 1989; Wilson and Henderson, 1966
-England and Wales	Restricted distribution	-
-Scotland	Present, no further details	Greig, 1987; Greig and Sharpe, 1991; Pei and Gibbs, 1991
Yugoslavia (former)	Present, no further details	CMI, 1989
Yugoslavia (Serbia and Montenegro)	Present, no further details	Widder, 1941

Potential Distribution

The pathogen is not currently known to exist in the United States, but poses a serious threat to the vast coniferous forest habitat in this country (*Figure 2-1* on page 2-11). *Cronartium flaccidum* is known to have many pine hosts. *Pinus sylvestris* is considered a preferred host, but the pathogen has been detected on more than 15 pine species. Likewise *Melampyrum sylvaticum* (small cow-wheat) and *Vinetoxicum hirundinaria* (Louise's swallow wort) are considered the primary alternate (telial) hosts, though several alternate hosts have been reported including: *Asclepias* spp., *Impatiens* spp., *Loasa* spp., *Melampyrum* spp., *Nemesia* spp., *Paeonia* spp., *Pedicularis* spp., *Ruellia* spp., *Schizanthus* spp., *Tropaeolum* spp., *Verbena* spp., and *Vincetoxicum* spp. Natural spread of rust aeciospores, urediniospores and sporidia is likely limited to a few miles (Alexopoulos et al., 1996; Hunt, 1997). Aeciospores and urediniospores may be disseminated greater distances than the basidiospores, which may be limited to less than 500 meters (Hunt, 1997). A potential association between rust spore dispersal and insect vectors has been proposed, but results were inconclusive (Pappinen and Weissenberg, 1994). The most probable means of introduction of either form of the rust would come from seedlings or young trees transported while the infections were still latent (USDA–APHIS, 2008).

With the exception of Ponderosa pine, most United States species were considered to have a high degree of resistance to *Cronartium flaccidum* by Raddi and Fagnani (1978). If this rust has or gains the capacity to infect North American pines, the economic and ecological impact would be incalculable (Geils et al., 2009).

For comparison, it has been estimated that control of white pine blister rust (caused by *C. ribicola*) has cost over 1 billion in current U.S. dollars since its introduction into North America in the 1900s (Geils et al. 2009). *Cronartium flaccidum* has caused much greater losses in forest productivity and ecological impacts across Europe.

In the United States, Scots pine has been planted for erosion control, as an ornamental, and also harvested for pulp and timber; however, its primary economic value is currently for Christmas trees (although other conifers are more recently favored). It has been widely planted in the colder regions of North America and is naturalized in the U.S. Northeast, Midwest, and Pacific Northwest (Geils et al., 2009). In 2002, Oregon, North Carolina, Michigan, Pennsylvania, Wisconsin, Washington, New York, and Virginia were the top Christmas tree producing States. The majority of Scots pine is grown primarily in the Great Lakes States (*Figure 2-2* on page 2-11). Michigan was the top producer of Christmas trees in 1998 (Geils et al., 2009). These areas would be at high risk based solely on host availability, which include *Pinus nigra* and *P. mugo* (*Figure 2-2* on page 2-11).

Based on a climatic model, a recent risk map (*Figure 2-3* on page 2-12) indicates that northeastern United States and portions of Washington State have the greatest risk for *Cronartium flaccidum* establishment.

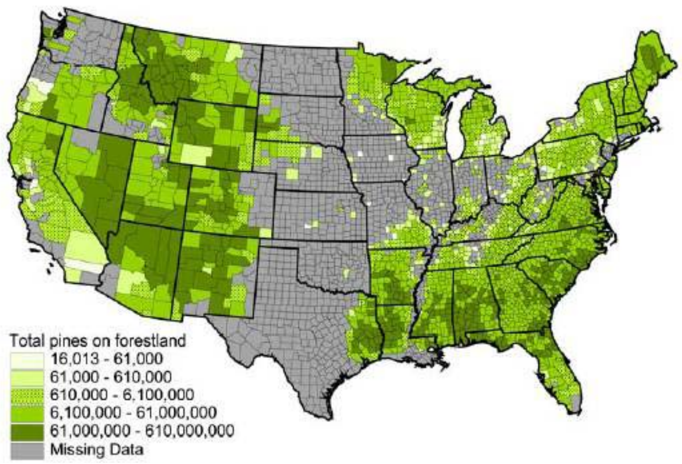

Figure 2-1 Number of Pines per County of Forestland Based on 2004 to 2006 Surveys (USDA Forest Service, Forest Inventory and Assessment Image from Venette, 2008)

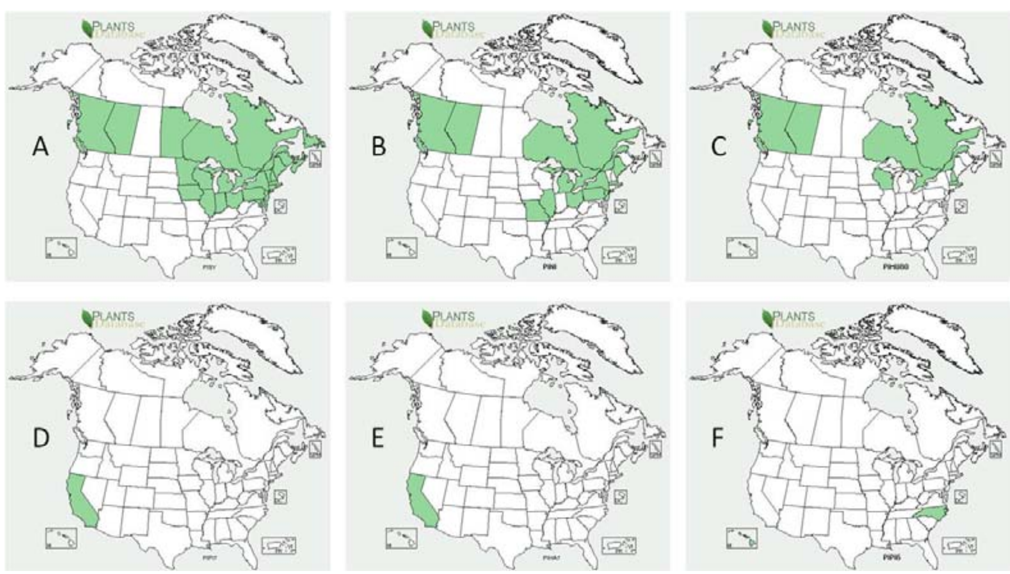

Figure 2-2 Distribution of Six Pine Species Known to be Potential Hosts of *Cronartium flaccidum* Within the United States and Canada. A- *Pinus sylvestris*; B- *P. nigra*; C- *P. mugo*; D- *P. pinea*; E- *P. halepensis*; F- *P. pinaster* (http://plants.usda.gov/java/profile?symbol=PISY)

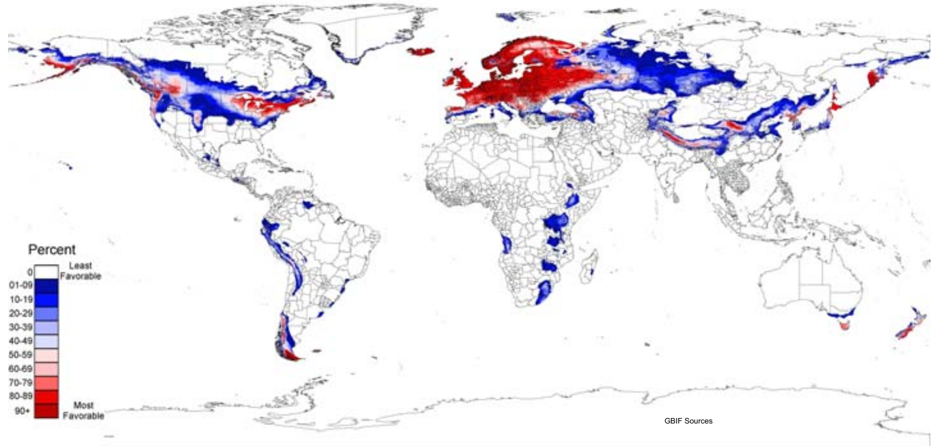

Figure 2-3 Risk Map for Climate Condition Favorable to *Cronartium flaccidum* Within the United States; Map Courtesy of USDA–APHIS–PPQ–CPHST (https://www.nappfast.org/)

Hosts

Cronartium flaccidum is known to have several pine species hosts with different levels of susceptibility. *Pinus sylvestris* (Scots pine) is considered a common (although moderately resistant) host, but the pathogen has been shown to cause disease on over 15 pine species. Species with an asterisk are reported by multiple authors as being important hosts of *C. flaccidum*.

Major Pine Hosts

*Pinus brutia** (brutian pine), *Pinus densiflora** (Japanese red pine), *Pinus halepensis** (aleppo pine), *Pinus koraienis** (fruit pine), *Pinus laricio* (black pine), *Pinus massoniana** (masson pine), *Pinus montana* (dwarf mountain pine), *Pinus mugo* (mountain, mugo pine), *Pinus nigra** (black, Austrian pine), *Pinus pallasiana*, *Pinus pinaster** (maritime pine), *Pinus pinea** (stone pine), *Pinus ponderosa* (ponderosa pine), *Pinus pumila** (dwarf Siberian pine), Pinus rotunda, *Pinus sylvestris** (Scots pine), *Pinus tabuliformis** (Chinese pine), *Pinus taiwanensis* (Taiwan red pine), *Pinus takahasii*, *Pinus uncinata** (mountain pine), *Pinus wallichiana** (blue pine), and *Pinus yunnanensis** (Yunnan pine) (CABI, 2010; Mordue and Gibson, 1978; Moricca et al., 1996; Ragazzi and Dellavalle Fedi, 1982).

Alternate hosts of this rust belong to a diverse group of herbaceous species in dicotyledonous families such as *Asclepiadaceae*, *Paeoniaceae*, and *Scrophulariaceae*.

Alternate Angiosperm Hosts

Asclepias spp. (milkweeds), *Asclepias cornuti* (milkweed), *Asclepias purpurascens* (purple milkweed), *Delphinium delavayi* (Delavayi larkspur), *Euphrasia brevipila* (drug eyebright), *Euphrasia maximowiczkii* (an-jeun-jop-ssal-pul), *Gentiana asclepiadea* (willow gentian), *Grammatocarpus* spp. (twining grammatocarpus), *Impatiens* spp. (impatiens, touch-me-knots), *Loasa* spp. (loasa), *Melampyrum* spp. (cow-wheats), *Melampyrum arvense* (field cow-wheat), *Melampyrum cristatum* (crested cow-wheat), *Melampyrum nemorusum* (wood cow-wheat), *Melampyrum pratense* (common cow-wheat) *Melampyrum sylvaticum* (small cow-wheat), *Nemesia* spp. (nemesia), *Paeonia* spp. (peony), *Paeonia albiflora* (white peony), *Paeonia anomala* (anomalous peony), *Paeonia arborea* (mu dan), *Paeonia broteri* (Brotero's peony), *Paeonia corallina* (peony), *Paeonia cultorum* (peony), *Peonia daurica* (peony), *Peonia edulis* (peony), *Paeonia japonica* (cao shao yao), *Paeonia lactiflora* (Chinese peony), *Paeonia mascula* (peony), *Paeonia moutan* (peony), *Paeonia obovata* (Chinese peony), *Paeonia officinalis* (common peony), *Paeonia peregrine* (peregrine peony), *Peonia suffruticosa* (Japanese tree peony), *Paeonia taurica* (peony), Paeonia tenuifolia (peony), *Paeonia triternata* (peony), *Pedicularis* spp. (louseworts), *Pedicularis lapponicum* (Lapland lousewort), *Pedicularis palustris* (marsh lousewort), *Pedicularis resupinata* (fan gu ma xian hao), *Pedicularis sceptrum-carolinum* (lousewort), *Phtheirospermum japonicum* (song hao), *Ruellia* spp. (wild petunia), *Schizanthus* spp. (butterfly flower, poor man's orchid), *Siphonostegia chinensis* (yin xing cao), *Tropaeolum* spp. (nasturtium), *Verbena* spp. (verbena), *Vincetoxicum* spp. (swallow wort), *Vincetoxicum albovianum* (swallow wort), *Vincetoxicum fuscatum* (swallow wort), *Vincetoxicum hirundinaria* (= *Cynanchum laxum*, *C. vincetoxicum*) (Louise's swallow wort), *Vincetoxicum mongolicum* (hua bei bai qian), *Vincetoxicum nigrum* (black swallow wort), *Vincetoxicum officinale* (white swallow wort), *Vincetoxicum rossicum* (European swallow wort), and *Vincetoxicum scandens* (Farr and Rossman, 2011; Kaitera, 1999; Kaitera and Hantula, 1998; Kaitera and Nuorteva, 2003a, 2003b; Kaitera et al., 2005; Kaitera et al., 1999a; Kasanen, 2001; Mordue and Gibson, 1978; Moricca and Ragazzi, 1998; Roll-Hansen, 1973).

Note: Considerable variation has been found in the susceptibility of alternate hosts from different locations and the virulence of *Cronartium flaccidum* spore sources (Kaitera, 1999; Kaitera et al., 1999a; Roll-Hansen, 1973).

Raddi and Fagnani (1978) grew several pines from the United States in Europe, inoculated them with *Cronartium flaccidum*, and found several species with no mycelium in needle tissue and no pycnia, aecia, or mycelium in the stem: *Pinus clausa* (sand pine), *P. contorta* (lodgepole pine), *P. echinata* (shortleaf pine), *P. elliottii* (slash pine), *P. glabra* (spruce pine), *P. radiata* (Monterey pine), *P. resinosa* (red pine), *P. serotina* (pond pine), *P. taeda* (loblolly pine), and *P. virginiana* (Virginia pine). They considered these pines to have a high degree of resistance to *C. flaccidum*, although some did display spotted seedlings. Kaitera and Nuorteva (2008) showed no disease symptoms on artificially inoculated *P. contorta* (lodgepole pine), *P. peuce* (Balkan pine), *P. strobus* (eastern white pine), *P. resinosa* (red pine), *P. banksiana* (jack pine), and *P. cembra* (swiss, arolla pine).

The USDA–APHIS produced a host density map based on *Pinus* spp. distribution data in the United States to help Cooperative Agricultural Pest Survey (CAPS) program cooperators plan surveys and for decision support (*Figure 2-4* on page 2-18).

Table 2-4 List of Reported Host Plants of *Cronartium flaccidum*

Scientific name	Common name	References
Asclepias spp.	Milkweed	Robert et al., 2005
Asclepias cornuti	Milkweed	Farr and Rossman, 2011
Cynanchum spp.		CABI, 2011a
Cynanchum fuscatum		Farr and Rossman, 2011
Cynanchum laxum		Farr and Rossman, 2011
Cynanchum nigrum	Swallow-wort, Black	Farr and Rossman, 2011
Cynanchum scandens		Farr and Rossman, 2011
Cynanchum vincetoxicum	Swallow-wort	Farr and Rossman, 2011
Delphinium delavayi	Larkspur, Delavayi	Farr and Rossman, 2011
Euphrasia brevipila	Eyebright, Drug	Farr and Rossman, 2011
Euphrasia maximowiczii	An-jeun-jop-ssal-pul	Farr and Rossman, 2011
Gentiana spp.	Gentian	CABI, 2011a
Gentiana asclepiadea	Gentian, Willow	Farr and Rossman, 2011
Grammatocarpus volubilis	Grammatocarpus, Twining	Farr and Rossman, 2011
Impatiens spp.	Touch-me-not	CABI, 2011a
Loasa spp.	Loasa	CABI, 2011a
Loasa triphylla var. vulcanica		CABI, 2011a
Loasa vulcanica	Loasa	Farr and Rossman, 2011
Melampyrum spp.		CABI, 2011a
Melampyrum arvense	Cow-wheat, Field	Farr and Rossman, 2011
Melampyrum cristatum	Cow-wheat, Crested	Farr and Rossman, 2011
Melampyrum nemorosum	Wood Cow-wheat	Kaitera and Nuorteva, 2003a
Melampyrum pratense	Cow-wheat	Kaitera, 1999
Melampyrum sylvaticum	Cow-wheat, Small	Kaitera, 1999
Myrica asplenifolia		Farr and Rossman, 2011
Nemesia spp.		CABI, 2011a
Nemesia strumosa	Nemesia	Farr and Rossman, 2011
Nemesia versicolor		Farr and Rossman, 2011
Paeonia spp.	Peony	CABI, 2011a
Paeonia albiflora	Peony, White	Farr and Rossman, 2011
Paeonia anomala	Peony, Anomalous	Farr and Rossman, 2011
Paeonia arborea	Mu Dan	Farr and Rossman, 2011
Paeonia broteri	Paeonia broteri	Farr and Rossman, 2011
Paeonia corallina	Peony	Farr and Rossman, 2011
Paeonia cultorum		Farr and Rossman, 2011
Paeonia daurica		Dudka et al., 2004
Paeonia edulis		Farr and Rossman, 2011

Table 2-4 List of Reported Host Plants of *Cronartium flaccidum*

Scientific name	Common name	References
Paeonia japonica	Cao shao yao	Farr and Rossman, 2011
Paeonia lactiflora	Peony	Farr and Rossman, 2011
Paeonia lactiflora var. tricho-carpa		Farr and Rossman, 2011
Paeonia lactiflora var. hor-tensis	Jak-yag	Farr and Rossman, 2011
Paeonia mascula		Farr and Rossman, 2011
Paeonia moutan	Peony, Tree	Farr and Rossman, 2011
Paeonia obovata	Peony, Chinese	Farr and Rossman, 2011
Paeonia officinalis	Paeony, Common	Farr and Rossman, 2011
Paeonia peregrine		Farr and Rossman, 2011
Paeonia suffruticosa	Peony, Japenese Tree	Farr and Rossman, 2011
Paeonia taurica		Farr and Rossman, 2011
Paeonia tenuifolia		Farr and Rossman, 2011
Paeonia triternata		Farr and Rossman, 2011
Pedicularis spp.	Lousewort	CABI, 2011a
Pedicularis lapponica		Kaitera and Hiltunen, 2011
Pedicularis palustris	Lousewort, Marsh	Kaitera and Hiltunen, 2011
Pedicularis resupinata	Fan Gu Ma Xian Hao	Farr and Rossman, 2011
Pedicularis sceptrum-caroli-num	Moor-king	Farr and Rossman, 2011
Phtheirospermum japoni-cum	Song Hao	CABI, 2011a
Pinus brutia	Pine, Brutian	Raddi and Fagnani, 1978
Pinus clausa	Pine, Alabama	Raddi and Fagnani, 1978
Pinus contorta	Pine, Lodgepole	Raddi and Fagnani, 1978
Pinus densiflora	Pine, Japanese Umbrella	Farr and Rossman, 2011
Pinus echinata	Pine, Shortleaf	Raddi and Fagnani, 1978
Pinus elliottii	Pine, American Pitch	Raddi and Fagnani, 1978
Pinus glabra	Pine, Spruce	Raddi and Fagnani, 1978
Pinus griffithii	Pine, Himalayan	Farr and Rossman, 2011
Pinus halepensis	Pine, Aleppo	Raddi and Fagnani, 1978
Pinus koraiensis	Pine, Fruit	Farr and Rossman, 2011
Pinus massoniana	Pine, Masson	Farr and Rossman, 2011
Pinus montana	Pine, Dwarf Mountain	Farr and Rossman, 2011
Pinus mugo	Pine, Mountain	Raddi and Fagnani, 1978
Pinus mugo var. rostrata		Farr and Rossman, 2011
Pinus mugo var. rotundata		Farr and Rossman, 2011
Pinus nigra	Pine, Black	Raddi and Fagnani, 1978
Pinus pallasiana	Pine, Crymea	Farr and Rossman, 2011

