Effects of Cronartium ribicola on Soluble Sugars in Pinus monticola Bark

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ABSTRACT

Fructose, glucose, sucrose, and raffinose were the soluble sugars identified from *Pinus monticola* bark, and bark infected by *Cronartium ribicola*. There were no qualitative differences between the two types of tissue, but glucose, sucrose, raffinose, and total sugar content were significantly reduced in invaded bark. In contrast, fructose showed a nonsignificant reduction. Trace amounts of two polyols were detected within infected bark. The physiological implications of these findings are discussed.

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Much information has accumulated concerning the quantitative effects of obligate and faculative parasites on fructose, glucose, and sucrose concentrations in diseased tissues of annual hosts (2, 3, 6, 13). However, very little information exists on the qualitative and quantitative effects of obligate parasites on woody perennial hosts (5). The objective of this study was to determine the qualitative and quantitative effects of *Cronartium ribicola* on ethanol-soluble sugars in *Pinus monticola*. Because of the heterotrophic nature of *C. ribicola,* we expected a reduction of the ethanol-soluble sugars in the infected *P. monticola* bark.

MATERIALS AND METHODS.—Forty 10- to 15-year-old western white pines, each supporting a single bole canker (free of secondary organisms) were selected in October 1971 from a natural stand of mixed conifers on the East Fork of Potlatch Creek, Bovill, Idaho. In June, 1972, 11 of these trees with cankers were harvested. The bole cankers and noninfected bark samples taken about 30.5 cm above the canker (11 infected and 11 noninfected samples) were quick-frozen on the sampling site and transferred to laboratory freezers (−5 C).

Infected bark located between the pycnia scars and 1 cm beyond the discolored canker margin was used (11). Bark samples were prepared and extracted in a Soxhlet apparatus with petroleum ether and 95% ethanol as previously described (11). Clearing of the ethanol extracts of substances that interfere with chromatography and the chromatographic and densitometric procedures for measuring quantities of individual free sugars have been described (10). For polyol determination, we used the chromatographic techniques described by Macek (7), and the spray reagent (0.1% periodate-benzidine) developed by Cifonelli and Smith (1).

Because we expected concentrations of individual ethanol-soluble sugars to be lower in rust-infected bark than in noninfected bark, one-tailed "t" tests were used to determine differences between the two bark classes. The "t" values were calculated by using the method of comparing sample means with paired observations outlined by Steel and Torrie (9).

RESULTS.—The free sugars identified by thin-layer and paper chromatographic analyses were fructose, glucose, sucrose, and raffinose. There were no qualitative differences between noninfected and infected bark, except that raffinose (which was detected in trace amounts in all extracts of noninfected bark) was found in only six extracts of infected bark. Trace quantities of two polyols were detected in all extracts of infected bark, but not in extracts of noninfected bark. The two polyols of infected bark had chromatographic (paper) migration characteristics of ribitol and sorbitol. However, chromatographic analysis was not extensive enough to merit positive identification.

The amount of fructose was not significantly lower in infected bark, but glucose, sucrose, and total free sugars were significantly reduced in infected bark (Table 1). Because raffinose was not detected in 5 of 11 extracts of infected bark, we believe this sugar was present in smaller quantities in infected than in noninfected bark.

DISCUSSION.—There appears to be an increased demand for glucose, sucrose, and probably raffinose in infected bark. That demand appears strong enough to lower sugar concentrations below the levels found in noninfected bark. This hypothesis assumes (i) that *C. ribicola* stem infections do not decrease the transport of these sugars from their points of synthesis in needles to the infection sites and (ii) that the rate of starch breakdown to glucose within diseased tissues is not reduced. We accept the first part of this hypothesis. Our opinion is based on other studies that have demonstrated that ^{14}C-labeled materials accumulate at infection sites of both obligate and nonobligate pathogens (4, 8, 13). As to the second part of the hypothesis, we cannot envision an obligatory host-pathogen relationship that would allow

TABLE 1. Concentrations of 95% ethanol-soluble sugars within *Pinus monticola* bark both noninfected and infected by *Cronartium ribicola*

Sugar	Ethanol-soluble sugars (mg/g) Noninfected bark[a]	Infected bark[a]	$\bar{d}$[b]	t
Fructose	13.2	12.2	1.07	1.45
Glucose	12.2	10.9	1.25	1.96*[c]
Sucrose	18.2	15.8	2.24	3.80*
Total	43.6	38.9	4.67	2.64*

[a]A mean of 11 samples expressed as milligrams of ethanol-soluble sugars per gram of freeze-dried tissue.
[b]$\bar{d}$ = the mean difference.
[c]Asterisk denotes a significant difference, $P = 0.05$.

depletion of a metabolically important sugar and, at the same time, reduce the breakdown of storage carbohydrates that would replenish the supply of this sugar. Therefore, it seems that the decrease of glucose, sucrose, and possibly raffinose in infected bark is due to increased utilization or assimilation of these molecules, and not to reduced rates of transport to infection sites or of starch breakdown.

Decrease of glucose and sucrose in infected bark tissue can be explained by considering metabolic demands for these sugars. These sugars may furnish carbon skeletons needed for building the materials soluble in 95% ethanol that were found in quantities significantly higher in infected bark tissues than in noninfected bark (11). This hypothesis assumes that ethanol-soluble substances are synthesized in infected bark and do not result from accumulation of substances transported from other parts of the tree (11, 12). Inasmuch as polyols are detectable in trace amounts in infected bark only, we further believe that these molecules are of fungal origin.

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