Histopathology of Blueberry Stems Naturally Infected with Godronia cassandrae

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ABSTRACT

Living sections of 1- to 3-year-old highbush blueberry stems naturally infected with *Godronia cassandrae* were studied microscopically. Initial necrosis occurred in cortical parenchyma and collenchyma immediately beneath stomata. Most rapid hyphal growth was in longitudinal air channels of the cortex. Chloroplasts of cortical parenchyma turned red 15-20 μm in advance of hyphae. In larger lesions most cortical parenchyma was necrotic, filled with opaque deposits, and invaded by hyphae. Wilting was apparently caused by occlusion of vessels with hyphae, various deposits, and possible tyloses.

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Godronia canker and stem blight of highbush blueberry (*Vaccinium corymbosum* L.), caused by *Godronia cassandrae* Peck, is a severe disease on blueberries in Michigan (1,7). Symptomology of the disease in the field has been recently described (7). This paper reports pathological histology of *G. cassandrae* on naturally infected blueberry stems.

MATERIALS AND METHODS.—One-to 3-year-old diseased stems of Jersey and Bluecrop cultivars were collected in the field during April and May. Tissue was divided into the following infection classes representing development of the disease in the field (7): (i) red lesions $<$ 2.0 mm in diameter; (ii) elliptical lesions 0.5-1.0 cm in length which were devoid of pycnidia; (iii) lesions $>$ 1.0 cm in length with active pycnidia; and (iv) brown-colored xylem from stems in initial stages of wilt. At least five stems with each infection class and five healthy stems were observed on each of the two sampling dates. Stems were immersed in water at 10-15 C for 12 hours to facilitate sectioning (2). Approximately 30, 10- to 15-μm thick sections each were made from the center, basal, and leading edges of each lesion or discolored area of the xylem. Tissues were neither fixed nor stained and were mounted in either distilled water or a 50% (v/v) glycerin solution. Presence or absence of *G. cassandrae* was verified by plating samples of tissue on half strength Difco potato-dextrose agar.

RESULTS.—The epidermis of lesions $<$ 1.0 mm in diameter was unaffected and initial necrosis was always observed in the cortical parenchyma and collenchyma immediately beneath stomata (Fig. 1-A). As lesions increased in size, hyphae were observed intercellularly among cortical parenchyma and closely adpressed to surfaces of living parenchyma which were adjacent to necrotic tissue (Fig. 1-C). Penetration of living cells was not observed. Chloroplasts of cortical parenchyma turned red 15-20 μm in advance of hyphae. Hyphal advance was principally through longitudinal air channels in the cortex (Fig. 1-B).

Dying cells and air channels in the cortex near centers of lesions $>$ 0.5 cm in length were filled with brown-colored opaque deposits and hyphae could not be seen within the cells. Profuse growth of hyphae was observed, however, in air channels and in dead cortical cells after apparent dissolution of cell contents. Pycnidia developed in outer layers of necrotic cortical tissue which was thoroughly ramified with hyphae. Hyphae were observed in necrotic tissue in axillary buds associated with lesions $>$ 0.5 cm in length (Fig. 1-D).

Incipient browning of xylem tissue was often observed by the time lesions developed to 1.0 cm in length. Hyphae and deposits in xylem vessels were usually confined to outermost cells, whereas 30-40% of the vessels in wilting stems were occluded with hyphae, various deposits, and possible tyloses. Hyphae grew longitudinally through vessels (Fig. 1-F) and accumulations of amber-brown-colored deposits were frequently observed between vessel elements (Fig. 1-E).

DISCUSSION.—The location of initial necrosis in incipient lesions suggests that stomata can serve as infection courts on 1- and 2-year-old stems. Other observations made in the field and in inoculation studies support this conclusion; however, petioles and unhealed leaf scars are probably more important as infection courts (6).

Godronia cassandrae is apparently a weak pathogen. In these studies the most rapid hyphal advance was through air channels of the cortex and the fungus failed to penetrate living cells. Our observation that wilting of blueberry plants was more severe following a period of dormancy than in plants growing at low temperatures would support this hypothesis (D. P. Weingartner, *unpublished*). Discoloration of chloroplasts several microns in advance of hyphae, however, suggests that cells are affected metabolically prior to infection by the fungus.

We previously reported that the xylem of all wilted stems was brown-colored at some point below the wilted leaves. These studies suggest that wilting and ultimate stem death is due to occlusion of vessels by hyphae, tyloses and various deposits as has been described for other blueberry diseases (3, 4, 5, 6).