Table 2-4 List of Reported Host Plants of *Cronartium flaccidum*

Scientific name	Common name	References
Pinus pinaster	Pine, Maritime	Raddi and Fagnani, 1978
Pinus pinea	Pine, Italian Stone	Raddi and Fagnani, 1978
Pinus ponderosa	Pine, Ponderosa	Raddi and Fagnani, 1978
Pinus pumila	Pine, Dwarf Siberian	Raddi and Fagnani, 1978
Pinus radiata	Pine, Monterey	Raddi and Fagnani, 1978
Pinus resinosa	Pine, Red	Raddi and Fagnani, 1978
Pinus rotundata		Farr and Rossman, 2011
Pinus serotina	Pine, Pond	Raddi and Fagnani, 1978
Pinus sylvestris	Pine, Scots	Ragazzi and Dellavalle Fedi, 1982
Pinus sylvestris var. mongolica		Farr and Rossman, 2011
Pinus tabuliformis	Pine, Chinese	Farr and Rossman, 2011
Pinus tabuliformis var. yunnanensis	Pine, Chinese Red	Farr and Rossman, 2011
Pinus taeda	Pine, Loblolly	Raddi and Fagnani, 1978
Pinus taiwanensis	Pine, Taiwan	Farr and Rossman, 2011
Pinus takahasii		CABI, 2011a
Pinus uncinata	Pine, Mountain	CABI, 2011a
Pinus virginiana	Pine, Virginia	Raddi and Fagnani, 1978
Pinus wallichiana	Pine, Bhutan	CABI, 2011a
Pinus yunnanensis	Pine, Yunnan	Farr and Rossman, 2011
Ruellia spp.		CABI, 2011a
Ruellia formosa	Wild Petunia	Farr and Rossman, 2011
Schizanthus spp.		CABI, 2011a
Siphonostegia chinensis	Yin Xing Cao	Zhuang and X., 2005
Tropaeolum spp.		CABI, 2011a
Tropaeolum majus	Nasturtium	Farr and Rossman, 2011
Tropaeolum minus	Nasturtium, Bush	Farr and Rossman, 2011
Verbena spp.		Robert et al., 2005
Verbena teucrioides	Verbena	Farr and Rossman, 2011
Vincetoxicum spp.	Angel-Pod	Farr and Rossman, 2011
Vincetoxicum albovianum		Farr and Rossman, 2011
Vincetoxicum amplexicaule	He Zhang Xiao	Farr and Rossman, 2011
Vincetoxicum fuscatum		Kaitera, 1999
Vincetoxicum hirundinaria	Swallowwort, White	Kaitera and Hiltunen, 2011
Vincetoxicum mongolicum	Hua Bei Bai Qian	Kaitera, 1999
Vincetoxicum nigrum	Swallowwort, Black	Kaitera, 1999
Vincetoxicum officinale	Swallowwort, White	Robert et al., 2005

Table 2-4 List of Reported Host Plants of *Cronartium flaccidum*

Scientific name	Common name	References
Vincetoxicum purpurascens		Farr and Rossman, 2011
Vincetoxicum rossicum	Swallowwort, European	CABI, 2011a

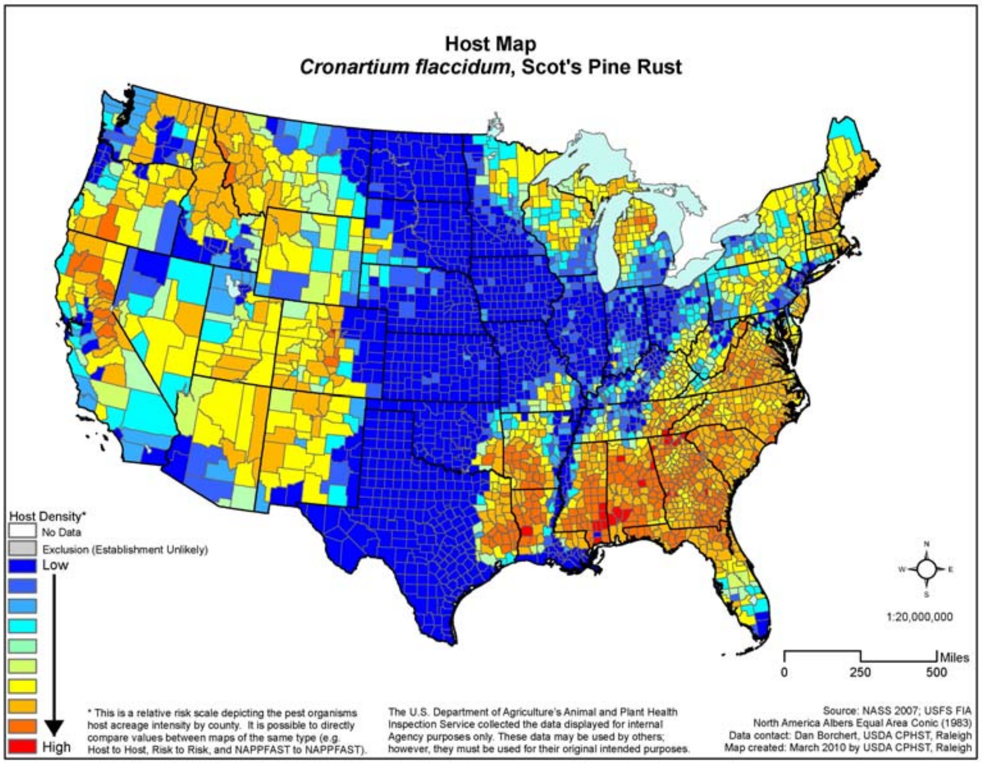

Figure 2-4 Host Map for Establishment Potential of *Cronartium flaccidum* Within the United States; Map Courtesy of USDA–APHIS–PPQ–CPHST (http://www.nappfast.org/)

Biology and Life cycle

Cronartium flaccidum is a heteroecious rust fungus completing different stages of its life cycle alternating between certain *Pinus* spp. and different angiosperm hosts including members of the genera *Paeonia, Asclepias, Delphinium, Impatiens*, *Myrica*, and *Tropaeolum*. In southern Europe, the main alternate host of *C. flaccidum* is *Vincetoxicum hirundinaria*, whereas *Melampyrum sylvaticum* is more common in northern Europe.

A rust fungus may produce as many as five distinct fruiting structures with five different spore stages in its life cycle in a definite sequence (*Table 2-5* on page

2-19). *Cronartium flaccidum* is macrocyclic and is known to produce all five spore stages. Like all rust fungi, *C. flaccidum* is an obligate parasite that requires living host cells to complete its life cycle. Genetic studies have shown that *C. flaccidum* is heteroecious and closely related to the autoecious rust *Peridermium pini* (Hantula et al., 2002). Autoecious refers to rust fungi that produce all stages of their life cycle on one species of host plant (in this case, pine); while heteroecious refers to rust fungi that require two unrelated host plants for completion of their life cycle (in this case, pine and another host).

Refer to *Table 2-5* on page 2-19 for a summary of each spore stage of *Cronartium flaccidum* and its role in the life cycle of the pathogen. *Cronartium flaccidum* infects hosts (*Pinus* spp.) by basidiospores (Stage IV) that are formed on leaves of alternate hosts and aerially dispersed (Ragazzi and Fedi, 1992). The basidia directly penetrate into the needle stomata to cause the initial infections on pine (Ragazzi and Fedi, 1992). Symptoms, however, only become apparent later in the branches and main stem (Geils et al., 2009). On pine shoots, spermagonia (Stage 0) and aecia (Stage 1) are developed, spreading the rust aerially among alternate hosts by aeciospores (Ragazzi et al., 1986b). A period of several years (2 to 4 years for the autoecious form but longer for heteroecious form) may elapse between infection and the appearance of the aecial state on infected tissue (Kaitera, 2000; Mordue and Gibson, 1978; Ragazzi and Moriondo, 1980). After successful disease establishment from aeciospores, uredinia (Stage III) are formed on alternate hosts, followed by telia (Stage IV) formation from uredinia or directly through the leaf epidermis (Kaitera and Nuorteva, 2003a; Ragazzi et al., 1987). After germination, basidia are formed on telia followed by basidiospore formation. The cycle then repeats by infection of the pine host through basidiospores. The pathogen survives as mycelium within host tissues.

Table 2-5 Five Spore Stages of *Cronartium flaccidum*

Stage	Description	Role
0	Spermagonia* bearing spermatia (n) and receptive hyphae (n)	Formed on pine; Sexual cycle of rust
I	Aecia bearing aeciospores (n+n)	Formed on pine; Infect alternative hosts
II	Uredinia (uredia) bearing urediniospores (uredospores) (n+n)	Formed on alternate host; Reinfects alternate hosts
III	Telia bearing teliospores (n+n → 2n)	Formed on alternate hosts
IV	Basidia bearing basidiospores (n)	Formed on alternate host; Cause of initial infections on pine

*Note: Spermagonia were formally known as pycnia and spermatia were formally known as pycniospores, and some references use the older nomenclature.

Several environmental factors influence the development of the disease and the life cycle of *Cronartium flaccidum*. Ragazzi et al. (1989) evaluated temperature, spore type, and host leaf age as variables in the production of uredia and telia of *C. flaccidum* on the alternate host *Vincetoxicum hirundinaria*. The authors found that 20°C (68°F) was optimal for the production of uredia and telia on host leaves (5 to 10 days old). The production of uredia was best, however, when urediniospores rather than aeciospores were used as inoculum. Ragazzi (1983) reported that the optimum temperature for formation of uredinia and telial columns was 20 to 22°C (68 to 72°F), and temperatures less than 18°C (64°F) or greater than 22°C (72°F) were detrimental to rust fructification.

The temperatures reported for germination of the different spore types are 5 to 30°C (41 to 86°F) for aeciospores, 5 to 30°C (41 to 86°F) for urediniospores, and 10 to 25°C (50 to 77°F) for basidiospores (Mordue and Gibson, 1978; Ragazzi et al., 1986a). The optimum temperature for germination of aeciospores, urediniospores, and basidiospores was reported as 15°C (59°F), 20°C (68°F), and 20°C, respectively (Ragazzi et al., 1986a). High moisture levels and precipitation increase the incidence of disease (CABI, 2010).

In addition, pathogenic variability in strains of *Cronartium flaccidum* have been observed. Differences in pathogenicity was correlated to different hosts and habitats with significant differences dependent on the *Pinus* spp. inoculated and the elevation from which *C. flaccidum* strains were obtained (Mittempergher and Raddi, 1977).

Insects may play a role in mating in *Cronartium flaccidum* based on the similarity of its life cycle to that of *C. ribicola* (Mordue and Gibson, 1978). Insects are attracted to the sweet liquid produced from the spermogonia of *Cronartium ribicola* and appear to promote fertilization by carrying spermatia between them.

Outbreaks of Scots pine blister rust are often associated with insect infestations (*Myleophilus piniperda*, *Bupalus piniaria*, *Pissodes notatus*), which aggravate the damage caused. Egg laying by *P. notatus* is localized on pines attacked by *Cronartium flaccidum* (Mordue and Gibson, 1978). Aeciospores have been shown to be transmitted by *Pissodes piniphilus* (Pappinen and Weissenberg, 1994). *Pissodes pini*, *Dioryctria splendidella*, *Laspeyresia coniferana*, *Lagria hirta*, and *Doryctria abietella* are reported as possible vectors for the rust on the basis of their occurrence and because they feed on *C. flaccidum* aecia (CABI, 2010).

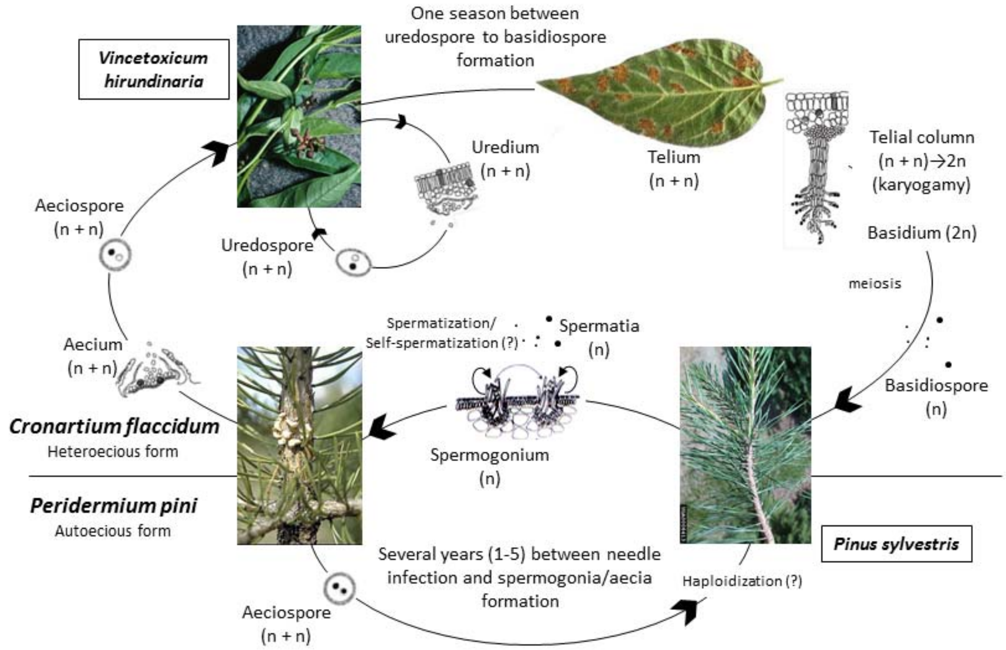

Figure 2-5 Disease Cycle of the Two Scots Pine Blister Rust Forms, *Cronartium flaccidum* (Heteroecious Form) and *Peridermium pini* (Autoecious Form)

Environmental Impact

Introduction of this pathogen could have some negative impacts on the environment. Plant hosts of the Scots pine blister rust may include several *Pedicularis* spp. which are listed as species of concern or endangered, (USFWS, 2011). Chemical control programs may be initiated in the event of an introduction of the *Cronartium flaccidum* in the United States, which may negatively impact non-target pests and the environment.

Table 2-6 List of Species of Concern and Endangered Potential Plant Hosts of *Cronartium flaccidum*

Scientific Name	Common Name	Listing Status
Pedicularis dudleyi	Dudley's lousewort	Species of Concern
Pedicularis furbishiae	Furbish lousewort	Endangered
Pedicularis semibarbata charlestonensis	No common name	Species of Concern
Gentiana pennelliana	Wiregrass gentian	Species of Concern
Gentiana plurisetosa	Klamath gentian	Species of Concern
Gentiana setigera	Mendocino gentian	Species of Concern

Table 2-6 List of Species of Concern and Endangered Potential Plant Hosts of *Cronartium flaccidum*

Scientific Name	Common Name	Listing Status
Cynanchum wigginsii	No common name	Species of Concern
Pinus contorta bolanderi	Bolander's beach pine	Species of Concern
Pinus radiata	Monterey pine	Species of Concern
Pinus torreyana insularis	Torrey Island pine	Species of Concern
Pinus torreyana torreyana	Torrey, Del Mar pine	Species of Concern

Endangered and Threatened Plants (50 CFR 17.12).
USFWS, 2011.

Chapter 3

Identification

Contents

Introduction

Use *Chapter 3 Identification* as a guide to recognizing Scots pine blister rust (*Cronartium flaccidum* and *Peridermium pini*). Accurate identification of the pathogen is pivotal to assessing its potential risk, developing a survey strategy, and determining the level and manner of control. Characteristic symptoms of the rusts on the host plants are not definitive and morphological and microscopic characteristics are necessary to identify *C. flaccidum* and *P. pini*.

Authorities

Qualified State, County, or cooperating university, personnel may perform preliminary identification and screening of suspect *Cronartium flaccidum* and *Peridermium pini*. Before survey and control activities are initiated in the United States, an authority recognized by USDA–APHIS–PPQ-National Identification Services must confirm the identity of such pathogens. Submit specimens to the USDA-National Identification Services (NIS). For further information refer to *How to Submit Plant Samples* on page C-1 and *Taxonomic Support for Surveys* on page D-1.

Reporting

Forward reports of positive identifications by national specialists to PPQ National Identification Service (NIS) in Riverdale, Maryland, according to Agency protocol. NIS will report the identification status of these tentative and confirmed records to PPQ-Emergency and Domestic Programs (EDP). EDP will report the results to all other appropriate parties. For further information refer to *Taxonomic Support for Surveys* on page D-1.

Symptoms

This section describes the plant symptoms that are characteristic of Scots pine blister rust.

Cronartium flaccidum causes blister rust in pines. The first symptoms of disease are yellowish, necrotic spots on the pine needles. Chlorosis and necrosis of the infection sites, yellowing and premature defoliation of leaves/ needles, branch death, bark discoloration, cankers (lesions) and deformed growth are also commonly observed symptoms of the disease (CABI, 2010). Resinosis (excessive resin exudation) can be seen in and on the lesions.

Cronartium flaccidum affects plants by growing within the vascular system and impeding nutrient and water uptake. Mycelia grow on young shoots. As the pathogen spreads within the host, it interferes with normal tree growth by killing the cambium and damaging vascular tissue. This damage results in the loss of conductive ability, premature leaf loss, and eventual death of the tree. The pathogen can girdle the tree and the part of the tree located above the girdle dies (Mordue and Gibson, 1978).

The disease may occur on pines of all ages. The development of disease is usually rapid and lethal to seedlings and young trees (Martinsson and Nilsson, 1987). Infection, which takes place primarily via needles, leads to swelling of young shoots and to production of blister-like structures in the cortex, which split to reveal masses of orange aeciospores (*Figure 3-1* on page 3-3). The time from infection to visible aeciospores can take several years. In England, the aeciospores are usually observed in early summer (Greig, 1987). Spermogonia with spermatial fluid (sweetish droplets) also occur on the infected bark. Uredinia and hair-like telia appear on the lower leaf surface of the alternate hosts in mid to late summer (*Figure 3-2* on page 3-3).

Figure 3-1 Aecia on Twig of *Pinus sylvestris* 7.5× (http://nt.ars-grin.gov/taxadescriptions/factsheets/index.cfm?thisapp=Cronartiumflaccidum)

Figure 3-2 Uredinia on Underside of Leaf of *Paeonia* sp. 10× (http://nt.ars-grin.gov/taxadescriptions/factsheets/index.cfm?thisapp=Cronartiumflaccidum)

Figure 3-3 Telia on Underside of Leaf of *Paeonia officinalis* 10× (http://nt.ars-grin.gov/taxadescriptions/factsheets/index.cfm?thisapp=Cronartiumflaccidum)

Figure 3-4 Telia on Underside of Leaf of *Vincetoxicum hirundinaria* 25× (http://nt.ars-grin.gov/taxadescriptions/factsheets/index.cfm?thisapp=Cronartiumflaccidum)

Morphological Descriptions

Spermagonia

Spreading beneath the periderm, flat, about 40 to 50 µm deep and 0.5 to 3 mm diameter, at first yellowish, exuding spermatia in orange droplets 1 to 2 µm, later darkening, gradually disrupted by enlarging aecia.

Aecia

Peridermioid, about 2 to 7 mm diam., dehiscence circumscissile or irregular. Peridium several cells thick, the cells rhomboid ellipsoid, elongated up to 80 µm long by 38 µm wide, the walls 4 to 8 µm thick, strongly verrucose (wart-like); rigid hair like peridial filaments are frequently present. Aeciospores are globose to ovoid-ellipsoid, 21 to 36 x 14 to 24 µm (mean 26 x 19 µm) with hyaline walls 2 to 4 µm thick; walls verrucose except for smooth area at base or side, the warts approx. 1 µm diam. and 1 to 2 µm high.

Uredinia

Hypophyllous (growing on underside of leaves), in groups or scattered, bullate (appearing puckered, blistered), 0.1 to 0.3 mm diam., peridiate (with protective layer enclosing spores), dehiscing (splitting open) by a central pore. Urediniospores broadly ellipsoid to obovoid, 18 to 30 x 11 to 20 µm (mean 24 x 15 µm), wall hyaline, 1.5 to 2.5 µm thick, echinulate (spiny) with the spines 2 to 4 µm apart and about 1 µm high, though some spores show almost smooth areas; germ pores inconspicuous.

