Scots stem pine rust - *Cronartium flaccidum*

*Cronartium flaccidum* is a heteroecious rust fungus, completing different stages of its life cycle on different plants. The spermogonial and aecial stages occurs on species of hard or two-needled pines, and the uredinial and telial stages on the leaves of herbaceous species in dicotyledonous families such as Asclepiadaceae, Paeoniaceae, and Scrophulariaceae. Mating of haploid strains occurs on species of *Pinus*, followed by production of aeciospores. An asexual stage producing urediniospores occurs on the dicot plants, followed by production of teliospores, the sexual stage, that germinate to form basidiospores that infect pines thus completing the cycle. A closely related autoecious rust, *Endocronartium* (*Peridermium*) *pini*, infects only *Pinus* hosts.

*Cronartium flaccidum* (Alb. & Schw.) Wint.

*Cronartium flaccidum* is known from Europe and parts of northern and eastern Asia in the Northern Hemisphere (Smith et al., 1988; CABI, 1989). The autoecious form, *Endocronartium 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. Azbukina (2008) expected to find *Endocronartium pini* in Siberia but did not. It is a Regulated Pest for the United States (USDA/APHIS, 2008). 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 overlooked, such that accidental introduction of the rust could occur through importation of conifer seedlings or trees.

Spermogonia caulicolous, on cankers, under bark, flat, yellow, turning brown, exuding spermatia in orange droplets.

Aecia caulicolous, blister-like, 2-7 mm diam, single or confluent; peridium several cells thick, white, peridial cells rhomboid-ellipsoid, walls thick, verrucose. Aeciospores in chains, globose, ovoid to ellipsoid, or polygonal, verrucose except for a smooth area, 21-36 x 14-24 µm, orange-yellow; wall hyaline, thick, 2-4 µm; warts 1-2 µm high.

Uredinia hypophyllous, scattered or in groups, blister-like, 100-300 µm diam. Peridium hemispherical, opening by a central pore. Urediniospores single on pedicel, ellipsoid to ovoid, sparsely echinulate, 18-30 x 11-22 µm, yellow; wall hyaline, 1.5-2.5 µm thick.

Telia hypophyllous, often developing in uredinia, erumpent; teliospores in chains, adhering in columns to 2 mm or more long, pale orange to cinnamon brown. Teliospores ellipsoid to cylindrical, not separating, 20-64 x 6-16 µm, smooth, with yellowish wall 1-3 µm thick.

Basidiospores globose-subglobose, smooth, 4-12 µm diam, their production giving a whitish appearance to upper ends of telia.

For a more detailed description, see Mordue and Gibson (1978), Ragazzi et al. (1987), and Kaitera et al. (1999b).

See also Kuprevich and Tranzschel (1957), Hiratsuka (1969), and Kasanen (1997).

**Notes:** The diverse interactions of this heteroecious fungus with its hosts has resulted in a complicated nomenclature. Several species of *Cronartium* were initially identified for the rust on the various dicotyledons 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).

Wilson and Henderson (1966) do not identify any morphological differences between the aecial stages of the two species. Hiratsuka (1968) reported that aeciospores of European isolates of E. pini germinated to produce, for the most part, determinate, septate, usually binucleate, but frequently uninucleate, germ tubes, while the aeciospores of *C. flaccidum* produced indeterminate, aseptate germ tubes that are more frequently binucleate. Subsequently, Gibbs et al. (1988), Kasanen (1997), and Kaitera et al. (1999b).
Another *Peridermium* species, *P. pini* (Wild.:Pers.) Lev., was found to be autoecious, living only on pines. A separate *Cronartium* name was established for it, but this species came to be viewed, instead, as a race of *C. flaccidum* (Wilson and Henderson, 1966; Hiratsuka, 1968). Hiratsuka (1969) created the genus, *EndoCronartium*, for such autoecious forms of *Cronartium*-related rusts. This genus units forms that are more closely related to distinct species of *Cronartium* than to each other (Vogler and Bruns, 1998). A number of studies have elucidated the close genetic relationship between the heteroecious *C. flaccidum* and the autoecious *Peridermium pini* (Moricca et al., 1996; Moricca and Ragazzi, 1998; Vogler and Bruns, 1998; Kasanen et al., 2000; Hautala et al., 2002) as well as the difficulty of separating them on a morphological basis (Gibbs et al., 1988; Kasanen, 1997, Kaitera et al., 1999b).