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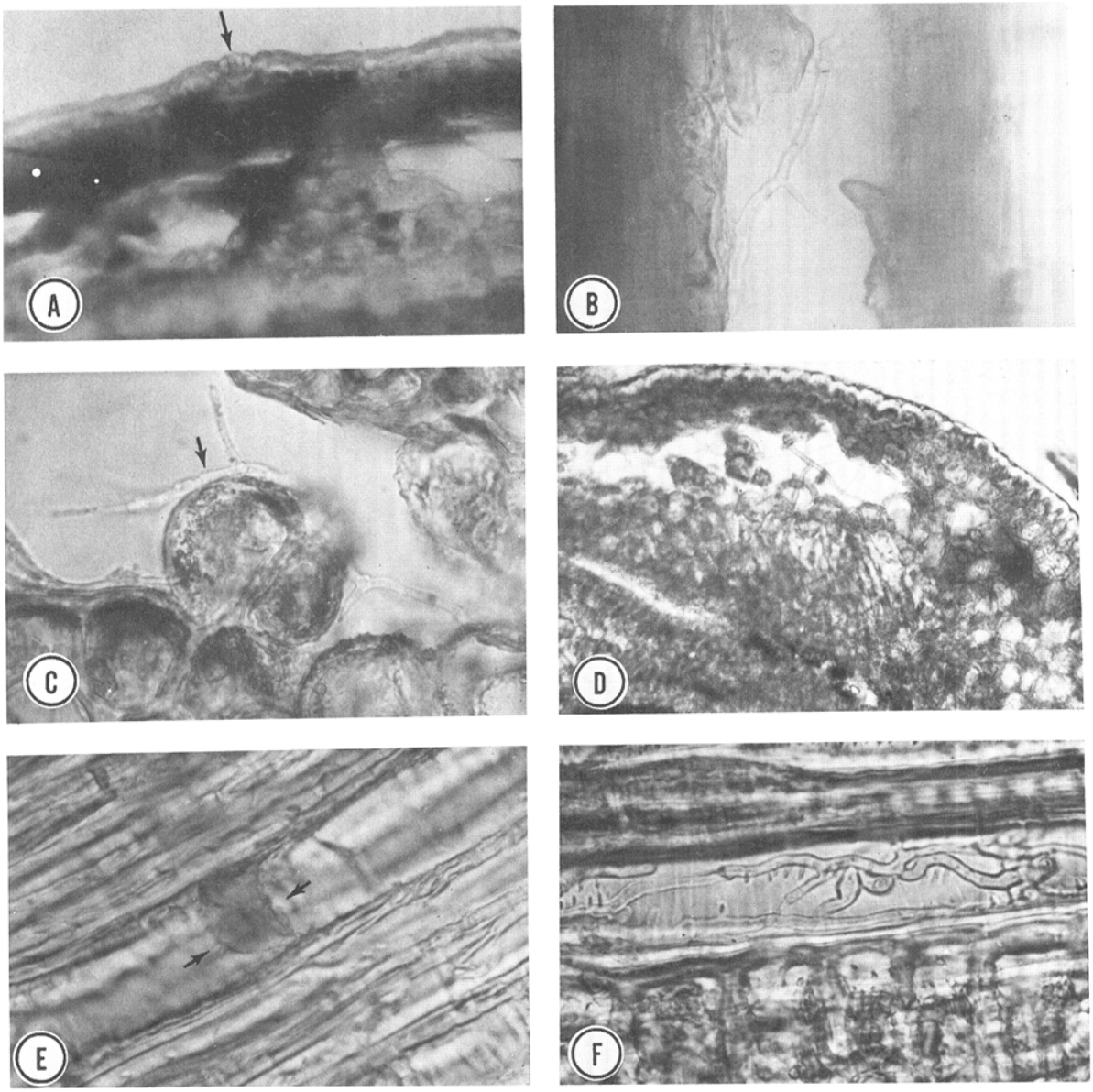

Fig. 1-(A to F). Histopathology of blueberry stems infected with *Godronia cassandrae*. **A)** Transverse section through the center of a small (< 1.0 mm in diameter) internodal lesion showing initial necrosis immediately below a stoma (arrow) (× 204). **B)** Radial section showing hyphae in air channel of cortex (× 816). **C)** Hyphae (arrow) growing along the surface of a living parenchyma cell in the cortex (× 816). **D)** Hyphae and general necrosis in tissue of axillary bud (× 204). **E)** Amber-brown deposit (arrows) between two vessel elements in discolored xylem of a wilted stem (× 816). **F)** Radial section showing hyphae in vessel of discolored xylem of wilted stem (× 816).

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