Telia

Develop in the uredinia or separately, producing basally peridiate teliospore columns up to 2 mm long and 0.1 to 0.2 mm wide, pale orange to cinnamon brown, sometimes closely grouped on clearly defined spots, sometimes more scattered. Teliospores catenate (arranged in chains), firmly adherent, fairly short ellipsoid at apex of telial columns, longer and more cylindrical below, ends rounded or truncate, 20 to 64 x 10 to 16 µm (commonly about 55 x 12 µm), wall hyaline, yellowish to golden, about 1 µm thick, often thickened at ends or corners (particularly at apex of spore) to 2 to 3 µm, smooth. The teliospores germinate without dormancy and the upper part of the telial columns usually has a whitish powdery appearance due to the presence of basidia and basidiospores.

Basidia

Mature basidium septate with four cells, 33 to 40 µm long; each with a conical protuberance called sterigma, about 4 µm in length. Each sterigma has a basidiospore at the apex. In total there are four basidiospores for each basidium. Basidiospores rounded, smooth-surfaced, hyaline, 3 to 4 µm in diameter (Ragazzi et al., 1987). Basidiospores produce germ tubes that are often ramified. They vary in length (some more than 200 µm after 4 days of incubation) with a diameter of 2 to 3.5 µm (Ragazzi et al., 1987).

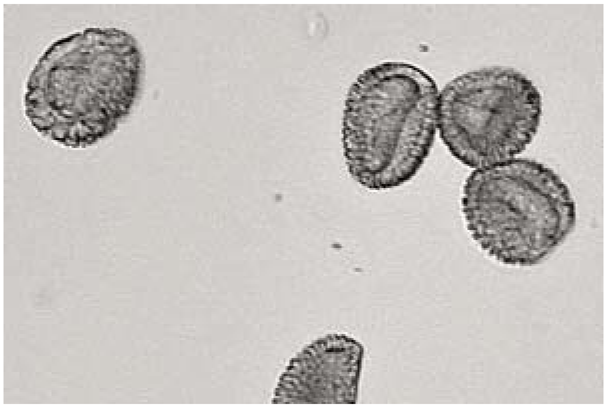

Figure 3-5 Aeciospores 400× (http://nt.ars-grin.gov/taxadescriptions/factsheets/index.cfm?thisapp=Cronartiumflaccidum)

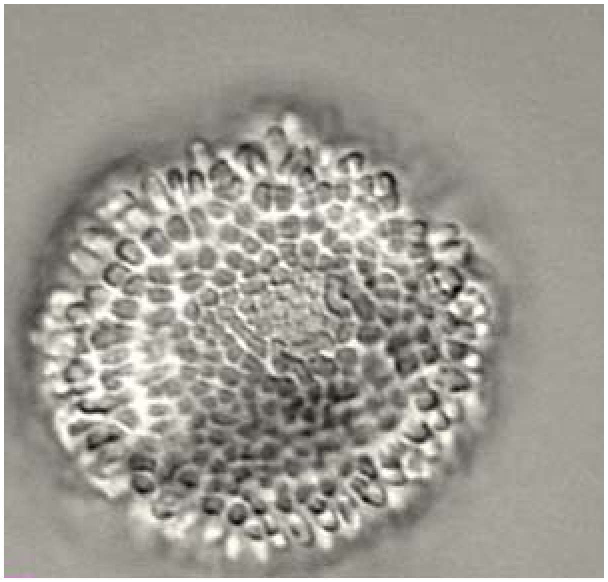

Figure 3-6 Aeciospore 1000× (http://nt.ars-grin.gov/taxadescriptions/factsheets/index.cfm?thisapp=Cronartiumflaccidum)

Figure 3-7 Aeciospore 1000× (http://nt.ars-grin.gov/taxadescriptions/factsheets/index.cfm?thisapp=Cronartiumflaccidum)

Figure 3-8 Urediniospores 400× (http://nt.ars-grin.gov/taxadescriptions/factsheets/index.cfm?thisapp=Cronartiumflaccidum)

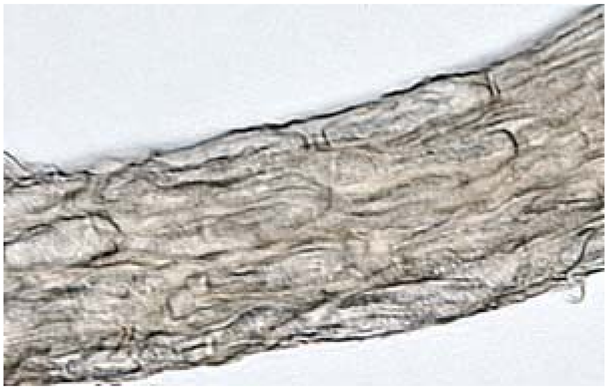

Figure 3-9 Urediniospores 1000× (http://nt.ars-grin.gov/taxadescriptions/factsheets/index.cfm?thisapp=Cronartiumflaccidum)

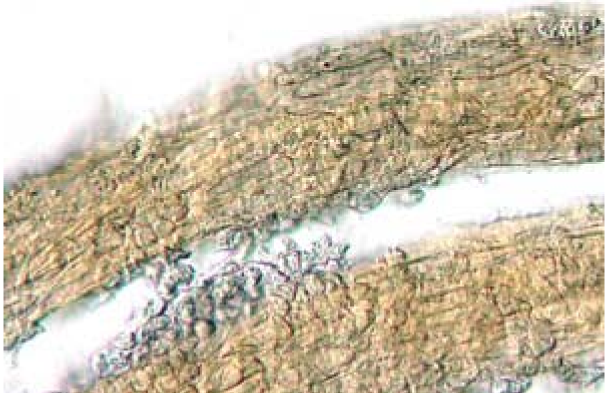

Figure 3-10 Teliospores in Telial Column 400× (http://nt.ars-grin.gov/taxadescriptions/factsheets/index.cfm?thisapp=Cronartiumflaccidum)

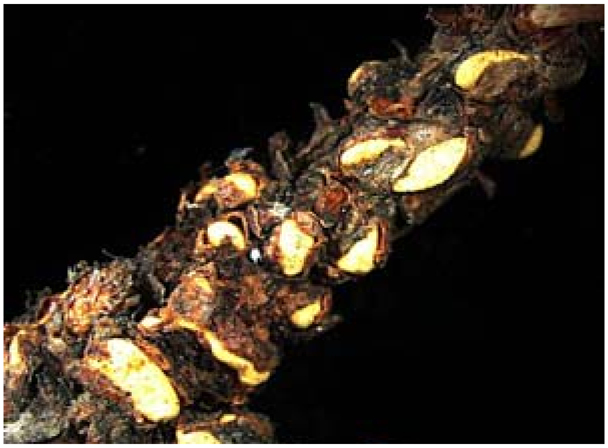

Figure 3-11 Teliospores in Telial Columns, with Basidiospores 200× (http://nt.ars-grin.gov/taxadescriptions/factsheets/index.cfm?thisapp=Cronartiumflaccidum)

Diagnostic Test

CAPS-Approved Method

Confirmation of *Cronartium flaccidum* requires a morphological identification. Characteristics of pycnia, aecia, aeciospores, uredinia, urediniospores, telia, and teliospores can be used to distinguish this species from other rust fungi (Mordue and Gibson, 1978).

Cronartium flaccidum can be cultured (axenically) by seeding aeciospores on modified Schenk and Hildebrandt's and Harvey and Grasham's media and incubating at 23 to 25°C (73 to 77°F) in the dark (Moricca and Ragazzi, 1994). Further study is possible *in vitro* on *Pinus* spp. callus tissue (Ragazzi et al., 1995).

Literature-Based Methods

The recovery plan for Scots pine blister rust suggests a morphological identification to genus and DNA sequencing to determine species (Geils et al., 2009).

Morphological

Cronartium flaccidum can be cultured by seeding aeciospores on modified Schenk and Hildebrandt's (1972) and modified Harvey and Grasham's (1974) media incubated at 21 to 24°C (70 to 75°F) (Moricca and Ragazzi, 1994, 1996). Incubation in the dark is suggested since the germ tubes of *C. flaccidum* are light sensitive. For *C. flaccidum*, the optimal seeding rate was found to be 400 to 1200 aecispores/mm^2 (Moricca and Ragazzi, 1994). Growth is slow and may take weeks to months to develop colonies. High variation was observed in hyphal length and morphology, and in colony appearance, margin, and morphology (Moricca and Ragazzi, 1994, 1996).

The modified Schenk and Hildebrandt’s medium (SH1) contains the following ingredients per liter: 300 mg $NH_4H_2PO_4$; 5 mg H_3BO_3; 151 mg $CaCl_2$; 0.100 mg $CoCl_2.6H_2O$; 0.200 mg $CuSO_4.5H_2O$; 20 mg Na_2.EDTA.$2H_2O$; 15 mg $FeSO_4.7H_2O$; 194.5 mg $MgSO_4$; 10 mg $MnSO_4$. H_2O; 1 mg KI; 2.5 g KNO_3; 0.100 mg Na_2MoO_4 .$2H_2O$; 1 mg $ZnSO_4.7H_2O$; 8 g Difco Bacto agar; 3 g oxoid broth; l g malt extract; 30 g sucrose; 2 mg kinetin, and 0.5 mg 2,4-dichlorophenoxyacetic acid (2,4-D) (Moricca and Ragazzi, 1994, 1996).

The modified Harvey and Grasham’s medium (HG1) contain the following ingredients per liter: 500 mg $CaNO_3.4H_2O$; 281.73 mg $MgSO_4.7H_2O$; 25 mg $(NH_4)2SO_4$; 250 mg $Fe_2(SO_4)_3.7H_2O$; 140 mg KH_2PO_4; 4.14 mg $MnSO_4.3H_2O$; 8 g Difco Bacto agar; 4 g oxoid broth; and 30 g sucrose

(Moricca and Ragazzi, 1994, 1996). The pH of both media was adjusted to 5.7 to 5.8 with 1N HCL and 1N NaOH before autoclaving at 121°C for 20 minutes (Moricca and Ragazzi, 1994). In general, isolates from Italy grew better at 21 than at 24°C and better on the HG1 medium than on the SH1 medium, but neither temperature nor medium significantly affected colony appearance and shape, sporulation, spore type, or hyphal type (Moricca and Ragazzi, 1996).

Moricca and Ragazzi (Moricca and Ragazzi, 2001) developed a technique to grow mycelial clones axenically of *Cronartium flaccidum* from basidiospores from single telia on HG1 medium containing 2 g/l of yeast extract, 0.5 g/l $CaCO_3$, and 10 g/l bovine serum albumin. Ragazzi et al. (1995) grew axenic cultures of *C. flaccidum* on pine callus tissue. The authors grew the pine calli on MS medium (Murashige and Skoog, 1962) supplemented 0.5 mg/l 2,4 D and used basidiospores to inoculate the callus tissue.

Biochemical

Cheng et al. (1995) were able to differentiate three *Cronartium* spp. (*C. ribicola*, *C. flaccidum*, and *C. quercum*) using isozyme analyses on the aeciospores.

Molecular

Kaitera and Hantula (1998) provide a protocol to compare restriction fragment length polymorphisms (RFLP) in ITS-region DNA based on digestion of PCR products with the restriction enzyme *Alu* I. This protocol was used to separate *Cronartium fraxinea* and *C. ribicola* telia from alternate hosts and to confirm aecia collected from Scots pine. *Cronartium ribicola* showed two bands with apparent sizes of 220 bp and 450 bp, *C. fraxinea* showed three bands with apparent sizes of 130 bp, 230 bp, and 350 bp. The 220 and 230 bp bands appeared to be twice as intense as the other bands, and assuming these two represent double restriction fragments, the summed fragment sizes of the two patterns were 890 and 940 bp, indicating the digestions were complete.

To distinguish alternating from non-alternating isolates of pine blister rust, PCR-amplified fragments of two regions of rDNA (ITS2 and IGS1) can be used in RFLP and SSCP analysis (Moricca and Ragazzi, 1998). Sequences for several regions of rDNA, particularly those for the 5.8s rRNA examined by Moricca et al. (1996), are available in GenBank for comparison (NCBI, 2011). A study from Samil et al. (2011), found that PCR–RFLP analysis of the IGS1 region using restriction enzyme *Hinf*I produced identical banding pattern between samples of *Cronartium flaccidum* and *Peridermium pini* and could not discriminate between the two rust forms as previously shown by Moricca and Ragazzi (1998). Samil et al. (2011) developed a set of microsatellite markers (*Table 2-6* on page 2-21) that could be used for detection and genetic variation studies both within and between lesions of Scots pine blister rust.

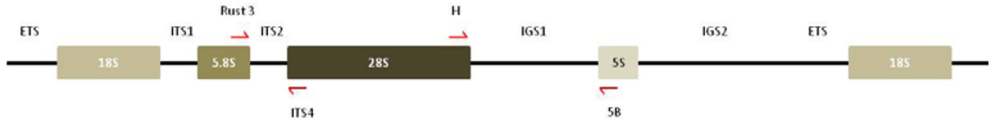

Figure 3-12 Schematic Diagram of rDNA in *Cronartium flaccidum* and *Peridermium pini*; Transcription Unit With Location of ITS and IGS Regions; adapted from Moricca and Ragazzi 1998

Table 3-1 List of PCR Primers for Detection of *Cronartium flaccidum* and *Peridermium pini*

Name	Oligonucleotide sequence (5'-3')	Product Size (bp)	Reference
Rust3	ACATCGAT-GAAGAACACAGT		Moricca et al., 1996
ITS4	TCCTCCGCTTATT-GATATGC	270	White et al., 1990
H	CCTCGATGTCG-GCTCTTC		Buchko and Klas-sen, 1990
5B	AGGATTCCCGC-GTGGTCCCC	1,300	Moricca and Raga-zzi, 1998
Pp1-F	ATTACCTCTCTT-GATGACAA		Samils et al., 2011
Pp1-R	GTACCAGGC-CAAAGG	153-195	Kasanen et al., 2000
Pp2-F	CTGGGTCAAGT-CAAATCTCC		Kasanen et al., 2000
Pp2-R	GGACCAAATTC-GATCATAGG	260-300	Samils et al., 2011
CqfSI_AAC27-F	TGGTGAGATATAG-ATAAGCATCAAGC		Burdine et al., 2007
CqfSI_AAC27-R	ACAAAGGGAGGA-CACATTGG	134-137	Burdine et al., 2007
CqfSI_AAC30-F	ATTTTCGCACGAA-CAGAGC		Burdine et al., 2007
CqfSI_AAC30-R	ATGTAAGTACT-GCCGGTGGC	341-515	Burdine et al., 2007
CqfSI_AAG13-F	AGCAGCA-CAAGCTGAGAATG		Burdine et al., 2007
CqfSI_AAG13-R	CGTTCTCATCC-GAATCCATC	104-107	Burdine et al., 2007
CqfSI_AAG18-F	AGTTTTCTTGGGT-GGTGGTG		Burdine et al., 2007
CqfSI_AAG18-R	ACCATCAGGTG-GTCAAGGAG	276-282	Burdine et al., 2007

Table 3-1 List of PCR Primers for Detection of *Cronartium flaccidum* and *Peridermium pini*

Name	Oligonucleotide sequence (5'-3')	Product Size (bp)	Reference
CqfSI_GATA06-F	ATCGAGAAC-GAGAGCGAGAG		Burdine et al., 2007
CqfSI_GATA06-R	AGAACAGATTG-GCATGAGCC	213-233	Burdine et al., 2007

Similar Species

At least eleven *Cronartium* species and six species of *Peridermium* occur in North America on pine (Chalkley, 2010). To a certain extent, these can be distinguished by the aeciospore and urediniospore morphology, as well as by symptomatology. While some cause stem cankers, other rusts produce galls or witches brooms in infected stems or branches. Others cause no symptoms at all (Chalkley, 2010). *Cronartium flaccidum* belongs to a distinct group of *Cronartium* species distinguished by their aeciospores (in which an echinulate surface alternates with smooth areas) (Moricca and Ragazzi, 1996). *Cronartium comandrae*, a widespread North American pine stem rust, that like *C. flaccidum* also infects two-needle species, produces unique tear-drop shaped aeciospores on pine (Chalkley, 2010).

Symptoms can be confused with those of *Cronartium ribicola*, the causal agent of white pine blister rust. *Cronartium ribicola* does not infect *Pinus sylvestris*, and *C. flaccidum* does not infect five-needle pines or *Ribes* species (Kaitera and Nuorteva, 2006b). Kaitera and Nuorteva (2006a) conducted inoculation studies with *C. ribicola* on the main alternate hosts of *C. flaccidum*. The authors found that neither uredinia nor telia developed on the leaves of *Vincetoxicum hirundinaria*, *V. nigrum*, *Melampyrum sylvaticum*, *M. pretense*, *M. nemorosum*, *M. arvense*, *M. cristatum*, or *M. polonicum*.

In Europe, other rusts that can attack pines also have a heteroecious life cycle similar to *Cronartium flaccidum*, but usually infect different alternate hosts. *Coleosporium tussilaginis*, the pine needle rust, shares a few telial hosts with blister rust, but produces its spermagonia and aecia on pine needles, not on the stems. Also, teliospores of this rust on species of *Melampyrum* are single to cylindrical, produced not in long columns but in waxy crusts. *Melampsora populnea* infects the shoots of two-needle pines, causing shoot bending and/or tip death. Its linear aecia lack a peridium and the aeciospores are significantly smaller than those of *C. flaccidum* (Chalkley, 2010).

Chapter

4

Survey Procedures

Contents

Introduction

Use *Chapter 4 Survey Procedures* as a guide when conducting a survey for Scots pine blister rust (*Cronartium flaccidum* and *Peridermium pini*) in potentially infected *Pinus* spp. hosts or when monitoring other natural areas where its alternate host plants may be present. After a new detection in the United States, or when detection in a new area is confirmed, conduct a delimiting survey to define the geographic location where diseased plants are present. Conduct a monitoring survey if you have applied a control procedure and need to measure its effectiveness.

Survey Types

Plant regulatory officials will conduct detection, delimiting, and monitoring surveys for *Cronartium flaccidum*. Conduct a detection survey to ascertain the presence or absence of *C. flaccidum* in an area where it is **not** known to occur. After a new U.S. detection, or when detection in a new area is confirmed, conduct a delimiting survey to define the extent of potentially infected plants in the U.S. Territories. Conduct a monitoring survey to determine the success of control or mitigation activities conducted against the pathogen.

Preparation, Sanitization, and Clean-Up

This section provides information that will help personnel prepare to conduct a survey; procedures to follow during a survey; and instructions for proper cleaning and sanitizing of supplies and equipment after the survey is finished.

1. Before starting a survey, determine if there have been recent pesticide applications that would make it unsafe to inspect the vineyards, rootstock nursery, or landscape planting. Contact the property owner or manager and ask if there is a re-entry period in effect due to pesticide application. Look for posted signs indicating recent pesticide applications, particularly in commercial fields or greenhouses.
2. Conduct the survey at the proper time. The aecia of *Cronartium flaccidum* appear on pine bark in early summer and other symptoms occur in the alternate hosts in mid- to late- summer (CABI, 2011a). Based upon the pests' reported global distribution, scientists believe *Cronartium flaccidum* and *Peridermium pini* could establish in the northeastern United States and portions of Washington State. General survey should focus on months when host plants are easily accessible and during active growing phases.
3. Obtain permission from the landowner before entering a property.
4. Determine if quarantines for other pests, or other crops, are in effect for the area being surveyed. Comply with any and all quarantine requirements.
5. When visiting the area to conduct surveys or to take samples, everyone must take strict measures to prevent contamination by *Cronartium flaccidum* and *Peridermium pini* or other pests between properties during inspections.

Before entering a new property, make certain that clothing and footwear are clean and free of pests and soil to avoid moving soilborne pests and arthropods from one property to another.

