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Compartmentalization of Discolored and Decayed Wood in Red Maple Branch Stubs

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ABSTRACT. Branch stubs from 12 red maples (*Acer rubrum* L.) were studied to determine differences in tree response to such injuries. Two basic patterns emerged from the 110 stubs dissected. Some stubs (type A) had a clearly visible green-colored boundary, which separated the discolored and decayed wood of the protruding stub from sapwood inside the stem. Other stubs (type B) lacked this boundary, and discolored wood extended into the stem. Fungi and bacteria were more abundant in sapwood and discolored wood of type B stubs than type A stubs. The green-colored boundary was enriched with phenols, and appeared to prevent movement of bacteria and fungi into the stem. This boundary should not be removed when a tree is pruned. FOREST SCI. 27:519–522.

ADDITIONAL KEY WORDS. *Acer rubrum*, pruning, tree wounds.

BRANCH STUBS EXPOSE the wood of living trees to colonization by bacteria and fungi that degrade wood. Branch stubs are one of the major points of initiating decay in living trees (Shigo 1975).

The importance of the size of dead branches to decay has been studied (Toole 1961). However, considerable variation exists in defects associated with branch stubs of similar size (Basham and Anderson 1977). The purpose of this study was to look at differences in the capacity of branch stubs to wall-off, or compartmentalize (Shigo and Marx 1977), discolored and decayed wood initiated with loss of branches in young red maples.

MATERIALS AND METHODS

Twelve red maples (*Acer rubrum* L.) were selected on the Massabesic Experimental Forest in Alfred, Maine. The trees were 15 to 30 years old, 10 to 25 cm diameter at 1.4 m above ground, and 8 to 14 m tall. Trees were growing on moist to very wet sites.

Trees were felled and bolts containing branch stubs were cut 5 cm above and 10 cm below the stub. Forty-five bolts were dissected to isolate microorganisms and collect wood samples for chemical analysis. An additional 65 bolts were split longitudinally through the stub to observe patterns of discoloration and decay. The split sections were sanded to expose the stem and branch pith as well as gross anatomical features of the stub.

Bolts used for isolations and chemical analysis were split through the stub. Wood chips were taken aseptically from one split surface and plated on malt-yeast agar medium containing benlate (g/l-20 agar, 10 malt extract, 2 yeast extract, +5 ppm benlate). Isolations were made from sapwood and discolored wood in type A and B stubs, and from the green-colored boundary tissues of type A stubs (Fig. 1). Isolations were also made from sapwood of five living branches as controls. The wood chips were incubated in the medium for 3 weeks at 25°C in darkness. Fungi which grew from them were identified to genus where possible.

On the opposite split face, a small wood gouge was used to collect samples of sapwood, discolored wood, and the green-boundary tissue. Chips of the same tissue type were air-dried, combined, and ground in a Wiley mill to pass a 425 μm sieve. Duplicate half-gram samples were extracted in 50 ml of distilled water in a boiling water bath under reflux with constant stirring for 1 hr. The extract was filtered through tared, fritted-glass crucible, porosity C, and used for phenol (Folin-Ciocolteau method, Horowitz 1960) and carbohy-

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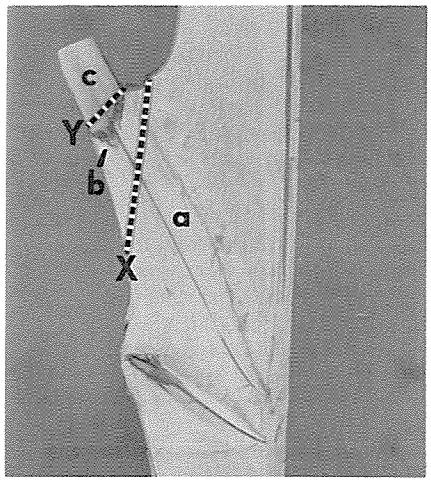

FIGURE 1. Type A stub, well compartmentalized; sampling sites a = sapwood, b = green-colored boundary, and c = discolored wood: and location of flush cut (X) versus pruning at callus collar (Y).