**Similarities to Other Species:** Hautala et al. (2002) showed that *Cronartium flaccidum* and the autoecious rust *EndoCronartium pini* are genetically almost indistinguishable despite differences in life cycle. The two cannot be distinguished by aeciospore morphology (Kasanen, 1997; Kaitera et al., 1999b). Inoculation tests on the dicot alternate hosts should establish the difference (Kaitera, 1999; Kaitera et al., 1999a; Kaitera and Nuorteva, 2008, but see Gibbs et al. 1988). Moricca and Ragazzi (1998) identified an RFLP technique that distinguished the two forms on the basis of one electrophoretic band resulting from Hinf1 restriction endonuclease digestion of the IGS1 region of rDNA.

In North America, there are at least eleven species of *Cronartium* including *Cronartium ribicola* J.C. Fisch. and six species of *Peridermium* that infect trees in the genus *Pinus* (Sinclair and Lyon, 2005). To a certain extent, these can be distinguished by 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 stem symptoms at all (Sinclair and Lyon, 2005). Although some of the common Eurasian hosts have been naturalized e.g., *Pinus sylvestris* (USDA-NCRS, 2009), tests indicate that the three-needed pines native to North America are not susceptible to *C. flaccidum* (Raddi and Fagnini, 1978; Kaitera and Nuorteva, 2008). *Cronartium comandrae* Peck, a widespread North American pine stem rust that also infects introduced two-needed species, also hosts for *C. flaccidum*, produces unique tear-drop-shaped aeciospores on pine (Sinclair and Lyon, 2005).

Other European rusts that can attack pines have a heteroecious life cycle similar to that of *C. flaccidum*, but usually infect different alternate hosts. *Coleosporium tussilaginis* (Pers.) Lev., the pine needle rust, shares a few telial hosts with the blister rust, but produces its spermospore and aecia on pine needles, not on stems (Smith et al., 1988). Also, teliospores of this rust on species of *Melampyrum* are single and cylindrical, produced not in long columns but in waxy crusts (Wilson and Henderson, 1966). *Melampsora populea* (Pers.) P. Karst infects the shoots of two-needed pines, causing shoot bending and/or tip death (Smith et al., 1988). Its linear aecia lack a peridium and the aeciospores are significantly smaller than those of *C. flaccidum* (Wilson and Henderson, 1966).

Although their aeciospores cannot be distinguished even at the level of scanning electron microscopy (Kasanen, 1997), the invasive species *C. ribicola* does not infect the primary hosts of *C. flaccidum* in Europe (Kaitera and Nuorteva, 2006) or the same species of pines, since it is restricted to 5-needled (soft) pines (Sinclair and Lyon, 2005). Butin (1995) notes that *C. flaccidum* tends to infect at the top of a tree rather than at the base of stem or of lower branches, where *C. ribicola* infection is usually found.

**Detection and Inspection Methods:** The delay of sporulation for months or years after infection of pines (Wilson and Henderson, 1966) reduces the reliability of single inspections of seedlings and young trees to detect this fungus. Spots may or may not result from infection of needles (Raddi et al., 1979; Ragazzi et al., 1986). Infection through wounded stems is also possible (Wilson and Henderson, 1966). Established infections produce cankers with blister-like aecia on stems or bark-bearing wood. In the field, nearby herbaceous alternate hosts can be examined for the presence of uredinia and/or bristle-like telial columns on the undersides of leaves (Kaitera et al., 2005).

**Diagnostic Method:** PCR-amplified fragments of two regions of rDNA can be used in RFLP and SSCP analysis to distinguish alternating from non-alternating isolates of pine blister rust (Moricca and Ragazzi, 1998). Sequences for several regions of rDNA, particularly those for the 5.8s rRNA examined by Moricca et al. (1996), are currently available in GenBank for comparison (NCBI, 2009).