Wash hands with an approved antimicrobial soap. If not using an antimicrobial soap, wash hands with regular soap and warm water to remove soil and debris. Then use an alcohol-based antimicrobial lotion, with an equivalent of 63 percent ethyl alcohol. If hands are free of soil or dirt, the lotion can be applied without washing. Unlike some antimicrobial soaps, antimicrobial lotions are less likely to irritate the hands and thereby improve compliance with hand hygiene recommendations.

6. Gather together all supplies. Confirm the equipment and tools are clean. When taking plant samples, disinfest tools with bleach to avoid spreading diseases or other pests. A brief spray or immersion of the cutting portion of the tool in a 5 percent solution of sodium hypochlorite (bleach) is an effective way to inactivate bacterial and other diseases and prevent their spread.
7. Mark the plant, tree or sampled location with flagging whenever possible, and draw a map of the immediate area and indicate reference points so that the areas can be found in the future if necessary. Do not rely totally on the flagging or other markers to re-locate a site as they may be removed. Record the GPS coordinates for each trap or infected tree location so that the area or plant may be re-sampled if necessary.
8. Survey task forces should consist of an experienced survey specialist or pathologist familiar with *Cronartium flaccidum* and *Peridermium pini* and the symptoms of their damage.

Detection Survey

The purpose of a detection survey is to determine that a pest is present in a defined area. This can be broad in scope, as when assessing the presence of the pest over large areas or it may be restricted to determining if a specific pest is present in a focused area.

Statistically, a detection survey is not a valid tool to claim that a pest does not exist in an area, even if results are negative. Negative results can be used to provide clues about mode of dispersal, temporal occurrence, or industry practices. Negative results are also important when compared with results from sites that are topographically, spatially, or geographically similar.

Procedure

Follow this procedure when conducting a detection survey for *Cronartium flaccidum* and *Peridermium pini*.

1. Use visual inspection to examine the cultivated host plants for symptoms. Refer to *Visual Inspection for Detection Survey* on page 4-7 for further information on inspection procedures.

Important: Detection surveys for pine tree infected by *Cronartium* flaccidum and *Peridermium pini* should be conducted by State inspectors in conjunction with Federal PPQ inspectors.

2. To confirm disease, collect plants showing typical symptoms. Place samples in plastic bags. Keep samples cool. Double bag the samples and deliver promptly to a diagnostic laboratory.

The CAPS-approved survey method for *Cronartium flaccidum* and *Peridermium pini* is based on visual survey, spore trapping, or a combination of these methods. For visual survey, collect twigs, bark, or leaves from symptomatic plants with signs (fruiting bodies) of the pathogen. Spore traps, similar to those used for soybean rust monitoring, can be used to detect spores.

Literature-Based Methods

Visually examine two-needle pines, especially Scots pine, for fruiting bodies (spermagonia and aecia) of the pathogen. Alternate hosts can also be examined for uredinia and telia of the pathogen. *Cronartium flaccidum* can be detected in the tree most easily when fruiting. Spermogonia with spermatial fluid occur on the infected bark (next to the aecial scars of early summer) in late summer; aecia appear on the bark in the early summer, and uredinia and hair-like telia appear on the lower leaf surface of the alternate hosts in mid-to-late summer. The infected part of the shoot (lesion) is often swollen. The disease is also revealed by resinosis in the lesion. After the leader of the shoot carrying the lesion is killed, the top of the tree is dead, but green shoots below the lesion are visible. As an indication of infection in the shoot, the color of the needles above the lesion may turn light green to yellow (CABI, 2010).

Delimiting Survey after Initial U.S. Detection

If *Cronartium flaccidum* and *Peridermium pini* are detected in the United States, surveys will be conducted in the area to determine the distribution of the infected plants. In large areas, locating the actual source of an infestation could be difficult depending on season, age of infected plants and time elapsed from the initial infection.

Procedure

Follow the same procedure used for *Detection Survey* on page 4-3. Once *Cronartium flaccidum* and *Peridermium pini* have been confirmed, surveys should be most intensive around the known positive detections and any discovered through traceback and trace-forward investigations.

Traceback and Trace-Forward Investigations

Traceback and trace-forward investigations help determine priorities for delimiting survey activities after an initial U.S. detection. Traceback investigations attempt to determine the source of infection. Trace-forward investigations attempt to define further potential dissemination through means of natural and artificial spread (commercial or private distribution of infected plant material). Once a positive detection is confirmed, investigations are conducted to determine the extent of the infestation or suspect areas in which to conduct further investigations.

Due to the risk of *Cronartium flaccidum* and *Peridermium pini* spreading through infected plants, USDA–APHIS–PPQ has prohibited the importation of plants for planting of the listed host genera, with the exception of seed, until a pest risk analysis has been completed and appropriate effective mitigation measures have been established. However, *C. flaccidum* and *P. pini* may enter through the illegal importation of nursery stock.

Homeowner Properties

For positive detections on homeowner properties, ask the owner of the infected material to determine where it originated (nursery, neighbors, etc.) and where it might have been further distributed.

Nursery Properties

For nursery hosts, a list of facilities associated with infected nursery stock from those testing positive for *Cronartium flaccidum* and *Peridermium pini* will be compiled. These lists will be distributed by the State to the field offices, and are **not** to be shared with individuals outside USDA–APHIS–PPQ regulatory cooperators. Grower names and field locations on these lists are strictly confidential, and any distribution of lists beyond appropriate regulatory agency contacts is prohibited.

Each State is only authorized to see locations within their State and sharing of confidential business information may be restricted between State and Federal entities. Check the privacy laws with the State Plant Health Director for the State.

When notifying growers on the list, be sure to identify yourself as a USDA or State regulatory official conducting an investigation of facilities that may have received *Cronartium flaccidum* and *Peridermium pini* -infected material. Speak to the growers or farm managers and obtain proper permission before entering private property.

Several actions need to occur immediately upon confirmation that a nursery sample is positive for either *Cronartium flaccidum* or *Peridermium pini*:

- Check nursery records to obtain names and addresses for all sales or distribution sites (if any sales or distribution has occurred from infested nursery during the previous 6 months).
- Evaluate the disease situation, including identification and inspection of the budwood source(s) of the diseased tree(s), the location within the nursery, and the disease severity.

Refer to *Regulatory Procedures* on page 5-1 and *Control Procedures* on page 6-1 for more information.

Monitoring Survey

Conduct a monitoring survey if you have applied a control procedure and need to measure its effectiveness. If *Cronartium flaccidum* and *Peridermium pini* are detected in the United States, CPHST personnel will assemble a technical working group to provide guidance on using a monitoring survey to measure the effectiveness of applied treatments on the pathogen population. Refer to *Control Procedures* on page 6-1 for further information on control options.

Procedure

Once *Cronartium flaccidum* and *Peridermium pini* have been confirmed from a particular field, and infected and potentially infected plants have been destroyed, additional monitoring will be necessary. Use the following tools:

- Visual inspection in the field
- Collection of samples from potential weed hosts for several years and multiple times per season

Refer to *Visual Inspection for Detection Survey* on page 4-7 and *Visual Inspection for Delimiting Survey* on page 4-7 for further information concerning the inspection of host plants.

Visual Inspection for Detection Survey

Use visual inspection as a tool when surveying for Scots pine blister rust (*Cronartium flaccidum* and *Peridermium pini*) in forest, nursery and Christmas tree production areas.

Conduct a visual inspection in a field by looking for plants with typical pine blister symptoms. The absence of symptoms, however, does not necessarily mean *Cronartium flaccidum* and *Peridermium pini* are not present in the area inspected. Some infected plants may not express symptoms, depending on the severity of the infection and in particular, less sensitive, pine species.

Visual Inspection for Delimiting Survey

Construct delimiting surveys in an area—based on known positive testing, associated positive testing, or potentially infested areas to define the geographic location of the pathogen population. However, it may be necessary to do random samples in a general growing area to detect new infestations not discovered through investigations. The delimiting survey in a general growing area can include random sampling of wild and cultivated host species throughout a geographical area, with more intensive sampling near known infestations. As the distance away from the epicenter of a known infestation increases, decrease the rate of random sampling. Based on the epidemiology and grower practices, an evaluation of risk and resources available will help determine the extent of these random sampling surveys.

Sentinel Sites

Sentinel sites are locations that are regularly inspected along the surveyor's normal route. The sites can be established using a known host plant. The plant used as a sentinel site should be inspected for visual signs of damage; if available, test the host plant. Use GPS to record the location of the host plant, and draw a map of the immediate area that includes reference points so that the area can be found by others if necessary. Once the sentinel site is established the surveyor should re-inspect the site on a regular basis (bimonthly or monthly) as permitted by the regular survey schedule. GIS can be use to map the sentinel site locations to help visualize an even coverage, particularly high risk areas.

Other Diseases

Other diseases can cause symptoms that are similar, so diagnostic tests must be performed on samples from symptomatic plants in order to confirm the presence of *Cronartium flaccidum*. See *Identification* on page 3-1 for more information.

Targeted Surveys

Conduct targeted surveys at nurseries associated with high risk pathways. Areas with regular traffic from countries with known infestations should also be targeted for regular surveys.

Procedure

A defined method is unavailable.

Survey Records

Records should be kept for each survey site. Negative survey data must be recorded even in the absence of *Cronartium flaccidum* and *Peridermium pini*. Record also the absence of samples at surveyed sites. Survey records and data recording formats should be consistent, to allow for standardized collection of information.

If automated field collection devices are used, such as the Integrated Survey Information System (ISIS), ensure that all surveyors are trained in the technology before beginning the survey. Use the appropriate ISIS templates for this pathogen. To reduce the burden on field data collectors, enter any known contact or address information into the database and hand-held data recorders before working in the field. At the end of the survey, all survey data should be entered into a designated State or national pest database.

Data Collection

Surveyors visiting sites to place holds or take samples should collect the following information:

- Date of collection or observations
- Collector's name

- Grower's field identification numbers
- GPS coordinates
- Variety of host plants grown
- History of farm machinery usage
- Observations of symptoms
- Other relevant information

In the absence of inspection officials, take the following actions immediately if yellowing symptoms are noticed:

1. Mark the location
2. Remove the plants and flag the location in the field
3. Notify the State or PPQ inspector
4. Place the whole plant inside two resealable plastic bags
5. Label the sealed bags with the following information:
6. Date
7. Name of person responsible
8. Location of sample collection
9. Keep bagged plants cool or refrigerated until the inspector arrives
10. Do not freeze the sample

Cooperation with Other Surveys

Other surveyors regularly sent to the field should be trained to recognize outbreaks that could be associated with *Cronartium flaccidum* and *Peridermium pini.*

Chapter 5

Regulatory Procedures

Contents

Introduction

Use *Chapter 5 Regulatory Procedures* as a guide to the procedures that must be followed by regulatory personnel when conducting pathogen survey and control programs against Scots pine blister rust (*Cronartium flaccidum* and *Peridermium pini*).

Instructions to Officials

Agricultural officials must follow instructions for regulatory treatments or other procedures when authorizing the movement of regulated articles. Understanding the instructions and procedures is essential when explaining procedures to people interested in moving articles affected by the quarantine and regulations. Only authorized treatments can be used in line with labeling restrictions. During all field visits, ensure that proper sanitation procedures are followed as outlined in *Preparation, Sanitization, and Clean-Up* on page 4-2.

Regulatory Actions and Authorities

After an initial suspect positive detection, an Emergency Action Notification may be issued to hold articles or facilities, pending positive identification by a USDA–APHIS–PPQ-recognized authority and/or further instruction from the PPQ Deputy Administrator. If necessary, the Deputy Administrator will issue a letter directing PPQ field offices to initiate specific emergency action under the Plant Protection Act until emergency regulations can be published in the *Federal Register.*

The Plant Protection Act of 2000 (Statute 7 USC 7701-7758) provides the authority for emergency quarantine action. This provision is for interstate regulatory action only; intrastate regulatory action is provided under State authority.

State departments of agriculture normally work in conjunction with Federal actions by issuing their own parallel hold orders and quarantines for intrastate movement. However, if the U.S. Secretary of Agriculture determines that an extraordinary emergency exists and that the States measures are inadequate, USDA can take intrastate regulatory action provided that the governor of the State has been consulted and a notice has been published in the Federal Register. If intrastate action cannot or will not be taken by a State, PPQ may find it necessary to quarantine an entire State.

PPQ works in conjunction with State departments of agriculture to conduct surveys, enforce regulations, and take control actions. PPQ employees must have permission of the property owner before entering private property. Under certain situations during a declared extraordinary emergency or if a warrant is obtained, PPQ can enter private property without owner permission. PPQ prefers to work with the State to facilitate access when permission is denied, however each State government has varying authorities regarding entering private property.

A General Memorandum of Understanding (MOU) exists between PPQ and each State that specifies various areas where PPQ and the State department of agriculture cooperate. For clarification, check with your State Plant Health Director (SPHD) or State Plant Regulatory Official (SPRO) in the affected State. Refer to *Resources* on page A-1 for information on identifying SPHD’s and SPRO’s.

Tribal Governments

USDA–APHIS–PPQ also works with federally-recognized Indian Tribes to conduct surveys, enforce regulations and take control actions. Each Tribe stands as a separate governmental entity (sovereign nation) with powers and authorities similar to State governments. Permission is required to enter and access Tribal lands.

Executive Order 13175, Consultation and Coordination with Indian and Tribal Governments, states that agencies must consult with Indian Tribal governments about actions that may have substantial direct effects on Tribes. Whether an action is substantial and direct is determined by the Tribes. Effects are not limited to Tribal land boundaries (reservations) and may include effects on off-reservation land or resources which Tribes customarily use or even effects on historic or sacred sites in States where Tribes no longer exist.

Consultation is a specialized form of communication and coordination between the Federal and Tribal governments. Consultation must be conducted early in the development of a regulatory action to ensure that Tribes have opportunity to identify resources which may be affected by the action and to recommend the best ways to take actions on Tribal lands or affecting Tribal resources. Communication with Tribal leadership follows special communication protocols. For more information, contact PPQ's Tribal Liaison. Refer to *Table A-1* on page A-2 for information on identifying PPQ's Tribal Liaison.

To determine if there are federally-recognized Tribes in a State, contact the State Plant Health Director (SPHD). To determine if there are sacred or historic sites in an area, contact the State Historic Preservation Officer (SHPO). For clarification, check with your SPHD or State Plant Regulatory Official (SPRO) in the affected State. Refer to *Resources* on page A-1 for contact information.

Overview of Regulatory Program After Detection

Once an initial U.S. detection is confirmed, holds will be placed on the property by the issuance of an Emergency Action Notification. Immediately put a hold on the property to prevent the removal of any host plants of the pest.

Traceback and trace-forward investigations from the property will determine the need for subsequent holds for testing and/or further regulatory actions. Further delimiting surveys and testing will identify positive properties requiring holds and regulatory measures.

Record-Keeping

Record-keeping and documentation are important for any holds and subsequent actions taken. Rely on receipts, shipping records and information provided by the owners, researchers or manager for information on destination of shipped plant material, movement of plant material within the facility, and any management (cultural or sanitation) practices employed.

Keep a detailed account of the numbers and types of plants held, destroyed, and/or requiring treatments in control actions. Consult a master list of properties, distributed with the lists of suspect nurseries based on traceback and trace-forward investigations, or nurseries within a quarantine area. Draw maps of the facility layout to located suspect plants, and/or other potentially infected areas. When appropriate, take photographs of the symptoms, property layout, and document plant propagation methods, labeling, and any other information that may be useful for further investigations and analysis.

Keep all written records filed with the Emergency Action Notification copies, including copies of sample submission forms, documentation of control activities, and related State issued documents if available.

Issuing an Emergency Action Notification

Issue an Emergency Action Notification to hold all host plant material at facilities that have the suspected plant material directly or indirectly connected to positive confirmations. Once an investigation determines the plant material is not infested, or testing determines there is no risk, the material may be released and the release documented on the EAN.

Regulated Area Requirements Under Regulatory Control

Depending upon decisions made by Federal and State regulatory officials in consultation with a Technical Working Group, quarantine areas may have certain other requirements for commercial or research fields in that area, such as plant removal and destruction, cultural control measures, or plant waste material disposal.

Any regulatory treatments used to control this pest or herbicides used to treat plants will be labeled for that use or exemptions will be in place to allow the use of other materials.

Establishing a Federal Regulatory Area or Action

Regulatory actions undertaken using Emergency Action Notifications continue to be in effect until the prescribed action is carried out and documented by regulatory officials. These may be short-term destruction or disinfestation orders or longer term requirements for growers that include prohibiting the planting of host crops for a period of time. Over the long term, producers, shippers, and processors may be placed under compliance agreements and permits issued to move regulated articles out of a quarantine area or property under an EAN.

Results analyzed from investigations, testing, and risk assessment will determine the area to be designated for a Federal and parallel State regulatory action. Risk factors will take into account positive testing, positive associated, and potentially infested exposed plants. Boundaries drawn may include a buffer area determined based on risk factors and epidemiology.

Regulatory Records

Maintain standardized regulatory records and databases in sufficient detail to carry out an effective, efficient, and responsible regulatory program.

Use of Chemicals

The PPQ *Treatment Manual* and the guidelines identify the authorized chemicals, and describe the methods and rates of application, and any special instructions. For further information refer to *Control Procedures* on page 6-1. Agreement by PPQ is necessary before using any chemical or procedure for regulatory purposes. No chemical can be recommended that is not specifically labeled for this pest.

Chapter 6

Control Procedures

Contents

Introduction

Use *Chapter 6 Control Procedures* as a guide to control an outbreak of Scots pine blister rust (*Cronartium flaccidum* and *Peridermium pini*) in the United States and collaborating territories. Consider the treatment options described within this chapter when taking action to eradicate, manage or contain an infestation by *Cronartium flaccidum* and *Peridermium pini*.

The control of these pathogens is generally obtained implementing a combination of strategies including removal and destruction of infected plants, cultural and management control, chemical treatments and use of genetic resistance.

Overview of Emergency Programs

Plant Protection and Quarantine develops and makes control measures available to involved States. United States Environmental Protection Agency-approved treatments will be recommended when available. If the selected treatments are not labeled for use against the pest or in a particular environment, PPQ's FIFRA Coordinator is available to explore the appropriateness in developing an Emergency Exemption under Section 18, or a State Special Local Need under section 24(c) of FIFRA (Federal Insecticide, Fungicide, and Rodenticide Act), as amended.

The PPQ FIFRA Coordinator is also available upon request to work with EPA to rush the approval of a product that may not be registered in the United States, or to get labeling for a new use. The PPQ FIFRA Coordinator is available for guidance pertaining to pesticide use and registration. Refer to *Resources* on page A-1 for information on contacting the Coordinator.

Treatment Options

Consider the treatment options described within this chapter when taking action to eradicate or control Scots pine blister rust (*Cronartium flaccidum* and *Peridermium pini*). Treatments may include the following:

- *Prevention* on page 6-4
- *Eradication* on page 6-4
- *Cultural Control and Sanitary Measures* on page 6-4
- *Chemical Control* on page 6-4
- *Biological Control* on page 6-6
- *Host Resistance* on page 6-6

Environmental Documentation and Monitoring

Obtain all required environmental documentation before beginning. For further information, refer to *Environmental Compliance* on page 7-1. Contact Environmental Services staff for the most recent documentation. Refer to *Resources* on page A-1 for contact information.

Efficacy of Treatment

Eradication measures should be continued for several years to ensure that populations of exotic *Cronartium flaccidum* and *Peridermium pini* have been eliminated. Once the pathogen has been eradicated, monitoring of the site should be continued for 1 to 2 years. For further information, refer to *Monitoring Survey* on page 4-6.

Site Assessment

When visiting a site keep a log of observations, flag the infested areas, and record the coordinates. Record also the name of the property owner. Some of this information may have been recorded during the survey. Communicate frequently with the person responsible for the site.