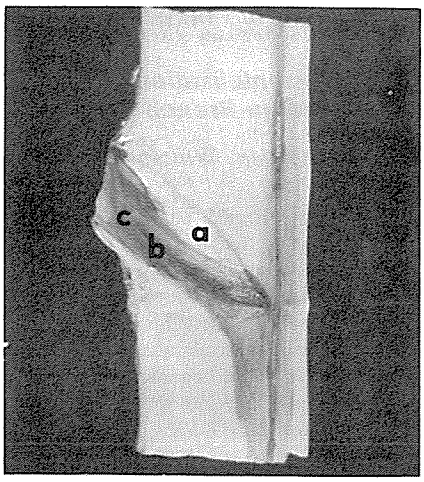

FIGURE 2. Type B stub, poorly compartmentalized; sampling sites a = sapwood, b = inner discolored wood, and c = outer discolored wood.

drate (Anthrone method, Loewus 1952) determinations. The extracted wood was washed with boiling distilled water and dried at 104°C to constant weight. Soluble dry matter was determined by subtracting the final oven-dry weight from the initial oven-dry weight calculated by using a moisture correction factor determined from separate half-gram samples.

Forty ml of the aqueous extract used for phenol and carbohydrate determinations were extracted 3 times with 20 ml chloroform : ethyl acetate (1:1) and the organic layer dried by filtration using PS 1 phase separators. The organic layer was evaporated to dryness and redissolved in 1 ml 95 percent ethanol. Thin-layer chromatograms were made on precoated silica gel 60 plates using 10 λ spots of extract and either chloroform : methanol (9:1) or toluene : ethyl formate : formic acid (5:4:1) as developing solvents. Spots were located by UV fluorescence and by spraying with 1:1 mixture of 0.6 percent aqueous solutions of ferric chloride and potassium ferricyanide (FCF reagent). Unsprayed spots were eluted with ethanol (95 percent with 1 percent HCl) and UV spectra determined with and without ionization using ethanolic NaOH.

RESULTS

Two major patterns of dead branch stubs were observed. Type A (Fig. 1) had a clearly defined green-colored boundary, which separated the discolored and decayed wood of the stub from sapwood inside the stem. This boundary began at the callus ridge around the stub and extended most deeply into the stem along the branch pith. Type B (Fig. 2) stubs lacked this boundary.

No bacteria or fungi were isolated from the sapwood of the five control branches. Bacteria and fungi were rarely isolated from the sapwood of type A stubs, but were commonly found in the sapwood of type B stubs (Fig. 3). Fungi occurring 4 or more times in sapwood of type B stubs were *Cytospora* sp. and *Penicillium* sp.

Fungi were abundant in the discolored wood of both type A and B stubs, but the discolored wood of B yielded twice as many isolates as A (Fig. 3). Fungi (genera) occurring 4 or more times in type B stubs included, in decreasing order of occurrence: *Cephalosporium, Gliocladium, Nodulisporium, Phialophora, Penicillium,* and *Trichocladium. Penicillium* was the only fungus that occurred 4 times in discolored wood of type A stubs. Only a few nonidentified decay-causing hymenomycetes were isolated, and they were in discolored wood of type B stubs.