**Notes on Habitat:** Moricca and Ragazzi (1996) found that *C. flaccidum* isolates had different cultural morphologies in different regions of Italy. Likewise, Gibbs et al. (1988) determined that *P. pini* isolates from different parts of Great Britain have different culture morphologies. Kaitera et al. (1999a; 2005) identify *C. flaccidum* as occurring in southern Finland, while *E. pini* is found throughout the country. Nevertheless, the two forms can occur in the same or adjacent stands of *P. sylvestris* (Kaitera et al., 1999a).
Risk of Introduction: 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, 1996). Single-aged monoculture populations could suffer epidemics, in particular, if trees are at a susceptible age and the autoecious Endocronartium 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, 2009). Other alternate hosts (Gentiana spp., Paeonia spp.) may be introduced as ornamentals and be grown near introduced ornamental pines.

North America has a significant number of stem rusts of the genera Cronartium, Endocronartium and Peridermium on its native species of three-needled pines (Sinclair and Lyon, 2005), which are in a different section of the genus Pinus from the European two-needled hosts (USDA-ARS, 2009). The ecological situation into which C. flaccidum might invade is therefore complex. Some evidence exists of resistance in the native pines (Raddi and Fagnini, 1978; Kaitera and Nuorteva, 2008). Nevertheless, native or introduced primary (telial) hosts for C. flaccidum are present in North America (USDA-ARS, 2009). Among the USDA/APHIS Regulated Plant Pests, Rossman et al. (2006) place this species in the category of Threat to major crop plants and forest trees. 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 (USDA/APHIS, 2008; Canadian Food Inspection Agency, 2009).

Introduction of Endocronartium pini 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.

Notes on Crops/Other Plants Affected: The commercially valuable aecial hosts of C. flaccidum, specifically two-needled pines of Europe and Asia, vary in susceptibility (Raddi and Fagnini, 1978; Kaitera and Nuorteva 2008). Scots pine, P. sylvestris, is the common host in northern Europe, but appears to be less susceptible than the Mediterranean species when tested in southern Europe (Raddi and Fagnini, 1978). The North American three-needled pines tested were found to be relatively resistant. Less is known about the pine species reported as hosts in China (Tai, 1979; Teng, 1996; Cao et al., 2000; Chen, 2002; Zhuang and Wei, 2005), Japan (Kobayashi et al., 2007), Korea (Cho and Shin, 2004) and Taiwan (Hiratsuksa and Chen, 1991). Raddi and Fagnini (1978) did not observe spotting on needles of young plants of the Asian species P. densiflora, P. massoniana and P. tabuliformis that were exposed to natural basidiospore inoculation in Italy; these species appeared to be resistant.

The telial hosts of C. flaccidum are herbaceous dicotyledonous plants in at least ten families, as demonstrated by inoculation tests (Wilson and Henderson, 1966; USDA/APHIS, 2008). Some of the species are not native to the geographic range of the rust, and infection was observed in gardens (Goncalves da Cunha, 1936; Gjaerum, 1974; BPI, 2009). Specialized forms referred to as formae speciales varying with respect to pathogenicity to various dicot hosts have been identified in Europe (Smith et al., 1988), but inoculation tests (Kaitera and Hantula, 1998; Kaitera, 1999; Kaitera et al., 1999a) have indicated that C. flaccidum has low host specificity. Although differences in host susceptibility within a plant genus were noted (Kaitera, 1999; Kaitera et al, 1999a), the broad host range of C. flaccidum suggests that additional host species are likely to be susceptible. Thus, the rust was recently reported from Paeonia daurica in Ukraine (Dudka et al., 2004) and Siphonostegia chinensis in China (Zhuang and Wei, 2005).