Classification

Information on the type of property needs to be recorded to help develop a control plan. Site access, security, containment, and ownership type may dictate a particular direction in control options. Prepare a concise overview of the infested area. Record information about the infested property, including the following:

- Location
- Type of property ownership (government, private, Tribal, commercial, residential, or agricultural
- Current and past users of the property
- Distribution of infected plants
- Status of security and containment
- Modes of artificial movement

Prevention

A preventive measure should include restrictions on importation of primary or alternate host plants and plant material from areas where this rust disease is known to occur. The importation of host plant seed would represent virtually no risk for this pathogen (Geils et al., 2009). Also, given the possibility of latent infections in *Pinus*, phytosanitary post-entry quarantine of any imported plants is necessary (USDA–APHIS, 2008). Accidental introduction would also likely be prevented by controlling bark-bearing wood in shipping materials from areas where the rust occurs (see Canadian Food Inspection Agency, 2008; MAF, 2009).

Eradication

Butin (1995) recommends removal of infected branches or trees in stands where the disease is already present. Pruning of infected branches may or may not prevent development of additional cankers on the trunk, although the general purpose of removing inoculum is achieved (Moricca and Ragazzi, 2008).

Cultural Control and Sanitary Measures

Because basidiospores are disseminated only over a short distance (Hunt, 1997), removal of the primary hosts from the vicinity of limited plantings of pine is a measure that can reduce infection by the heteroecious *Cronartium flaccidum* (Butin, 1995; Mordue and Gibson, 1978). In Italy, the alternate host is too common for this effort to be effective or worthwhile, however, development of a hazard map of the known distribution of the alternate host allows for planting of susceptible pines away from sources of inoculum (Moricca and Ragazzi, 2008).

Chemical Control

Use of fungicides may be practical for rust control on plantation, nursery and garden trees, but is impractical in forests (Moricca and Ragazzi, 2008).

Triadimefon has been demonstrated (Pitt et al., 2006) to be a useful prophylactic for white pine blister rust (*Cronartium ribicola*). Triadimefon is registered in the United States for use on Christmas trees. Yao and Peixin

(1991) report that application of thiophanate-methyl and triadimefon to a canker surface was effective in eliminating aecial sporulation of Scots pine blister rust in China. Salt spray can kill alternate hosts, but eradication was only practical in proximity to a pine plantation. Maloy (1997) reviewed the history of control of *C. ribicola* in the United States, including aborted efforts with antibiotics (phytoactin and cycloheximide) and herbicides. Pappinen and von Weissenburg (1996) describe the role of the pine-top weevil wounding pine twigs and increasing rust infection. We are not aware of any studies on the effective use of insecticides to reduce insect vectors (carrying either spermatia or aeciospores).

Application

At the initiation of an eradication program, evaluate the available fungicides for their use in program operations. Select a fungicide after considering local conditions along with survey results.

Labeling

While the proposed formulation is approved for an effective eradication program, it may not be labeled, at the time of pest detection, for the specific use-site where treatment is required. If a formulation is not labeled for the needed use, it may be possible to request a Federal Crisis or Quarantine Exemption from the U.S. Environmental Protection Agency (EPA) under Section 18 of the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA). For further information refer to *Regulatory Procedures* on page 5-1. The prescribed formulation must be labeled for use on the site where it is to be applied and must be registered for use in the State where the eradication program is occurring. All applicable label directions must be followed, including requirements for personal protection equipment, maximum treatment rates, storage and disposal.

If the pathogen population was discovered too late for eradication measures to be effective, new measures should rely on containment or management options. Containment means keeping the target population of infected plants confined to a specific area, and perhaps later developing tools to eradicate it. Using this approach requires strong regulatory procedures.

A variation of containment is known as Slow the Spread (USDA–APHIS–PPQ, 2003). In Slow the Spread, the spread of the pest population is slowed as much as possible, resources permitting. In contrast, management is used when the population of the pathogen is so large or widely spread that resources are better directed at limiting the impacts caused by the infestation. The following control options are best suited for both containment programs and long term management. They could be used in an eradication program if the intent is to bring population numbers down to better achieve this goal.

Biological Control

Biological control agents are useful for suppressing pathogen populations, but do not eradicate them. Biological control can be useful if rigorous screening on non-target organisms is tested. Obtain the proper permits from PPQ-Plant, Organism, and Soil Permits, prior to testing.

The hyperparasite *Cladosporium tenuissimum* is proposed by Moricca et al. (2001) as a possible means of control for stem rust. The aeciospores are directly penetrated and parasitized by the conidial fungus. Tests on two-year-old pine seedlings in the greenhouse showed that treatment with the parasite prevented new rust infections by an average of 42 percent. *Cladosporium tenuissimum* has been reported as a hyperparasite of *Cronartium flaccidum* and has been isolated from the aeciospores of *Cronartium flaccidum* and its autoecious form *Peridermium pini* (Moricca et al., 1999; Moricca et al., 2001; Nasini et al., 2004). Based on its ability to reduce aeciospore germination, reduce viability of aeciospores, reduce rust development under greenhouse conditions over 2 years, and survive and multiply in forest ecosystems without rusts being present, *C. tenuissimum* appears to be a promising agent for the biological control of pine stem rusts in Europe (Moricca et al., 2001).

Another naturally occurring hyperparasite of *Cronartium flaccidum* is *Tuberculina maxima* (purple mold) described as a parasite on aecia of rust fungi may suppress rust sporulation to a limited extent.

Host Resistance

Raddi et al. (1979), Raddi and Ragazzi (1980), and Raddi et al. (1980) discuss current progress and issues with breeding for resistance to *Cronartium flaccidum* in pines.

Selection of more resistant species or provenances of pines for growing in areas of stem blister rust is a feasible and promising means of control, although Moricca et al. (2001) state that breeding efforts were not successful. The testing methodology may be a major factor in the usefulness of results obtained. Because the fungus develops slowly even in susceptible plants, progress in rating the plants for resistance must be slow.

Raddi and Fagnani (1978) used three methods to inoculate seedlings and young plants of different pine species with basidiospores of *Cronartium flaccidum*. Species from southern Europe were susceptible while North American and Asian species appeared resistant. In later tests, differing levels of susceptibility were found in three of the European species (Raddi et al.,

1979) and results indicated that selection for resistance might be possible in *Pinus pinaster.*

Although *Pinus sylvestris* appeared resistant in the limited tests in Italy (Raddi and Fagnani, 1978), it is a major host species in northern Europe, and differences in susceptibility have been observed (Mordue and Gibson, 1978). Over a number of years, Kaitera and Nuorteva (Kaitera and Nuorteva, 2008) tested seedlings 1 to 7 years old with aeciospores of *Peridermium pini* and basidiospores of *Cronartium flaccidum*. Little disease was obtained and no significant differences among provenances of Finnish trees were observed. The apparent resistance of the introduced American species *P. contorta* in the same tests led the researchers to recommend use of that species as an alternative to *P. sylvestris* in Finland.

Although levels of rust disease caused by both forms were low (ten percent or less), Kuzmina and Kuzmin (2008) did find variation in the resistance of climatypes of *Pinus sylvestris* from different parts of Russia in trials in Western Siberia. Soil type and humidity affected the severity of disease as well as the strength of the provenance tests.

Among the several alternate hosts of *Cronartium flaccidum*, differences in susceptibility would be expected as well. Roll-Hansen (Roll-Hansen, 1973) found strong resistance in some *Paeonia* (ornamental peony) cultivars and suggested that use of those in gardens could assist in control of *C. flaccidum*. Kaitera et al. (1999a) also observed variation in susceptibility among telial hosts in several genera, including *Paeonia*, but found the rust to have a low host specificity in general, since infections by Finnish isolates occurred on both native and non-native species.

Chapter

7

Environmental Compliance

Contents

Introduction

Use *Chapter 7 Environmental Compliance* as a guide to the environmental regulations when conducting a program against Scots pine blister rust (*Cronartium flaccidum* and *Peridermium pini*) absent from the United States and collaborating territories.

Overview

Program managers of Federal emergency response or domestic pest control programs must ensure that their programs comply with all Federal Acts and Executive Orders pertaining to the environment, as applicable. Two primary Federal Acts, the National Environmental Policy Act (NEPA) and the Endangered Species Act (ESA), often require the development of significant documentation before program actions may begin.

Program managers should also seek guidance and advice as needed from Environmental and Risk Analysis Services (ERAS), a unit of APHIS' Policy and Program Development (PPD) staff. ERAS is available to give guidance and advice to program managers and prepare drafts of applicable environmental documentation.

In preparing draft NEPA documentation ERAS may also perform and incorporate assessments that pertain to other acts and executive orders described below, as part of the NEPA process. The Environmental Compliance Team (ECT), a part of PPQ's Emergency Domestic Programs (EDP), will assist ERAS in the development of documents, and will implement any environmental monitoring.

Leaders of programs are strongly advised to meet with ERAS and/or ECT early in the development of a program in order to conduct a preliminary review of applicable environmental statutes and to ensure timely compliance. Environmental monitoring of APHIS pest control activities may be required as part of compliance with environmental statutes, as requested by program managers, or as suggested to address concerns with controversial activities. Monitoring may be conducted with regards to worker exposure, pesticide quality assurance and control, off-site chemical deposition, or program efficacy. Different tools and techniques are used depending on the monitoring goals and control techniques used in the program. Staff from ECT will work with the program manager to develop an environmental monitoring plan, conduct training to carry out the plan, give day-to-day guidance on monitoring, and provide an interpretive report of monitoring activities.

National Environmental Policy Act

The National Environmental Policy Act (NEPA) requires all Federal agencies to examine whether their actions may significantly affect the quality of the human environment. The purpose of NEPA is to inform the decisionmaker before taking action, and to tell the public of the decision. Actions that are excluded from this examination, that normally require an Environmental Assessment, and that normally require Environmental Impact Statements, are codified in APHIS' NEPA Implementing Procedures located in 7 CFR 372.5.

The three types of NEPA documentation are Categorical Exclusions, Environmental Assessments, and Environmental Impact Statements.

Categorical Exclusion

Categorical Exclusions (CE) are classes of actions that do not have a significant effect on the quality of the human environment and for which neither an Environmental Assessment (EA) nor an environmental impact statement (EIS) is required. Generally, the means through which adverse environmental impacts may be avoided or minimized have been built into the actions themselves (7 CFR 372.5(c)).

Environmental Assessment

An Environmental Assessment (EA) is a public document that succinctly presents information and analysis for the decisionmaker of the proposed action. An EA can lead to the preparation of an environmental impact statement (EIS), a finding of no significant impact (FONSI), or the abandonment of a proposed action.

Environmental Impact Statement

If a major Federal action may significantly affect the quality of the human environment (adverse or beneficial) or the proposed action may result in public controversy, then prepare an Environmental Impact Statement (EIS).

Endangered Species Act

The Endangered Species Act (ESA) is a statute requiring that programs consider their potential effects on federally-protected species. The ESA requires programs to identify protected species and their habitat in or near program areas, and document how adverse effects to these species will be avoided. The documentation may require review and approval by the U.S. Fish and Wildlife Service and the National Marine Fisheries Service before program activities can begin. Knowingly violating this law can lead to criminal charges against individual staff members and program managers.

Migratory Bird Treaty Act

The statute requires that programs avoid harm to over 800 endemic bird species, eggs, and their nests. In some cases, permits may be available to capture birds, which require coordination with the U.S. Fish and Wildlife Service.

Clean Water Act

The statute requires various permits for work in wetlands and for potential discharges of program chemicals into water. This may require coordination with the Environmental Protection Agency, individual States, and the Army Corps of Engineers. Such permits would be needed even if the pesticide label allows for direct application to water.

Tribal Consultation

The Executive Order requires formal government-to-government communication and interaction if a program might have substantial direct effects on any federally-recognized Indian Nation. This process is often incorrectly included as part of the NEPA process, but must be completed before public involvement under NEPA. Staff should be cognizant of the conflict that could arise when proposed Federal actions intersect with Tribal sovereignty. Tribal consultation is designed to identify and avoid such potential conflict.

National Historic Preservation Act

The statute requires programs to consider potential impacts on historic properties (such as buildings and archaeological sites) and requires coordination with local State Historic Preservation Offices. Documentation under this act involves preparing an inventory of the project area for historic properties and determining what effects, if any, the project may have on historic properties. This process may need public involvement and comment before the start of program activities.

Coastal Zone Management Act

The statute requires coordination with States where programs may impact Coastal Zone Management Plans. Federal activities that may affect coastal resources are evaluated through a process called Federal consistency. This process allows the public, local governments, Tribes, and State agencies an opportunity to review the Federal action. The Federal consistency process is administered individually by states with Coastal Zone Management Plans.

Environmental Justice

The Executive Order requires consideration of program impacts on minority and economically disadvantaged populations. Compliance is usually achieved within the NEPA documentation for a project. Programs are required to consider if the actions might impact minority or economically disadvantaged populations and if so, how such impact will be avoided.

Protection of Children

The Executive Order requires Federal agencies to identify, assess, and address environmental health risks and safety risks that may affect children. If such a risk is identified, then measures must be described and carried out to minimize such risks.

Chapter 8

Pathways

Contents

Introduction

Use *Chapter 8 Pathways* as a source of information on the pathways of introduction in the United States of Scots pine blister rust (*Cronartium flaccidum* and *Peridermium pini*). The rust fungus could accidentally be introduced through transport of infected seedlings, young trees or on pine wood while symptoms are still latent. Scots pine blister rust should be regarded as a moderate threat to the U.S. Christmas tree production and forest ecosystem.

Natural Movement

Natural spread of *Cronartium flaccidum* and *Peridermium pini* into the continental United States is considered a rare possibility, although, like many other rust fungi, the Scots pine blister rust fungus produces spores that can be occasionally transported long distance in the atmosphere by wind. A report by Hunt (1997), indicated that the rust's spores least easily transported are the basidiospores, which may be disseminated less than 500 meters away from its original source. Insects can act as vectors for all spore stages. *Pissodes pini*, *Dioryctria splendidella*, *Laspeyresia coniferana*, *Lagria hirta* and *Dioryctria abietella* are reported as possible vectors for the rust on the basis of their occurrence and because they feed on *C. flaccidum* aecia.

The risk of introduction of either form of this rust is greater for those temperate parts of the Southern Hemisphere, such as Australia, where introduced *Pinus* species are grown in plantations (Neumann and Marks, 1990). Single-aged monoculture populations could suffer epidemics, in particular, if trees are at a susceptible age and the autoecious *Peridermium pini* form is introduced. In addition, dicotyleconous hosts for the heteroecious *C. flaccidum* are native to the temperate countries of the Southern Hemisphere (USDA–ARS, 2011).

Other alternate hosts (*Gentiana* spp., *Paeonia* spp.) may be introduced as ornamentals and be grown near introduced ornamental pines.

Commerce

Signs of *Cronartium flaccidum* and *Peridermium pini* and symptoms of the disease may be latent (inactive, hidden, or dormant) for 2 or more years in infected pine host material and up to a month in leafy hosts. The chance of introduction into the United States is high because visual survey of propagative material may not be effective due to this latency (Geils et al., 2009). According to Geils et al. (2009), Japanese black pine (*Pinus thunbergii*), mugo pine (*Pinus mugo*) or other 2 or 3-needled pines, commonly used for bonsai, pose a significant risk for the introduction of *C. flaccidum* if imported as whole plants.

Plant parts liable to carry the pest in trade/transport are bark, leaves, stems, shoots, trunks, branches and wood. Accidental introduction of the rust as a latent infection in pines or on pine wood, appears more likely than on ornamental dicots, but the continent has no lack of pines for planting and regulatory agencies have established phytosanitary procedures (Canadian Food Inspection Agency, 2008; USDA–APHIS, 2008).

Introduction of *Cronartium flaccidum* to new temperate regions would require importation of infected seedlings without quarantine or the occurrence of viable aecia on bark-bearing pine wood materials or products. Apparently, due to regulatory vigilance or other circumstances, this has not happened yet.

Cronartium flaccidum is an imminent threat that could be introduced into the United States with imported leaves, bark and stems of infected plants (CABI, 2010). Due to the relatively slow rate of disease spread through natural secondary dissemination, containment and eradication should be actively pursued. The success of such strategy would rely on accurate and timely identification of the pathogen matched by quarantine action and targeted eradication implementing all necessary control measures.

Once a positive identification has been made confirming the presence of infected plants by *Cronartium flaccidum*, investigations should be initiated to determine the probable origin of the initial infections and the extent of distribution of potentially infected plants in the U.S. Territories.

After investigations are performed and the risk of pathogen establishment is evaluated the Deputy Administrator will issue a letter directing PPQ field offices to initiate specific actions under the Plant Protection Act. The Plant Protection Act of 2000 provides for authority for emergency quarantine action.

Program personnel must maintain records and maps noting the location of all detections, the number and type of plants subjected to control actions, and the materials and chemical formulations used in each treated area.

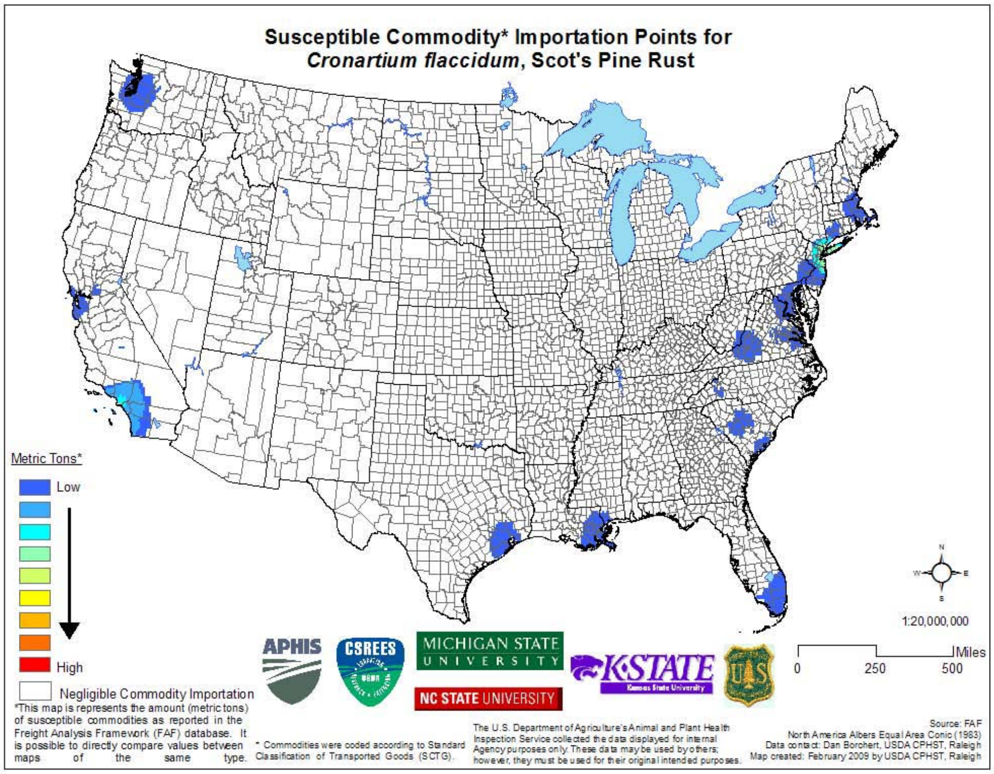

Figure 8-1 Importation Points for *Cronartium flaccidum* Within the Conterminous United States; Map Courtesy of USDA–APHIS–PPQ–CPHST; (For More Information on the Importation Points Map Please Visit http://www.nappfast.org/)

Scots Pine Blister Rust

References

Use *References* to learn more about the publications, Web sites, and other resources that were consulted during the production of the guidelines.

Publications, Web Sites, and Other

Canadian Food Inspection Agency, 2008. Wood packaging. Ottawa, Canada: Government of Canada. http://www.inspection.gc.ca/english/plaveg/for/cwpc/wdpkge.shtml (Archived at PERAL).

Alexopoulos, C. J., C. W. Mims, and M. Blackwell. 1996. Introductory Mycology (Fourth edition). John Wiley and Sons, Inc, New York, New York, USA. 868 pp.

Azbukina, Z. M. 1995. *Cronartium* species on *Pinus* species in the Russian far east. Pages 65-69 in S. Kaneko, K. Katsuya, M. Kakishima, and Y. Ono, (eds.). Proceedings of the 4th IUFRO Rusts of pines Working Party Conference, Tsukuba, Japan.