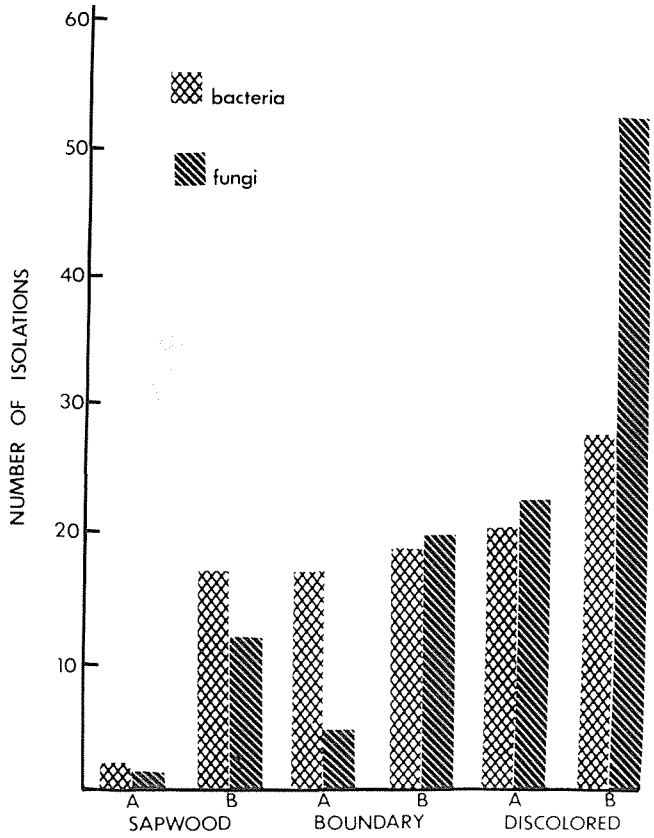

FIGURE 3. Number of isolations of bacteria and fungi from the sapwood, the green-colored boundary tissue (type A) or inner discolored wood (type B) and discolored wood of 13 paired stubs.

Bacteria were more common than fungi in the green-colored boundary of type A stubs (Fig. 3). Fungi in this tissue were less abundant than in discolored wood.

The green-colored boundary tissue of type A stubs had more phenol than either sapwood or discolored wood of type A or B stubs when expressed per unit weight of moisture-free wood, soluble dry matter, or total soluble carbohydrate (Table 1).

Sapwood and discolored wood of type A and B stubs are similar in amounts of dry matter, phenols, and carbohydrates.

Chromatograms of extracts from green-colored boundary tissue had more spots detected with UV light, FCF reagents, and aerial oxidation than those of sapwood or discolored wood. Substances eluted from major spots of the green-colored tissue had absorbances of >300 nm and bathochromic shifts of >20 nm. Those from sapwood and discolored wood were <300 and <20 nm.

DISCUSSION

These results indicate that the green-colored boundary tissues of type A stubs are similar in composition and function to those observed around discolored wood resulting from drill wounds in red maple (Shortle 1979). The discolored and decayed wood initiated by loss of branches appeared to be walled-off or compartmentalized by the phenol-enriched tissue in type A stubs. Similar protective boundary tissues have been described in branches of both broadleaved and coniferous trees (Aufsess 1975). Failure of the boundary tissues to form, or destruction of the tissues by microbial activity, may facilitate spread of micro-organisms, discoloration, and decay into the stems.

TABLE 1. Analysis of water soluble dry matter of sapwood, green-colored boundary tissue, and discolored wood of type A stubs and of sapwood and discolored wood of type B stubs.

Soluble substance	Sapwood		Boundary	Discolored	
	A	B	A	A	B
Phenol (mg/g wood)[a]	15	15	45	10	13
Phenol (mg/g dry matter)	225	210	310	105	185
Phenol (mg/g carbohydrate)	300	300	1,500	500	520
Dry matter (mg/g wood)	75	70	145	70	70
Carbohydrate (mg/g wood)	50	50	30	20	25

[a] Mean of duplicate observations.

Factors determining whether a stub will be type A or B are not known at present. Genes of the tree may be important (Shigo and others 1977), and also genes of the fungi (Shortle and Cowling 1978).

One major consequence of understanding how branch stubs compartmentalize discolored and decayed wood relates to pruning. Flush cutting branch stubs of type A removes the boundary tissue which appears to prevent internal spread of discoloration and decay (Fig. 1). Cutting such stubs at the callus collar would maintain the defensive boundary tissues and reduce inoculum in the protruding dead stubs. Consequences of removing the callus collar, and probably the internal defensive boundary tissues, have been observed in black walnut (Shigo and others 1978, 1979).

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