Witches' Broom Disease of Black Raspberry in Oregon

R. H. CONVERSE, Research Plant Pathologist, U.S. Department of Agriculture, Agricultural Research Service, Oregon State University, Corvallis 97331; R. G. CLARKE, formerly Assistant Professor of Entomology, Department of Entomology, Oregon State University; P. W. OMAN, SR., Professor Emeritus, Department of Entomology, Oregon State University; and G. M. MILBRATH, Plant Pathologist, Oregon Department of Agriculture, Salem 97310

ABSTRACT

A lethal disease that caused witches' broom of Munger black raspberry was found in a commercial planting east of Portland, OR, in 1980 and 1981. The disease appeared to spread rapidly in the field, eventually killing about 75% of the plants. Nearby Willamette red raspberry and wild Rubus spp. in and near the fields of infected Munger appeared to be unaffected. Mycoplasmalike bodies were common in sieve tubes of petioles from infected plants but were absent from symptomless Munger plants. The leafhopper Macrosipis fuscata (Zett.), a vector of Rubus stunts mycoplasmata in Europe, was common on wild R. laciniatus in and near the fields of infected Munger. Attempts to transmit black raspberry witches' broom disease by Macrosipis fuscata or by leaf graft or to find additional infected fields were unsuccessful. Antiserum against Spiroplasma citri failed to give positive reactions in enzyme-linked immunosorbent assay serological tests against leaf sap from black raspberry diseased with witches' broom.

Witches' brooming is an unusual condition in Rubus in North America. The few reported instances of witches' brooming or shoot proliferation in Rubus spp. in North America were summarized by van der Meer and de Flitter (8). A disease known as Rubus stunt, presumably caused by a mycoplasmalike organism and transmitted by several leafhoppers, including Macrosipis fuscata (Zett.), is common in Europe (4,5,8). M. fuscata was first reported in North America by Tonks in British Columbia (7) and was found in wild R. procerus P. J. Muell. only as far south as Seattle, WA, in 1969 (D. S. Jackson, personal communication).

In 1978, a commercial black raspberry grower east of Portland, OR, noticed some Munger black raspberry plants with a witches' brooming condition that was followed by rapid decline and death of affected plants. Our attention was called to this problem in June 1980 by members of the Oregon State University Agricultural Extension Service, R. L. Smith and R. Garron, Jr. The first Oregon Munger field to show witches' broom symptoms, field A (12 ha), was plowed under in April 1980 because more than 75% of the plants had died, presumably from this disorder. Rooted runner tips from field A had been used to establish field B (12 ha) in 1974. Field A had been planted in 1972, half with certified Munger-70 stock (1) from Washington State and half with Munger-70 plants obtained from a field that had been planted with certified stock the year before. None of the stock had previously shown symptoms of black raspberry witches' broom disease (BRWBD). There were no foreign plant imports on the grower's farm. Munger, which originated in Ohio in 1897 (3), is virtually the only black raspberry cultivar grown on a total of 736 ha in Oregon and Washington.

A survey of eight Munger plantings in the northern Willamette Valley of Oregon in June 1981 failed to identify any other fields infected with BRWBD.

Symptomatology. In infected Munger floricanes, flower buds broke at the normal time and developed into miniature shoots with tiny leaves and compressed internodes (Fig. 1A). Flowers formed on these shoots were small and sterile, often with elongated sepals (Fig. 1B). Infected and seemingly normal floricanes sometimes appeared on the same plant. Most plants with infected floricanes died over the winter.

Two types of symptoms appeared in infected Munger primocanes. After harvest, primocanes that developed symptoms in otherwise healthy-appearing plants lost apical dominance and forced numerous lateral buds, leading to an unusual, somewhat dwarfed, umbrella-shaped growth habit of the primocane with mostly normal-looking, slightly chlorotic leaves (Fig. 1C). Precocious fruiting with virulent, retentive petals sometimes occurred on such canes (Fig. 1D). Examination of 20,000 plants in field B in August 1980 revealed 15 widely scattered plants with umbrella-shaped growth. All of these plants had died by May 1981.

In other instances, perhaps related to the time of infection, basal buds produced numerous, minute vegetative shoots that were extremely shortened, with flattened, stellate whorls of small leaves (Fig. 1E). It seems likely that infections that caused symptoms only on primocanes after harvest became quiescent during late summer, autumn, and winter but became active again in floricanes the following spring, as noted above.

A severe nonmycoplasmalike witches' brooming frequently develops in Rubus spp. that have been sprayed with the commonly used herbicide glyphosate before the plants are killed by the action of this herbicide. Extensive electron microscopy of glyphosate-treated wild R. laciniatus Willd. shoots from field B and nearby fields failed to reveal the presence of mycoplasmalike bodies.