Buchko, J., and G. R. Klassen. 1990. Detection of length heterogeneity in the ribosomal DNA of *Pythium ultimum* by PCR amplification of the intergenic region. Current Genetics 18:203-205.

Burdine, C. S., T. L. Kubisiak, G. N. Johnson, and C. D. Nelson. 2007. Fifty-two polymorphic microsatellite loci in the rust fungus, *Cronartium quercuum* f.sp. fusiforme. Molecular Ecology Notes 7:1005-1008.

Butin, H. 1995. Tree Diseases and Disorders. Oxford University Press, Oxford, United Kingdom. 252 pp.

CABI. 2010. Crop Protection Compendium. in. Commonwealth Agricultural Bureau International (CABI), Wallingford, UK.

CABI. 2011a. Crop Protection Compendium: *Cronartium flaccidum*. Accessed November 21, 2011. http://www.cabi.org/cpc/, Wallingford, UK

CABI. 2011b. Species Fungorum. Accessed September 26, 2011. http://www.speciesfungorum.org/Names/Names.asp

Cao, Z. M., Z. Q. Li, and J. Y. Zhuang. 2000. Uredinales from the Qinling Mountains. Mycosystema 19(1):13-23.

Chalkley, D. 2010. Invasive Fungi. Scots stem pine rust - *Cronartium flaccidum*. Systematic Mycology and Microbiology Laboratory, ARS, USDA. http://nt.ars-grin.gov/sbmlweb/fungi/index.cfm (Archived at PERAL)

Chen, M. M. 2002. Forest Fungi Phytogeography: Forest Fungi Phytogeography of China, North America, and Siberia and International Quarantine of Tree Pathogens. Pacific Mushroom Research and Education Center, Sacramento, California. 469 pp.

Cheng, D., Y. Xue, X. Pan, and W. Li. 1998. Population genetic structures of three *Cronartium* species from China based upon allozyme analysis. Mycosystema 17(1):32-39.

Cheng, D. S., Y. Xue, and L. P. Shaeo. 1995. Differentiation among *Cronartium* species form northeast China by isozyme analysis. Pages 71-75 in Proceedings of the Fourth Rust of Pines Working Party Conference, Institute of Agriculture and Forestry. University of Tsukuba, Tsukuba, Japan.

Cho, W. D., and H. D. Shin. 2004. List of plant diseases in Korea (Fourth edition). Korean Society of Plant Pathology, Seoul, Republic of Korea. 779 pp.

Churakov, B. P. 1989. Infection of different forms of Scots pine by phytopathogenic fungi. Lesovedenie (1):69-73.

CMI. 1989. *Cronartium flaccidum*. [Distribution map]. in Distribution Maps of Plant Diseases, No. 616, edition 1. CAB International, Wallingford; UK.

Denchev, C. M. 1995. Bulgarian Uredinales. Mycotaxon 55:405-465.

Diamandis, S., and M. d. Kam. 1986. A severe attack of Scots pine by resin top disease in N. Greece. European Journal of Forest Pathology 16(4):247-249.

Dudka, I. O., V. P. Heluta, Y. Y. Tykhonenko, T. V. Anrianova, V. P. Hayova, M. P. Prydiuk, V. V. Dzhagan, and V. P. Isikov. 2004. Fungi of the Crimean Peninsula. National Academy of Sciences of Ukraine, Kiev, Ukraine. 452 pp.

Farr, D. F., and A. Y. Rossman. 2011. Fungal Databases, Systemic Mycology and Microbiology Laboratory. ARS, USDA http://nt.ars-grin.gov/fungaldatabases/ (Archived at PERAL)

Geils, B. W., N. B. Klopfenstein, M.-S. Kim, P. Spaine, B. A. Richardson, P. J. Zambino, C. G. Shaw, J. Walla, R. Bulluck, L. Redmond, and K. Smith. 2009. Recovery plan for Scots pine blister rust caused by *Cronartium flaccidum* (Alb. & Schwein.) G. Winter and *Peridermium pini* (Pers.) Lev. [syn. C. asclepiadeum (Willd.) Fr., *Endocronartium pini* (Pers.) Y. Hiratsuka].

Gibbs, J. N., N. England, and R. Wolstenholme. 1988. Variation in the pine stem rust fungus *Peridermium pini* in the United Kingdom. Plant Pathology 37(1):45-53.

Gibbs, J. N., B. J. W. Greig, and I. T. Hickman. 1987. An analysis of *Peridermium* stem rust of Scots pine in Thetford forest in 1984 and 1985. Forestry 60(2):203-218.

Gonçalves, D. C. A. 1936. Uredineae of Portugal. (Uredineas de Portugal.). Bol. Soc. broteriana, 11:169-265.

Greig, B. J. W. 1987. History of *Peridermium* stem rust of Scots pine (*Pinus sylvestris* L.) in Thetford forest, East Anglia. Forestry 60(2):193-202.

Greig, B. J. W., and A. L. Sharpe. 1991. Pine stem rust (*Peridermium pini*) in a Scots pine provenance trial at Teindland forest. Scottish Forestry 45(3):169-174.

Hantula, J., R. Kasanen, J. Kaitera, and S. Moricca. 2002. Analyses of genetic variation suggest that pine rusts *Cronartium flaccidum* and *Peridermium pini* belong to the same species. Mycological Research 106(2):203-209.

Hantula, J., E. M. Niemi, J. Kaitera, R. Jalkanen, and T. Kurkela. 1998. Genetic variation of the resin top fungus in Finland as determined by random amplified microsatellites (RAMS). European Journal of Forest Pathology 28(6):361-372.

Harvey, A. E., and J. L. Grasham. 1974. Axenic culture of the mononucleate stage of *Cronartium ribicola*. Phytopathology 64:1028-1035.

Hiratsuka, N. 1932. Inoculation experiments with some heteroecious species of the Melampsoraceae in Japan. Japanese Journal of Botany 6:1-33.

Hiratsuka, N., and Z. C. Chen. 1991. A list of Uredinales collected from Taiwan. Transactions of the Mycological Society of Japan 32(1):3-22.

Hiratsuka, Y. 1968. Morphology and cytology of aeciospores and aeciospore germ tubes of host-alternating and pine-to-pine races of *Cronartium flaccidum* in northern Europe. Canadian Journal of Botany 46(9):1119-1122.

Hiratsuka, Y. 1969. *Endocronartium*, a new genus for autoecious Pine stem rusts. Canadian Journal of Botany 47(9):1493-1495.

Hunt, R. 1997. Stem rusts. in E. M. Hansen and K. J. Lewis, (eds.). Compendium of conifer diseases. American Phytopathological Society, St. Paul, Minnesota, USA.

Jing, Y., W. H. Li, and S. G. Zhao. 1995. Study on pine rusts in Northwest China. Pages 37-41 in S. Kaneko, K. Katsuya, M. Kakishima, and Y. Ono, (eds.). Proceedings of the Fourth IUFRO Rusts of Pines Working Party Conference, Tsukuba. Laboratory of Plant Pathology & Mycology, University of Tsukuba, Tsukuba, Ibaraki, Japan.

Jing, Y., and P. Wang. 1989. A study of the blister rust of *Pinus* massoniana of China. Pages 302-312 in Y. Hiratsuka, J. K. Samoil, P. V. Blenis, P. E. Crane, and B. L. Laishley, (eds.). Rusts of Pine. Proceedings of the IUFRO Rusts of Pine Working Party Conference. Forestry Canada, Canada.

Jurc, D. 2007. Diseases of shoots, branches and trunk. Gremeniella abietina, *Cronartium flaccidum*, *Melampsora pinitorqua*. / Bolezni poganjkov, vej in debla. *Gremeniella abietina*, *Cronartium flaccidum*, *Melampsora pinitorqua*. Gozdarski Vestnik 65(2):89-104.

Kaitera, J. 1999. *Cronartium flaccidum* fruitbody production on *Melampyrum* spp. and some important alternate hosts to pine. European Journal of Forest Pathology 29(6):391-398.

Kaitera, J. 2000. Analysis of *Cronartium flaccidum* lesion development on pole-stage Scots pines. Silva Fennica 34(1):21-27.

Kaitera, J., T. Aalto, and R. Jalkanen. 1994. Effect of resin-top disease caused by *Peridermium pini* on the volume and value of *Pinus sylvestris* saw timber and pulpwood. Scandinavian Journal of Forest Research 9(4):376-381.

Kaitera, J., and J. Hantula. 1998. *Melampyrum sylvaticum*, a new alternate host for pine stem rust *Cronartium flaccidum*. Mycologia 90(6):1028-1030.

Kaitera, J., and R. Hiltunen. 2011. Susceptibility of *Pedicularis* spp. to *Cronartium ribicola* and *C. flaccidum* in Finland. Forest Pathology 41(3):237-242.

Kaitera, J., and H. Nuorteva. 2003a. *Cronartium flaccidum* produces uredinia and telia on *Melampyrum nemorosum* and on Finnish *Vincetoxicum hirundinaria*. Forest Pathology 33(4):205-213.

Kaitera, J., and H. Nuorteva. 2003b. Relative susceptibility of four *Melampyrum* species to *Cronartium flaccidum*. Scandinavian Journal of Forest Research 18(6):499-504.

Kaitera, J., and H. Nuorteva. 2006a. Finnish *Cronartium ribicola* does not infect alternate hosts of *Cronartium flaccidum*. Forest Pathology 36(4):247-252.

Kaitera, J., and H. Nuorteva. 2006b. Susceptibility of *Ribes* spp. to pine stem rusts in Finland. Forest Pathology 36(4):225-246.

Kaitera, J., and H. Nuorteva. 2008. Inoculations of eight *Pinus* species with *Cronartium* and *Peridermium* stem rusts. Forest Ecology and Management 255(3/4):973-981.

Kaitera, J., H. Nuorteva, and J. Hantula. 2005. Distribution and frequency of *Cronartium flaccidum* on *Melampyrum* spp. in Finland. Canadian Journal of Forest Research 35(2):229-234.

Kaitera, J., L. Seitamäki, J. Hantula, R. Jalkanen, and T. Kurkela. 1999a. Inoculation of known and potential alternate hosts with *Peridermium pini* and *Cronartium flaccidum* aeciospores. Mycological Research 103(2):235-241.

Kaitera, J., L. Seitamäki, J. Hantula, R. Jalkanen, and T. Kurkela. 1999b. Morphological variation of *Peridermium pini* and *Cronartium flaccidum* aeciospores. Mycological Research 103(6):677-683.

Kakishima, M., M. Imazu, K. Katsuya, Z. M. Azbukina, Y. Ono, S. Kaneko, Y. Hiratsuka, and S. Sato. 1995. Preliminary survey of pine blister rusts in the Russian Far East. Pages 49-63 in S. Kaneko, K. Katsuya, M. Kakishima, and Y. Ono, (eds.). Proceedings of the Fourth IUFRO Rusts of Pines Working Party Conference, Tsukuba. Laboratory of Plant Pathology & Mycology, University of Tsukub, Tsukuba, Ibaraki, Japan.

Karadzic´, D., and V. Vujanovic´. 2009. Aleppo pine defence against Sphaeropsis sapinea, *Cronartium flaccidum* and other fungal pathogens in the Mediterranean part of Montenegro. Glasnik Šumarskog Fakulteta, Univerzitet u Beogradu (99):59-74.

Kasanen, R. 1997. Aeciospores of *Cronartium flaccidum*, C. *ribicola* and *Endocronartium pini* show no differences in morphology. European Journal of Forest Pathology 27(4):251-260.

Kasanen, R. 2001. Relationship between *Cronartium flaccidum* and *Peridermium pini*. Academic dissertation, University of Helsinki.

Kasanen, R., J. Kaitera, and J. Hantula. 2000. The genetic composition of *Peridermium pini* and *Cronartium flaccidum* cankers on Scots pine as revealed by two multi-allelic loci. Forest Pathology 30(4):221-230.

Klebahn, H. 1938. Offene Fragen und neue Beobachtungen nber die rindenwohnenden Blasenroste der Kiefern nebst Bemerkungen nber einige andere Rostpilze. Zeitschrift fnr Pflanzenkrankheiten 48:369-410.

Klingstrom, A. 1973. Studies on free amino acid contents in the aeciospores of *Endocronartium pini* and *Cronartium flaccidum*. European Journal of Forest Pathology 3(4):209-214.

Kobayashi, T. 2007. Index of Fungi Inhabiting Woody Plants in Japan. Host, Distribution and Literature. Zenkoku-Noson-Kyiku Kyokai Publishing Co., Tokyo, Japan. 1227 pp.

Kuprevich, V., and V. Transchel. 1957. Cryptogamic plants of the USSR, vol. IV. Rust Fungi. No. 1, Family Melampsoraceae. USSR Academy of Sciences, Moscow, USSR.

Kuzmina, N. A., and S. R. Kuz'Min. 2008. Intraspecific response of Scots pine (*Pinus sylvestris* L.) to pathogens in a provenance trial in middle Siberia. Eurasian Journal of Forest Research 11-2:51-59.

Leont'eva, S. I., and N. P. Stenina. 1990. Phytopathological state of pine and spruce forests cultivated in the north-west of the Russian Federation. Mikologiya i Fitopatologiya 23(4):389-392.

MAF. 2009. Search results - *Endocronartium pini*. Biosecurity New Zealand, Ministry of Agriculture and Forestry. Last accessed July 2,2009. http://www.biosecurity.govt.nzlsearch/node/Endocronartium+pini

Maloy, O. C. 1997. White pine blister rust control in North America: A case history. Pages 87-109 in Annual Review of Phytopathology.

Martinsson, O., and B. Nilsson. 1987. The impact of *Cronartium flaccidum* on the growth of *Pinus sylvestris*. Scandinavian Journal of Forest Research 2(3):349-357.

Mittempergher, L., and P. Raddi. 1977. Variation of diverse sources of *Cronartium flaccidum*. European Journal of Forest Pathology 7(2):93-98.

Mordue, J. E. M., and l. A. S. Gibson. 1978. *Cronartium flaccidum*. CMI Descriptions of Phytopathogenic Fungi and Bacteria 580:1-2.

Moricca, S., T. Kasuga, K. Mitchelson, A. Ragazzi, and S. Diamandis. 1996. Heterogeneity in intergenic regions of the ribosomal repeat of the pine-blister rusts *Cronartium flaccidum* and *Peridermium pini*. Current Genetics 29(4):388-394.

Moricca, S., and A. Ragazzi. 1994. Axenic culture of the aecial state of *Cronartium flaccidum* from Italy. Mycological Research 98(11):1258-1262.

Moricca, S., and A. Ragazzi. 1996. Culture characteristics and variation of *Cronartium flaccidum* isolates. Canadian Journal of Botany 74(6):924-933.

Moricca, S., and A. Ragazzi. 1998. Use of RFLP and SSCP analysis to differentiate the pine rusts *Cronartium flaccidum* and *Peridermium pini*. Mycological Research 102(6):666-670.

Moricca, S., and A. Ragazzi. 2001. Establishment of single-genotype axenic cultures from the haploid stage of the pine blister rust *Cronartium flaccidum*. Mycological Research 105(12):1527-1532.

Moricca, S., and A. Ragazzi. 2008. Biological and integrated means to control rust diseases. Pages 303-329 in A. Ciancio and K. G. Mukerji, (eds.). Integrated Management of Plant Pests and Diseases. Springer, Dordrecht, The Netherlands.

Moricca, S., A. Ragazzi, and K. R. Mitchelson. 1999. Molecular and conventional detection and identification of *Cladosporium tenuissimum* on two-needle pine rust aeciospores. Canadian Journal of Botany 77(3):339-347.

Moricca, S., A. Ragazzi, K. R. Mitchelson, and G. Assante. 2001. Antagonism of the two-needle pine stem rust fungi *Cronartium flaccidum* and *Peridermium pini* by *Cladosporium tenuissimum* in vitro and in planta. Phytopathology 91(5):457-468.

Moriondo, F. 1975. Characteristics of epidemics of blister rust of Pines, *Cronartium flaccidum* [C. asclepiadeum], in Italy. Annali, Accademia Italiana di Scienze Forestali 24:331-406.

Mulenko, W., M. Kozlowska, and B. Salata. 2004. Microfungi of the Tatra National Park. A checklist. Poland: W. Szafer Institute of Botany, Polish Academy of Sciences. 72 pp.

Murashige, T., and F. Skoog. 1962. A revised medium for the rapid growth and bioassay with tobacco tissue cultures. Physiologia Plantarum 15:473-497.

Nasini, G., A. Arnone, G. Assante, A. Bava, S. Moricca, and A. Ragazzi. 2004. Secondary mould metabolites of *Cladosporium tenuissimum*, a hyperparasite of rust fungi. Phytochemistry 65(14):2107-2111.

NASS. 2010. Census of Horticultural Specialties (2009). National Agricultural Statistics Service. United States Department of Agriculture (NASS), Washington, D.C. 595 pp.

NCBI. 2011. Entrez cross-database search engine. National Center for Biotechnology Information, U.S. National Library of Medicine, Bethesda, Maryland, USA. http://www.ncbi.nlm.nih.gov/sites/gquery Accesses November 30, 2011.

Neumann, F. G., and G. C. Marks. 1990. Status and management of insect pests and diseases in Victorian softwood plantations. Australian Forestry 53(2):131-144.

Pappinen, A., and K. Von Weissenberg. 1996. Weevil feeding on Scots pine affects germination of *Endocronartium pini*. European Journal of Forest Pathology 26(5):225-234.

Pappinen, A., and K. v. Weissenberg. 1994. The ability of the pine-top weevil to carry spores and infect Scots pine with *Endocronartium pini*. European Journal of Forest Pathology 24(5):258-263.

Parke, J. L., and N. J. Grünwald. 2012. A Systems Approach for Management of Pests and Pathogens of Nursery Crops. Plant Disease 96(9):1236-1244.

Pei, M. H., and J. N. Gibbs. 1991. Cultural variation in *Peridermium pini* from two parts of the United Kingdom. European Journal of Forest Pathology 21(3):164-167.

Pitt, D. G., T. Meyer, M. Park, L. MacDonald, T. Buscarini, and D. G. Thompson. 2006. Application of slow-release tablets to enhance white pine regeneration: Growth response and efficacy against white pine blister rust. Canadian Journal of Forest Research 36(3):684-698.

Raddi, P., and A. Fagnani. 1978. Relative susceptibility to blister rust caused by *Cronartium flaccidum* of several species of pine. European Journal of Forest Pathology 8(1):58-61.

Raddi, P., L. Mittempergher, and A. Fagnani. 1980. Artificial inoculation of large numbers of pine seedlings with *Cronartium flaccidum*. Phytopathologia Mediterranea 19:44-50.

Raddi, P., L. Mittempergher, and F. Moriondo. 1979. Testing of *Pinus* pinea and *P. pinaster* progenies for resistance to *Cronartium flaccidum*. Phytopathology 69(6):679-681.

Raddi, P., and A. Ragazzi. 1980. Italian studies on resistance to pine blister rust (*Cronartium flaccidum*). Resist. Dis. Pests. for Trees:435-440.

Ragazzi, A., and I. Dellavalle Fedi. 1982. Observations under fluorescence on progress of basidiospore germination in *Cronartium flaccidum* (Alb. et Schw.) Wint. on the needle surface of certain pine species. European Journal of Forest Pathology 12(4/5):246-251.

Ragazzi, A., and I. Dellavalle Fedi. 1983. Effect of a fluorescent brightener on the germinative power of basidiospores of *Cronartium flaccidum*. European Journal of Forest Pathology 13(5/6):372-376.

Ragazzi, A., I. Dellavalle Fedi, and L. Mesturino. 1989. The effects of some variables on the production of uredia and telia of *Cronartium flaccidum*. Phytopathologia Mediterranea 28(1):43-45.

Ragazzi, A., A. Fagnani, and I. Dellavalle Fedi. 1987. Telial and basidiospore stages of *Cronartium flaccidum*: light and scanning electron microscopy observations. Phytopathologia Mediterranea 26(2):113-116.