Symptoms/Description: Infection of pine needles may or may not cause yellow-red spots on needles (Ragazzi et al., 1986; Smith et al., 1988); spots may appear only after some months (Ragazzi, 1989). Spermogonia appear within one or two years (Ragazzi, 1989) or later on older trees (Smith et al., 1988). The fungus grows into the shoots, which become swollen, and aecia are produced after one or two additional years (Grieg, 1987). Cankers on stems also bear blister-like pustules, containing orange-yellow spores, especially at bases of branch whorls (Butin, 1995), and the perennial growth of the fungus results in concentric zones of sporulation. Stems are flattened and deformed (Butin, 1995). Resin is produced from the cankers and in the wood underlying it. If cankers on the trunk grow large enough or separate cankers meet, girdling of the stem may result in death of the top or of the entire tree (Smith et al., 1988). The Resin top name of the disease derives from the combined death of the tree top with copious visible Resin generation. However, years may be required for infections to have this effect (Grieg, 1987). On Pinus sylvestris in Finland, Kaitera (2000) found that the average size of lesions bearing aecia on three-to twenty-years-old stems was about 4 cm in length.

Life Cycle: Teliospores in bristle-like telia on dicot primary host leaves germinate in spring to early summer and produce basidiospores that infect the young pine needles through stomates (Mordue and Gibson, 1978; Ragazzi et al., 1987). Mycelium grows through the needles into the cambium of stems and into the rays (Butin, 1995). Haploid spermogonia are the mating organs produced on shoots within one or two years; after fertilization, aecia are produced on the canker tissues during the summer of the next year or the year after that (Smith et al., 1988). Aeciospores are wind-blown to young leaves of herbaceous
distances than the basidiospores, which may be limited to less than 500 meters (Hunt, 1997). Aeciospores and urediniospores may be disseminated greater

On primary hosts, scattered small chlorotic or necrotic spots appear on the upper side of leaves, with orange uredinia and, later, bristle-like yellowish to red-brown telia below (Wilson and Henderson, 1966; Ragazzi et al., 1987; Kaitera, 1999; Kaitera and Nuorteva, 2006). Spots are sometimes limited by veins (Ragazzi et al., 1987).

In the autoecious Endocronartium pini, the spores produced by aecia are capable of infecting pine directly, without an intervening telial stage. When free water is present, aeciospores germinate on needles and infect through stomates (Van der Kamp, 1970). Wounding of the stem can enhance infection (Wilson and Henderson, 1966; Gibbs et al. 1988), and infections through stem wounds are more damaging (Van der Kamp, 1970).

Genetics: Moricca et al. (1996) identified three regions of DNA with high similarity in C. flaccidum and E. pini isolates from Europe. The 5.8s ribosomal gene sequence was highly conserved, but some heterogeneity appeared in the ITS1 and ITS2 regions. Nevertheless, the same sequence variants occurred in isolates of both fungi. Moricca and Ragazzi (1998) observed virtually identical RFLP profiles, again suggesting a high degree of affinity between the two fungi. A diagnostic difference between them, observed in single-strand conformation polymorphism (SSCP) analysis of a highly variable region, was of a type found in genetic analyses of differences within a species.

Hantula et al. (1998) also observed a high degree of similarity in two genetic regions in European C. flaccidum and E. pini. Results of additional tests using the two markers led Hantula et al. (2002) to consider the two forms to be the same species. For one highly variable marker, differentiation was lower between the two rusts than within populations of each form.

Reproductive biology: The two rusts are separated on the basis of their reproduction, since E. pini is autoecious, lacking the stage of sexual recombination on any primary host (Wilson and Henderson, 1966; Hantula et al., 2002). On the other hand, Gibbs et al. (1988) reported infection of a dicot host in vitro, with production of telia, by isolates considered to be the autoecious form. This suggests that E. pini may be "facultatively heteroecious", although Hantula et al. (2002) consider this unlikely. Kasanen et al. (2000) found support for E. pini as homothallic, if not clonal, while allele distribution among aecia of C. flaccidum provided evidence of both heterothallic and homothallic matings.

The spermatonia of E. pini appear to be self-fertilizing or non-functional (Hantula et al., 2002). Spermatogonial fluid was observed on only a few infected plants in tests in Finland (Kaitera and Nuorteva, 2008).

Physiology and phenology: Three specialized types of Cronartium flaccidum in Europe have been proposed depending on the telial host genera and varying in geographic distribution (Smith et al., 1988). Scientific attention has focused on f. sp. typica in western Europe. Kaitera (1999) identified new primary hosts in the genus Melampyrum (Scrophulariaceae, now Orobanchaceae) infected by C. flaccidum in Finland; their susceptibility to the other two formae speciales was not examined.