Electron microscopy and serology. Samples of petioles from trees with three witches' broom-diseased Munger primocanes and one floricane were collected in field B in May and August. These samples were fixed, embedded in Epon, sectioned, stained with lead citrate, and examined in a Phillips EM 300 electron microscope. Numerous mycoplasmalike bodies were found in 31% of 140 sieve tubes examined from primocanes (Fig. 2A) and floricanes (Fig. 2B). Such bodies were absent in sieve tubes of petioles from three symptomless, screenhouse-grown, indexed Munger-70 plants that were free from known viruses and virosislike disease agents.

Antiserum against Spiroplasma citri Saglio et al (2), kindly supplied by R. E. Davis, gave negative reactions (Auds = 0.015) in enzyme-linked immunosorbent assay tests (alkaline phosphatase, p-nitrophenyl phosphate system) with sap from witches'-broomed Munger shoots under conditions where strong homologous reactions (Auds = 0.75) were observed with S. citri cultured in vitro and with sap from Catharanthus (Vinca)
roseus (L.) G. Don infected with S. citri, also supplied by Dr. Davis.

Presence of M. fuscula (Zett.). This leafhopper was common on wild, symptomless R. laciniatus growing in ditch banks near field B when collections were made in July 1980. A few M. fuscula also occurred on Munger black raspberry in field B and in an adjoining Willamette red raspberry field, but these were not favored hosts. M. fuscula was also found abundantly on R. laciniatus, R. procerus, and R. parviflorus Nutt. in the Willamette Valley and adjoining areas of the Coast and Cascade mountain ranges, up to 1,200 m. This leafhopper is now well established in several wild Rubus spp. in western Oregon and occurs in low numbers on cultivated black raspberry in western Oregon.

Attempted transmission of witches' broom disease. In September 1980 we attempted to transmit BRWBD from infected to healthy Munger in the greenhouse by leaf graft to six plants and by approach graft to two plants. Graft sources died within a few weeks, and all Munger plants remained symptomless for 12 mo after grafting. The maximum latent period after grafting for Rubus stunt disease (8). M. fuscula collected in August 1980 on R. laciniatus near field B were caged, 10 insects each, on five healthy Munger plants where they died within 3 wk. These Munger plants were then fumigated and held for observation in the greenhouse. They remained symptomless for the following 4 mo. Parallel untreated, healthy Munger plants were also symptomless during the observation periods. Preliminary attempts to prolong the life of BRWBD-infected Munger plants by immersing canes in hot water (8) and in tetracycline solutions (2) were unsuccessful.

Possible relationships of witches' broom disease. Lethal witches' brooming associated with mycoplasmalike bodies in sieve tubes of affected Munger black raspberry is a previously unreported disease in Rubus in North America. In

Fig. 1. Witches' broom disease on Munger black raspberry: (A) Floricanes showing numerous dwarf, compact shoots; May 1981. (B) Floricanes with flowers having elongated sepals; May 1981. (C) Infected primocane (left) showing characteristic umbrella-shaped growth and lack of apical dominance; (right) healthy shoot showing strong apical dominance; September 1980. (D) Precocious fruiting on diseased primocane, with virescent and retentive petals; August 1980. (E) Production of numerous dwarfed shoots (witches' brooms) at base of diseased plant; August 1981.

Fig. 2. Electron micrographs of sieve tubes of Munger black raspberry plants infected with witches' broom disease: (A) Primocane with mycoplasmalike bodies; insert shows enlargement of individual mycoplasmalike body. (B) Floricanes with mycoplasmalike bodies. Bars represent 1,000 nm.
symptomatology, in the presence of mycoplasmalike organisms in diseased plants, and in the common presence of *M. fuscula* on or near diseased plants, BRWBD resembles European Rubus stunt disease (6,8). However, *R. occidentalis* is not commonly grown in Europe and has not been studied there as an experimental host of Rubus stunt disease. In Europe, no red raspberries have been found to be resistant to Rubus stunt disease. In Oregon, no symptoms resembling BRWBD or Rubus stunt disease were found in Willamette red raspberry adjoining a BRWBD-infected Munger field. European Rubus stunt causes phyllody and flower proliferation in infected red raspberry. Such symptoms were absent in Munger infected with BRWBD. Thus, the relationship of BRWBD to European Rubus stunt is uncertain. However, its possible transmission by a vector of European Rubus stunt, *M. fuscula*, deserves further investigation in any case. One possibility is that BRWBD is native to the Willamette Valley of western Oregon, in some plant host where it is normally latent or unrecognized. The recent entry of *M. fuscula* into the area possibly has provided a suitable vector for its spread into *R. occidentalis*.

In view of our failure to demonstrate that BRWBD is infectious, further studies of the nature of the cause and natural spread of this disease are needed. Because the potential for economic damage by BRWBD is so great, *Rubus* crops in the Pacific Northwest, and perhaps elsewhere in North America, should be monitored for the further occurrence of this disease.

ACKNOWLEDGMENT
We thank Chris K. Weiss for electron microscopy.

LITERATURE CITED