Ragazzi, A., and I. D. Fedi. 1992. Penetration of *Cronartium flaccidum* into pine needles. European Journal of Forest Pathology 22(5):278-283.

Ragazzi, A., I. D. Fedi, and L. Mesturino. 1986a. *Cronartium flaccidum* (Alb. et Schw.) Wint. spores: temperature requirements for germination. Phytopathologia Mediterranea 25(1-3):57-60.

Ragazzi, A., I. D. Fedi, and L. Mesturino. 1986b. *Cronartium flaccidum* on *Pinus* spp.: relation of inoculum concentration to symptom development. European Journal of Forest Pathology 16(1):16-21.

Ragazzi, A., S. Moricca, and I. Dellavalle. 1995. Growth of axenic cultures of *Cronartium flaccidum* on callus tissue from *Pinus nigra* var. laricio and *Pinus sylvestris*. European Journal of Forest Pathology 25(1):31-37.

Ragazzi, A., and F. Moriondo. 1980. Results of *Cronartium flaccidum* (Alb. & Schw.) Wint. inoculations on eight-year-old plants of *Pinus* pinea L. Phytopathologia Mediterranea 19:51-56.

Robert, V., G. Stegehuis, and J. Stalpers. 2005. The MycoBank engine and related databases. http://www.mycobank.org. (Archived at PERAL).

Roll-Hansen, F. 1973. Resistance of Paeonia cultivars to *Cronartium flaccidum* in Norway. European Journal of Forest Pathology 3(3):142-145.

Rozhkov, A. A. 1975. Resin canker. Zashchita Rastenii 1:44-45.

Samils, B., K. Ihrmark, J. Kaitera, J. Stenlid, and P. Barklund. 2011. New genetic markers for identifying *Cronartium flaccidum* and *Peridermium pini* and examining genetic variation within and between lesions of Scots pine blister rust in Sweden. Fungal Biology 115(12):1303-1311.

Schenk, R. U., and A. C. Hildebrandt. 1972. Medium and techniques for induction and growth of monocotyledonous and dicotyledonous plant cell cultures. Canadian Journal of Botany 50(1):199-204.

Siwecki, R., and B. Chojnacki. 1989. Epidemiology of Scots pine rusts in Poland. Pages 158-163 in Y. Hiratsuka, J. K. Samoil, P. V. Blenis, P. E. Crane, and B. L. Laishley, (eds.). Rusts of pine. Proceedings of the IUFRO Rusts of Pine Working Party Conference, Canada. Forestry Canada.

Smith, I. M., J. Dunez, R. A. Lelliott, D. H. Phillips, and S. A. Archer. 1988. European Handbook of Plant Diseases. Blackwell Scientific Publications, Oxford, UK. 583 pp.

Storozhenko, V. G. 1987. Fungus diseases in recreation forests. Lesnoe Khozyaistvo 3:63-65.

Szabo, I. 1998. Occurrence and importance of rust fungi in Hungarian forests. Finnish Forest Research Institute, Research Paper (712):9-13.

Tai, F. L. 1979. Sylloge Fungorum Sinicorum. Science Press, Beijing, China. 1527 pp.

USDA-APHIS. 2000. Regulated Plant Pest List.

USDA-ARS. 2011. Germplasm Resources Information Network (GRIN). Online Database. National Germplasm Resources Laboratory, Beltsville, Maryland, USA. http://www.ars-grin.gov/cgi-bin/npgs/html/tax_search.pl (Archived at PERAL)

USDA-NASS. 2010. 2007 Census of Agriculture. Census of Horticultural Specialties (2009) (AC-07-SS-3). 595 pp.

USDA/APHIS. 2008. Circular 34: Diseases of Picea spp. Post-entry Quarantine Manual for State Inspectors, Washington, D.C., USA. 10-151-153 pp.

USFWS. 2011. Threatened and Endangered Species System (TESS). United States Fish and Wildlife Service (USFWS). Last accessed September 8, 2011, http://ecos.fws.gov/tess_public/

Venette, R. C. (ed.). 2008. Pine Commodity-Based Survey Reference. Cooperative Agricultural Pest Survey. Animal and Plant Health Inspection Service, U.S. Dept. of Agriculture. [Available on-line at: http://ceris.purdue.edu/caps/adm2008/adm2008000032.pdf]. 187 pp.

Vogler, D. R., and T. D. Bruns. 1998. Phylogenetic relationships among the pine stem rust fungi (*Cronartium* and *Peridermium* spp.). Mycologia 90(2):244-257.

White, T. J., T. Bruns, S. Lee, and J. Taylor. 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. Pages 315-322 in M. A. Innis, D. H. Gelfand, J. J. Sninski, and T. J. White, (eds.). PCR Protocols : A Guide to Methods and Applications. Academic Press, San Diego, California.

Widder, F. 1941. Untersuchungen nber forstschSdliche *Cronartium*-arten. Sterreichischer Botanischer Zeitschrift 90:107-117.

Wilson, M., and D. M. Henderson. 1966. British rust fungi. Cambridge U.P., Cambridge, UK. 384 pp.

Yi, C. K., H. J. Kim, and Y. J. La. 1985. *Cronartium flaccidum* found on *Pinus* densiflora in Korea. Pages 149-161 in J. H. Ko and Y. J. La, (eds.). Proceedings of the Joint Conference IUFRO Working Party on Forest Gall Midges and Rusts of Pines. Korean Forestry Society, Suweon, Korea.

Zhuang, J. Y., and W. S. X. 2005. Urediniomycetes, Uredinales. Pages 233-290 in W. Y. Zhuang, (ed.). Fungi of Northwestern China. Mycotaxon Ltd, New York, USA.

Glossary

Scots Pine Blister Rust

Use this glossary to find the meaning of specialized words, abbreviations, acronyms, and terms used by PPQ–EDP. To locate where in the manual a given definition, term, or abbreviation is mentioned, refer to the index.

Definitions, Terms, and Abbreviations

amplicon. piece of DNA synthesized using amplification techniques such as PCR

anamorph. asexual form of a fungus

APA. American Phytopathological Society

APHIS. USDA–Animal and Plant Health Inspection Service

approved landfill. state licensed municipal or private landfill managed under state regulation to prevent leaching of potential pollutants into groundwater

autoecious. parasitic fungus that completes the entire life cycle on a single host

CAPS. Cooperative Agricultural Pest Survey Program, partnership between all 50 States and USDA to detect and monitor exotic pests of economic impact

chlorosis. yellowing of normally green tissue due to chlorophyll destruction in infected plants

CPB. U.S. Department of Homeland Security-Customs and Border Protection

CPHST. PPQ-Center for Plant Health Science and Technology

decontamination. application of approved chemical or other treatment to contaminated implements, material, or buildings for killing or deactivating a pathogen

detection survey. survey conducted in an environmentally favorable area where the pathogen is not known to occur

DHS. U.S. Department of Homeland Security

dieback. death of branches on woody plants, shrubs, trees; typically young shoots, twigs, and distal portions of branches die progressively toward older plant parts

disposal. method used to eliminate diseased plant material or material associated with diseased plant material, usually at approved landfill

EDP. PPQ-Emergency and Domestic Programs

EM. PPQ-Emergency Management

FIFRA. Federal Insecticide, Fungicide, and Rodenticide Act

ICS. Incident Command System

heteroecious. parasitic fungus that develops different stages of the life cycle on different host species

host. plant which is invaded by a parasite or pathogen and from which it obtains its nutrients

infection. establishment of a parasite on or within a host plant

ISIS. Integrated Survey Information System

macrocyclic. rust fungi that display a long life cycle with five stages, each with a characteristic type of spore
monitoring survey. survey conducted at a site where a disease was found and where an eradication program is being performed; also known as evaluation survey
NASS. National Agricultural Statistics Service
necrosis. dead or discolored plant tissue
NEPA. National Environmental Policy Act
NIS. PPQ-National Identification Service
NPAG. PPQ New Pest Advisory Group
NPRG. New Pest Response Guidelines
pathogen. organism that can incite disease
PCR. polymerase chain reaction, laboratory technique that amplifies DNA sequences in order to determine if host is infected with known pathogen
PCR-primers. short fragments of single stranded DNA (15 to 30 nucleotides in length), complementary to DNA sequences that flank the target region of interest; necessary components for the polymerase chain reaction
PERAL. Plant Epidemiology and Risk Analysis Laboratory
pest. insects, weeds, plant disease agents, and microorganisms
PPQ. APHIS-Plant Protection and Quarantine
SEL. USDA–ARS-Systematic Entomology Laboratory
SPHD. State Plant Health Director
SPRO. State Plant Regulatory Official
symptom. external and internal reactions or alterations of plant as the result of disease
teleomorph. sexual form of fungus
traceback. to investigate the origin of infested plants through intermediate steps in commercial distribution channels to the origin
trace-forward. to investigate where infected plants may have been distributed from a source through steps in commercial distribution channels
TWG. Technical Working Group
USDA. United States Department of Agriculture
USFWS. United States Fish and Wildlife Service

Appendix

A

Resources

Use *Appendix A Resources* to find the Web site addresses, street addresses, and telephone numbers of resources mentioned in the guidelines. To locate where in the guidelines a topic is mentioned, refer to the index.

Table A-1 Resources for Scots Pine Blister Rust (*Cronartium flaccidum* and *Peridermium pini*)

Resource	Contact Information
Center for Plant Health, Science, and Technology (USDA–APHIS–PPQ–CPHST)	http://www.aphis.usda.gov/plant_health/cphst/index.shtml
Emergency and Domestic Programs, Emergency Management (USDA–APHIS–PPQ–EDP–EM)	http://www.aphis.usda.gov/plant_health/plant_pest_info/index.shtml
PPQ *Manual for Agricultural Clearance*	http://www.aphis.usda.gov/import_export/plants/manuals/online_manuals.shtml
PPQ *Treatment Manual*	http://www.aphis.usda.gov/import_export/plants/manuals/online_manuals.shtml
Host or Risk Maps	http://www.nappfast.org/caps_pests/CAPs_Top_50.htm
Plant, Organism, and Soil Permits (APHIS–PPQ	http://www.aphis.usda.gov/plant_health/permits/index.shtml
National Program Manager for Native American Program Delivery and Tribal Liaison (USDA–APHIS–PPQ)	14082 S. Poston Place Tucson, AZ 85736 Telephone: (520) 822-544
Biological Control Coordinator (USDA–APHIS–CPHST)	http://www.aphis.usda.gov/plant_health/cphst/projects/arthropod-pests.shtml
FIFRA Coordinator (USDA–APHIS–PPQ–EDP)	4700 River Road Riverdale, MD 20737 Telephone: (301) 734-5861
Environmental Compliance Coordinator (USDA–APHIS–PPQ–EDP)	4700 River Road Riverdale, MD 20737 Telephone: (301) 734-7175
PPQ Form 391	http://www.aphis.usda.gov/library/forms/
List of State Plant Health Directors (SPHD)	http://www.aphis.usda.gov/services/report_pest_disease/report_pest_disease.shtml
List of State Plant Regulatory Officials (SPRO)	http://nationalplantboard.org/member/index.html
National Climatic Center, Data Base Administration, Box 34, Federal Building, Asheville, North Carolina 28801	http://www.ncdc.noaa.gov/oa/ncdc.html
CAPS Survey Manuals	http://caps.ceris.purdue.edu/
Leafhopper and treehopper genera in New Zealand	http://www1.dpi.nsw.gov.au/keys/leafhop/deltocephalinae/opsiini.htm
GenBank®	http://www.ncbi.nlm.nih.gov/
iPhyClassifier	http://plantpathology.ba.ars.usda.gov/cgi-bin/resource/iphyclassifier.cgi

Appendix

B

Forms

Use *Appendix B Forms* to learn how to complete the forms mentioned in the guidelines. To locate where in the guidelines a form is mentioned, refer to the index.

Contents

PPQ Form 391 Specimens For Determination

This report is authorized by law (7 U.S.C. 147a). While you are not required to respond your cooperation is needed to make an accurate record of plant pest conditions.

See reverse for additional OMB information.

FORM APPROVED
OMB NO. 0579-0010

U.S. DEPARTMENT OF AGRICULTURE
ANIMAL AND PLANT HEALTH INSPECTION SERVICE

SPECIMENS FOR DETERMINATION

Instructions: Type or print information requested. Press hard and print legibly when handwritten. Item 1 - assign number for each collection beginning with year, followed by collector's initials and collector's number. Example (collector, John J. Dingle): 83-JJD-001.
Pest Data Section – Complete Items 14, 15 and 16 or 19 or 20 and 21 as applicable. Complete Items 17 and 18 if a trap was used.

FOR IIBIII USE
LOT NO.
PRIORITY

1. COLLECTION NUMBER

2. DATE — MO | DA | YR

3. SUBMITTING AGENCY — ☐ State Cooperator ☐ PPQ ☐ Other ______

SENDER AND ORIGIN

4. NAME OF SENDER

6. ADDRESS OF SENDER

ZIP

INTERCEPTION SITE

5. TYPE OF PROPERTY *(Farm, Feedmill, Nursery, etc.)*

7. NAME AND ADDRESS OF PROPERTY OR OWNER

COUNTRY/ COUNTY

PURPOSE

8. REASON FOR IDENTIFICATION *("x" ALL Applicable Items)*

A. ☐ Biological Control (Target Pest Name)
B. ☐ Damaging Crops/Plants
C. ☐ Suspected Pest of Regulatory Concern *(Explain in* REMARKS*)*
D. ☐ Stored Product Pest
E. ☐ Livestock, Domestic Animal Pest
F. ☐ Possible Immigrant *(Explain in* REMARKS*)*
G. ☐ Survey *(Explain in* REMARKS*)*
H. ☐ Other *(Explain in* REMARKS*)*

9. IF PROMPT OR URGENT IDENTIFICATION IS REQUESTED, PLEASE PROVIDE A BRIEF EXPLANATION UNDER "REMARKS".

HOST DATA

10. HOST INFORMATION
NAME OF HOST *(Scientific name when possible)*

11. QUANTITY OF HOST
NUMBER OF ACRES/PLANTS
PLANTS AFFECTED *(Insert figure and indicate* ☐ Number ☐ Percent*):*

12. PLANT DISTRIBUTION
☐ LIMITED
☐ SCATTERED
☐ WIDESPREAD

13. PLANT PARTS AFFECTED
☐ Leaves, Upper Surface ☐ Trunk/Bark ☐ Bulbs, Tubers, Corms ☐ Seeds
☐ Leaves, Lower Surface ☐ Branches ☐ Buds
☐ Petiole ☐ Growing Tips ☐ Flowers
☐ Stem ☐ Roots ☐ Fruits or Nuts

PEST DATA

14. PEST DISTRIBUTION
☐ FEW
☐ COMMON
☐ ABUNDANT
☐ EXTREME

15. ☐ INSECTS ☐ NEMATODES ☐ MOLLUSKS

NUMBER SUBMITTED	LARVAE	PUPAE	ADULTS	CAST SKINS	EGGS	NYMPHS	JUVS.	CYSTS
ALIVE								
DEAD								

16. SAMPLING METHOD

17. TYPE OF TRAP AND LURE

18. TRAP NUMBER

19. PLANT PATHOLOGY – PLANT SYMPTOMS *("X" one and describe symptoms)*
☐ ISOLATED ☐ GENERAL

20. WEED DENSITY
☐ FEW ☐ SPOTTY ☐ GENERAL

21. WEED GROWTH STAGE
☐ SEEDLING ☐ VEGETATIVE ☐ FLOWERING/FRUITING ☐ MATURE

22. REMARKS

23. TENTATIVE DETERMINATION

24. DETERMINATION AND NOTES *(Not for Field Use)*

SIGNATURE

DATE

FOR IIBIII USE
DATE RECEIVED
NO.
LABEL
SORTED
PREPARED
DATE ACCEPTED
RR

PPQ FORM 391
(AUG 02)
Previous editions are obsolete.

This is a 6-Part form. Copies must be disseminated as follows:

☐ PART 1 – PPQ ☐ PART 2 – RETURN TO SUBMITTER AFTER IDENTIFICATION ☐ PART 3 – IIBIII OR FINAL IDENTIFIER
☐ PART 4 – INTERMEDIATE IDENTIFIER ☐ PART 5 – INTERMEDIATE IDENTIFIER ☐ PART 6 – RETAINED BY SUBMITTER

Figure B-1 Example of PPQ Form 391 Specimens For Determination, side 1

OMB Information

According to the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 0579-0010. The time required to complete this information collection is estimated to average .25 hours per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information.

Instructions

Use PPQ Form 391, Specimens for Determination, for domestic collections (warehouse inspections, local and individual collecting, special survey programs, export certification).

BLOCK	INSTRUCTIONS
1	1. Assign a number for each collection beginning the year, followed by the collector's initials and collector's number **EXAMPLE** In 2001, Brian K. Long collected his first specimen for determination of the year. His first collection number is 01-BLK-001 2. Enter the collection number
2	Enter date
3	Check block to indicate Agency submitting specimens for identification
4	Enter name of sender
5	Enter type of property specimen obtained from (farm, nursery, feedmill, etc.)
6	Enter address
7	Enter name and address of property owner
8A-8L	Check all appropriate blocks
9	Leave Blank
10	Enter scientific name of host, if possible
11	Enter quantity of host and plants affected
12	Check block to indicate distribution of plant
13	Check appropriate blocks to indicate plant parts affected
14	Check block to indicate pest distribution
15	• Check appropriate block to indicate type of specimen • Enter number specimens submitted under appropriate column
16	Enter sampling method
17	Enter type of trap and lure
18	Enter trap number
19	Enter X in block to indicate isolated or general plant symptoms
20	Enter X in appropriate block for weed density
21	Enter X in appropriate block for weed growth stage
22	Provide a brief explanation if Prompt or URGENT identification is requested
23	Enter a tentative determination if you made one
24	Leave blank

Distribution of PPQ Form 391

Distribute PPQ Form 391 as follows:
1. Send Original along with the sample to your Area Identifier.
2. Retain and file a copy for your records.

Figure B-2 Example of PPQ Form 391 Specimens For Determination, side 2

Purpose

Submit PPQ Form 391, Specimens for Determination, along with specimens sent for positive or negative identification.

Instructions

Follow the instructions in *Table B-1* on page B-5. Inspectors must provide all relevant collection information with samples. This information should be shared within a State and with the regional office program contact. If a sample tracking database is available at the time of the detection, please enter collection information in the system as soon as possible.