The pine blister rust organisms also exhibit variations in morphology in culture. Pei and Gibbs (1991) found two morphological types of colonies derived from aeciospores that corresponded to two regions of E. pini in Great Britain. Likewise, Moricca and Ragazzi (1996) observed two groupings of C. flaccidum isolates with different cultural characters that were correlated to their geographic origins in Italy.

Mittempergher and Raddi (1977) observed differences in regional sources of C. flaccidum in Italy. A montane isolate was more virulent on Pinus nigra from the same region than were isolates from other habitats. Endocronartium pini spore collections from different areas of Finland also differ in virulence on the pine host, but no significant differences were observed among three C. flaccidum collections (Kaitera and Nuorteva, 2008).

Associations: Insect may play a role in mating in C. flaccidum based on the similarity of its life cycle to that of Cronartium ribicola (Mordue and Gibson, 1978; Smith et al., 1988). In that species, Insects are attracted to sweet liquid produced from spermatogonia and appear to promote fertilization by carrying spermatia between them (Hunt, 1997).

Endocronartium pini is parasitized by the conidial fungus Tuberculina maxima (Wilson and Henderson, 1966; Gibbs et al., 1987), but that does not substantially affect losses due to rust (Moricca et al., 2001). Growth on the aecia apparently reduces sporulation rather than lesion expansion (Van der Kamp, 1970).

Natural dispersal: Rust aeciospores, urediniospores and sporidia (basidiospores) are distributed by wind (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).

**Vector transmission:** An association of *Endocronartium pini* with pine top weevils may involve transmission, but results were inconclusive (Pappinen and von Weissenberg, 1994).

**Accidental introduction:** Introduction of either form of the rust would be possible if seedlings or young trees were transported while the systemic infections were latent (USDA/APHIS, 2008).

**Economic impact:** The fungus generally does not have a major impact on forest management in the United Kingdom (Gibbs, 1997). On the other hand, whole plantations were destroyed in Italy during the 1960s due to the presence of new plantings and favorable conditions (Smith et al., 1988; Ragazzi, 1989). According to Martinsson and Nilsson (1987), *C. flaccidum* rust reduced radial stem increment by 40-70% in severe attacks and by 20-40% in minor attacks on *Pinus sylvestris* in Sweden. Kaitera et al. (1994) estimated that disease caused by *E. pini* in Finland reduced the market value of saw timber trees by 18% and pulpwod trees by 3% as a result of stem lesions, and by 15% and 14%, respectively, due to the death of tree tops.

**Prevention:** 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, 2009: 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* (Mordue and Gibson, 1978; Butin, 1995). 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).

**Biological control:** 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%.

**Host resistance:** 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 Fagnini (1978) used three methods to inoculate seedlings and young plants of different pine species with basidiospores of *C. 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 *P. pinaster*.

Although *P. sylvestris* appeared resistant in the limited tests in Italy (Raddi and Fagnini, 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 (2008) tested seedlings 1-7 years old with aeciospores of *E. pini* and basidiospores of *C. 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 *P. 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 (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.

**References**


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Aecia on twig of *Pinus sylvestris* 7.5×. BPI 032637. BPI 032637

Telia on underside of leaf of *Paeonia officinalis* 10×. BPI 032592. BPI 032592

Uredinia on underside of leaf of *Paeonia* sp. 10×. BPI 032563. BPI 032563

Telia on underside of leaf of *Vincetoxicum hirundinaria* 25×. BPI 032650. BPI 032650

Aeciospores 400×. BPI 032637. BPI 032637

Aeciospore 1000×. BPI 032637. BPI 032637
Urediniospores 400×. BPI 032563. BPI 032563

Teliospores in telial column 400×. BPI 032649. BPI 032649

Aecia of *Endocronartium pini* on twig of *Pinus sylvestris* 7.5×. BPI 154221. BPI 154221

Urediniospores 1000×. BPI 032563. BPI 032563

Teliospores in telial columns, with basidiospores 200×. BPI 032649. BPI 032649