Distribution

Distribute PPQ Form 391 as follows:

1. Send the original along with the sample to your area identifier
2. Keep and file a copy for your records

Table B-1 Instructions for Completing PPQ Form 391, Specimens for Determination

Block	Description	Instructions
1	COLLECTION NUMBER	1. ASSIGN a collection number for each collection as follows: 2-letter State code–5-digit sample number (Survey Identification Number in Parentheses) Example: PA-1234 (04202010001) 2. CONTINUE consecutive numbering for each subsequent collection 3. ENTER the collection number
2	DATE	ENTER the date of the collection
3	SUBMITTING AGENCY	PLACE an X in the PPQ block
4	NAME OF SENDER	ENTER the sender's or collector's name
5	TYPE OF PROPERTY	ENTER the type of property where the specimen was collected (farm, feed mill, nursery, etc.)
6	ADDRESS OF SENDER	ENTER the sender's or collector's address
7	NAME AND ADDRESS OF PROPERTY OR OWNER	ENTER the name and address of the property where the specimen was collected
8A-8H	REASONS FOR IDENTIFICATION	PLACE an X in the correct block
9	IF PROMPT OR URGENT IDENTIFICATION IS REQUESTED, PLEASE GIVE A BRIEF EXPLANATION UNDER "REMARKS"	LEAVE blank; ENTER remarks in *Block 22*
10	HOST INFORMATION NAME OF HOST	If known, ENTER the scientific name of the host
11	QUANTITY OF HOST	If applicable, ENTER the number of acres planted with the host
12	PLANT DISTRIBUTION	PLACE an X in the applicable box
13	PLANT PARTS AFFECTED	PLACE an X in the applicable box
14	PEST DISTRIBUTION FEW/COMMON/ ABUNDANT/EXTREME	PLACE an X in the appropriate block
15	INSECTS/NEMATODES/ MOLLUSKS	PLACE an X in the applicable box to indicate type of specimen
	NUMBER SUBMITTED	ENTER the number of specimens submitted as ALIVE or DEAD under the appropriate stage
16	SAMPLING METHOD	ENTER the type of sample
17	TYPE OF TRAP AND LURE	ENTER the type of sample
18	TRAP NUMBER	ENTER the sample numbers
19	PLANT PATHOLOGY-PLANT SYMPTOMS	If applicable, check the appropriate box; otherwise LEAVE blank
20	WEED DENSITY	If applicable, check the appropriate box; otherwise LEAVE blank

Table B-1 Instructions for Completing PPQ Form 391, Specimens for Determination (continued)

Block	Description	Instructions
21	WEED GROWTH STAGE	If applicable, check the appropriate box; otherwise LEAVE blank
22	REMARKS	ENTER the name of the office or diagnostic laboratory forwarding the sample; include a contact name, email address, phone number of the contact; also include the date forwarded to the State diagnostic laboratory or USDA–APHIS–NIS
23	TENTATIVE DETERMINATION	ENTER the preliminary diagnosis
24	DETERMINATION AND NOTES (**Not** for Field Use)	LEAVE blank; will be completed by the official identifier

PPQ 523 Emergency Action Notification

According to the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information is 0579-0102. The time required to complete this information collection is estimated to average 1 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information.

FORM APPROVED - OMB NO. 0579-0102

U.S. DEPARTMENT OF AGRICULTURE
ANIMAL AND PLANT HEALTH INSPECTION SERVICE
PLANT PROTECTION AND QUARANTINE

EMERGENCY ACTION NOTIFICATION

SERIAL NO.

1. PPQ LOCATION
2. DATE ISSUED
3. NAME AND QUANTITY OF ARTICLE(S)
4. LOCATION OF ARTICLES
5. DESTINATION OF ARTICLES
6. SHIPPER
7. NAME OF CARRIER
8. SHIPMENT ID NO.(S)
9. OWNER/CONSIGNEE OF ARTICLES

   Name:

   Address:

   PHONE NO. FAX NO.

   SS NO. TAX ID NO.

10. PORT OF LADING
11. DATE OF ARRIVAL
12. ID OF PEST(S), NOXIOUS WEEDS, OR ARTICLE(S)

12a. PEST ID NO.

12b. DATE INTERCEPTED

13. COUNTRY OF ORIGIN
14. GROWER NO.
15. FOREIGN CERTIFICATE NO.

15a. PLACE ISSUED

15b. DATE

Under Sections 411, 412, and 414 of the Plant Protection Act (7 USC 7711, 7712, and 7714) and Sections 10404 through 10407 of the Animal Health Protection Act (7 USC 8303 through 8306), you are hereby notified, as owner or agent of the owner of said carrier, premises, and/or articles, to apply remedial measures for the pest(s), noxious weeds, and or article(s) specified in Item 12, in a manner satisfactory to and under the supervision of an Agriculture Officer. Remedial measures shall be in accordance with the action specified in Item 16 and shall be completed within the time specified in Item 17.

AFTER RECEIPT OF THIS NOTIFICATION, ARTICLES AND/OR CARRIERS HEREIN DESIGNATED MUST NOT BE MOVED EXCEPT AS DIRECTED BY AN AGRICULTURE OFFICER. THE LOCAL OFFICER MAY BE CONTACTED AT:

16. ACTION REQUIRED

- ☐ TREATMENT:
- ☐ RE-EXPORTATION:
- ☐ DESTRUCTION:
- ☐ OTHER:

Should the owner or owner's agent fail to comply with this order within the time specified below, USDA is authorized to recover from the owner or agent cost of any care, handling, application of remedial measures, disposal, or other action incurred in connection with the remedial action, destruction, or removal.

17. AFTER RECEIPT OF THIS NOTIFICATION COMPLETE SPECIFIED ACTION WITHIN (Specify No. Hours or No. Days):
18. SIGNATURE OF OFFICER:

ACKNOWLEDGMENT OF RECEIPT OF EMERGENCY ACTION NOTIFICATION

I hereby acknowledge receipt of the foregoing notification.

SIGNATURE AND TITLE: DATE AND TIME:

19. REVOCATION OF NOTIFICATION

ACTION TAKEN:

SIGNATURE OF OFFICER: DATE:

PPQ FORM 523 (JULY 2002) Previous editions are obsolete.

Figure B-3 Example of PPQ 523 Emergency Action Notification

Purpose

Issue a PPQ 523, Emergency Action Notification (EAN), to hold all host plant material at facilities that have the suspected plant material directly or indirectly connected to positive confirmations. Once an investigation determines the plant material is not infested, or testing determines there is no risk, the material may be released and the release documented on the EAN.

The EAN may also be issued to hold plant material in fields pending positive identification of suspect samples. When a decision to destroy plants is made, or in the case of submitted samples, once positive confirmation is received, the same EAN which placed plants on hold also is used to document any actions taken, such as destruction and disinfection. More action may be warranted in the case of other fields testing positive for this pest.

Instructions

If plant lots or shipments are held as separate units, issue separate EAN's for each unit of suspected plant material and associated material held. EAN's are issued under the authority of the Plant Protection Act of 2000 (statute 7 USC 7701-7758). States are advised to issue their own hold orders parallel to the EAN to ensure that plant material cannot move intrastate.

When using EAN's to hold articles, it is most important that the EAN language clearly specify actions to be taken. An EAN issued for positive testing and positive-associated plant material must clearly state that the material must be disposed of, or destroyed, and areas disinfected. Include language that these actions will take place at the owner's expense and will be supervised by a regulatory official. If the EAN is used to issue a hold order for further investigations and testing of potentially infested material, then document on the same EAN, any disposal, destruction, and disinfection orders resulting from investigations or testing.

Find more instructions for completing, using, and distributing this form in the PPQ *Manual for Agricultural Clearance*.

Appendix

C

How to Submit Plant Samples

Plant Samples for Plant Pathology Analysis

1. Sampling

   Please submit adequate amounts of suspect leaf material when possible. This helps ensure that there is sufficient material if downstream diagnostic techniques are required. Twelve or more leaves per sample are desired.

2. Storing

   Refrigerate samples while awaiting shipment to the diagnostic laboratory. Place leaves without paper towel in a sealed and labeled ziplock bag.

3. Documentation

   Each sample should be documented on, and accompanied by its own completed PPQ Form 391 'Specimens for Determination'. It is good practice to keep a partially filled electronic copy of this form on your computer with your address and other information filled out in the interest of saving time. Please make sure all fields that apply are filled out and the bottom field (block 24: Determination and Notes) is left blank to be completed by the Identifier. Include the phone number and/or e-mail address of the submitter. Other documentation in the form of notes, images, etc. can be sent along with this if it useful to the determination. It is important that there be a way to cross-reference the sample with the accompanying form. For example, write the "Collection Number" both on the Form 391 and on the sample bag.

4. Packing

   To provide extra insurance against accidental release during shipping, specimens should be double-bagged – i.e. first place the specimen in a self-locking plastic bag and then place that bag within a second self-locking plastic bag. **The Form 391 should not be placed in the bag holding the sample! Rather, it should be placed inside the outer bag**

Place double-bagged samples in a sturdy cardboard box or heavy styrofoam container so that the samples are not damaged during shipping and handling. Ideally, samples should be packed with freezer blocks or wet ice to maintain their integrity during the shipping process. Thoroughly seal all seams on the container with shipping tape.

5. Shipping

The Identifier Laboratory should be contacted prior to forwarding samples. It is helpful to know how many samples are being forwarded, what types of samples they are (e.g. SOD-suspect Camellia leaves), when the samples will be shipped, and the package tracking number. Label the shipping box as 'URGENT' and send via overnight express courier (FedEx, UPS, Airborne, DHL, etc) to the appropriate Identifier.

Appendix

D Taxonomic Support for Surveys

Contents

Background

The National Identification Services (NIS) coordinates the identification of plant pests in support of USDA's regulatory programs. Accurate and timely identifications are the foundation of quarantine action decisions and are essential in the effort to safeguard the nation's agricultural and natural resources.

NIS employs and collaborates with scientists who specialize in various plant pest groups, including weeds, insects, mites, mollusks and plant diseases. These scientists are stationed at a variety of institutions around the country, including federal research laboratories, plant inspection stations, land-grant universities, and natural history museums. Additionally, the NIS Molecular Diagnostics Laboratory is responsible for providing biochemical testing services in support of the agency's pest monitoring programs.

On June 13, 2007, the PPQ Deputy Administrator issued PPQ Policy No. PPQ-DA-2007-02 which established the role of PPQ NIS as the point of contact for all domestically- detected, introduced plant pest confirmations and communications. A Domestic Diagnostics Coordinator (DDS) position was established to administer the policy and coordinate domestic diagnostic needs for NIS. This position was filled in October of 2007 by Joel Floyd (USDA, APHIS, PPQ-PSPI,NIS 4700 River Rd., Unit 52, Riverdale, MD 20737, phone (301) 734-4396, fax (301) 734-5276, e-mail: joel.p.floyd@aphis.usda.gov).

Taxonomic Support and Survey Activity

Taxonomic support for pest surveillance is basic to conducting quality surveys. A misidentification or incorrectly screened target pest can mean a missed opportunity for early detection when control strategies would be more viable and cost effective. The importance of good sorting, screening, and identifications in our domestic survey activity cannot be overemphasized.

Fortunately most states have, or have access to, good taxonomic support within their states. Taxonomic support should be accounted for in cooperative agreements as another cost of conducting surveys. Taxonomists and laboratories within the State often may require supplies, develop training materials, or need to hire technicians to meet the needs of screening and identification. As well, when considering whether to survey for a particular pest a given year, consider the challenges of taxonomic support.

Sorting and Screening

For survey activity, samples that are properly sorted and screened before being examined by an identifier will result in quicker turn around times for identification.

Sorting

Sorting is the first level of activity that assures samples submitted are of the correct target group of pests being surveyed, that is, after removal of debris, ensure that the correct order, or in some cases family, of insects is submitted; or for plant disease survey samples, select those that are symptomatic if appropriate. There should be a minimum level of sorting expected of surveyors depending on the target group, training, experience, or demonstrated ability.

Screening

Screening is a higher level of discrimination of samples such that the suspect target pests are separated from the known non-target, or native species of similar taxa. For example, only the suspect target species or those that appear similar to the target species are forwarded to an identifier for confirmation. There can be first level screening and second level depending on the difficulty and complexity of the group. Again, the degree of screening appropriate is dependent on the target group, training, experience, and demonstrated ability of the screener.

Check individual survey protocols to determine if samples should be sorted, screened or sent entire (raw) before submitting for identification. If not specified in the protocol, assume that samples should be sorted at some level.

Resources for Sorting, Screening, and Identification

Sorting, screening, and identification resources and aids useful to CAPS and PPQ surveys are best developed by taxonomists who are knowledgeable of the taxa that includes the target pests and the established or native organisms in the same group that are likely to be in samples and can be confused with the target. Many times these aids can be regionally based. They can be in the form of dichotomous keys, picture guides, or reference collections. NIS encourages the development of these resources, and when aids are complete, post them in the CAPS Web site so others can benefit. If local screening aids are developed, please notify Joel Floyd, the Domestic Diagnostics Coordinator, as to their availability. Please see the following for some screening aids available: http://pest.ceris.purdue.edu/caps/screening.php

Other Entities for Taxonomic Assistance in Surveys

When taxonomic support within a state is not adequate for a particular survey, in some cases other entities may assist including PPQ identifiers, universities and state departments of agriculture in other states, and independent institutions. Check with the PPQ regional CAPS coordinators about the availability of taxonomic assistance.

Universities and State Departments of Agriculture

Depending on the taxonomic group, there are a few cases where these two entities are interested in receiving samples from other states. Arrangements for payment, if required for these taxonomic services, can be made through cooperative agreements. The National Plant Diagnostic Network (NPDN) also has five hubs that can provide service identifications of plant diseases in their respective regions.

Independent Institutions

The Eastern Region PPQ office has set up multi-state arrangements for Carnegie Museum of Natural History to identify insects from trap samples. They prefer to receive unscreened material and work on a fee basis per sample.

PPQ Port Identifiers

There are over 70 identifiers in PPQ that are stationed at ports of entry who primarily identify pests encountered in international commerce including conveyances, imported cargo, passenger baggage, and propagative material. In some cases, these identifiers process survey samples generated in PPQ conducted surveys, and occasionally from CAPS surveys. They can also enter into our Pest ID database the PPQ form 391 for suspect CAPS target or other suspect new pests, prior to being forwarded for confirmation by an NIS recognized authority.

PPQ Domestic Identifiers

PPQ also has a limited number of domestic identifiers (three entomologists and two plant pathologists) normally stationed at universities who are primarily responsible for survey samples. Domestic identifiers can be used to handle unscreened, or partially screened samples, with prior arrangement through the PPQ regional survey coordinator. They can also as an intermediary alternative to sending an unknown suspect to, for example, the ARS Systematic Entomology Lab (SEL), depending on their specialty and area of coverage.

They can also enter into our Pest ID database the PPQ form 391 for suspect CAPS target or other suspect new pests, prior to being forwarded for confirmation by an NIS recognized authority.

PPQ Domestic Identifiers
Bobby Brown
Domestic Entomology Identifier
Specialty: forest pests (coleopteran, hymenoptera)
Area of coverage: primarily Eastern Region

USDA, APHIS, PPQ
901 W. State Street
Smith Hall, Purdue University
Lafayette, IN 47907-2089
Phone: 765-496-9673
Fax: 765-494-0420
e-mail: robert.c.brown@aphis.usda.gov

Julieta Brambila
Domestic Entomology Identifier
Specialty: adult Lepidoptera, Hemiptera
Area of Coverage: primarily Eastern Region
USDA APHIS PPQ
P.O. Box 147100
Gainesville, FL 32614-7100
Office phone: 352- 372-3505 ext. 438, 182
Fax: 352-334-1729
e-mail: julieta.bramila@aphis.usda.gov

Kira Zhaurova
Domestic Entomology Identifier
Specialty: to be determine
Area of Coverage: primarily Western Region
USDA, APHIS, PPQ
Minnie Belle Heep 216D
2475 TAMU
College Station, TX 77843
Phone: 979-450-5492
e-mail: kira.zhaurova@aphis.usda.gov

Grace O'Keefe
Domestic Plant Pathology Identifier
Specialty: Molecular diagnostics (citrus greening, P. ramorum, bacteriology, cyst nematode screening)
Area of Coverage: primarily Eastern Region

USDA, APHIS, PPQ
105 Buckhout Lab
Penn State University
University Park, PA 16802
Lab: 814 - 865 - 9896
Cell: 814 – 450- 7186
Fax: 814 - 863 – 8265
e-mail: grace.okeefe@aphis.usda.gov

Craig A. Webb, Ph.D.
Domestic Plant Pathology Identifier
Specialty: Molecular diagnostics (citrus greening, P. ramorum, cyst nematode screening)
Area of Coverage: primarily Western Region
USDA, APHIS, PPQ
Department of Plant Pathology
Kansas State University
4024 Throckmorton Plant Sciences
Manhattan, KS 66506-5502
Cell (785) 633-9117
Office (785) 532-1349
Fax: 785-532-5692
e-mail: craig.a.webb@aphis.usda.gov

Final Confirmations

If identifiers or laboratories at the state, university, or institution level suspect they have detected a CAPS target, a plant pest new to the United States, or a quarantine pest of limited distribution in a new state, the specimens should be forwarded to an NIS recognized taxonomic authority for final confirmation. State cooperator and university taxonomists can go through a PPQ area identifier or the appropriate domestic identifier that covers their area to get the specimen in the PPQ system (for those identifiers, see table G-1-1 in the Agriculture Clearance Manual, Appendix G link below). They will then send it to the NIS recognized authority for that taxonomic group.

State level taxonomists, who are reasonably sure they have a new United States. record, CAPS target, or new federal quarantine pest, can send the specimen directly to the NIS recognized authority, but must notify their State Survey Coordinator (SSC), PPQ Pest Survey Specialist (PSS), State Plant Health Director (SPHD), and State Plant Regulatory Official (SPRO).

Before forwarding these suspect specimens to identifiers or for confirmation by the NIS recognized authority, please complete a PPQ form 391 with the tentative determination. Also fax a copy of the completed PPQ Form 391 to

"Attention: Domestic Diagnostics Coordinator" at 301-734-5276, or send a PDF file in an e-mail to mailto:nis.urgents@aphis.usda.govwith the overnight carrier tracking number.

The addresses of NIS recognized authorities of where suspect specimens are to be sent can be found in The Agriculture Clearance Manual, Appendix G, tables G-1-4 and G-1-5: http://www.aphis.usda.gov/import_export/plants/manuals/ports/downloads/mac_pdf/g_app_identifiers.pdf

Only use Table G-1-4, the "Urgent" listings, for suspected new United States records, or state record of a significant pest, and Table G-1-5, the "Prompt" listings, for all others.

When the specimen is being forwarded to a specialist for NIS confirmation, use an overnight carrier, insure it is properly and securely packaged, and include the hard copy of the PPQ form 391 marked "Urgent" if it is a suspect new pest, or "Prompt" as above.

Please contact Joel Floyd, the Domestic Diagnostics Coordinator if you have questions about a particular sample routing, at phone number: 301-734-5276, or e-mail: joel.p.floyd@aphis.usda.gov

Digital Images for Confirmation of Domestic Detections

For the above confirmations, do not send digital images for confirmation. Send specimens in these instances. For entry into NAPIS, digital imaging confirmations can be used for new county records for widespread pests by state taxonomists or identifiers if they approve it first. They always have the prerogative to request the specimens be sent.

Communications of Results

If no suspect CAPS target, program pests, or new detections are found, communication of these identification results can be made by domestic identifiers or taxonomists at other institutions directly back to the submitter. They can be in spread sheet form, on hard copy PPQ form 391's, or other informal means with the species found, or "no CAPS target or new suspect pest species found". Good record keeping by the intermediate taxonomists performing these identifications is essential.

All confirmations received from NIS recognized authorities, positive or negative, are communicated by NIS to the PPQ Emergency and Domestic Programs (EDP) staff in PPQ headquarters. EDP then notifies the appropriate PPQ program managers and the SPHD and SPRO simultaneously. One of these contacts should forward the results to the originating laboratory, diagnostician, or identifier.

Data Entry

Cooperative Agricultural Pest Survey (CAPS)

For survey data entered into NAPIS, new country and state records should be confirmed by an NIS recognized authority, while for others that are more widespread, use the identifications from PPQ identifiers or state taxonomists.

Appendix

E

Research Needs

1. Conduct phylogeographic analysis of Scots pine blister rust across its native range relative to its genetic diversity.
2. Determine the host range and environmental requirements for each genetic group of Scots pine blister rust.
3. Conduct phylogenetic analysis of known and potential hosts of Scots pine blister rust.
4. Develop prediction models of potential spread of Scots pine blister rust based on distribution of aecial and telial hosts combined with present and future climate models.
5. Conduct phylogenetic analysis of Scots pine blister rust and other pine stem rusts present in North America (e.g., *Cronartium arizonicum*, *C. coleosporioides, C. comandrae, C. ribicola*, and *Peridermium harknessii*).
6. Use inoculation tests in infested countries to determine potential aecial and telial hosts of Scots pine blister rust in North America.
7. Assess potential sources of resistance in aecial host populations of North America.
8. Begin assessments of potential biological control agents.
9. Evaluate potential genetic information and assess status of germplasm collections for diverse geographic sources of potential pine hosts in North America.
10. Establish sentinel tree plantings in areas with Scots pine blister rust to establish a baseline for genetic screening.
11. Expand current knowledge on pathogenicity, host range and susceptibility and factors affecting disease development.