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May 1998

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Kimball, Bruce A.; Turnblom, Eric C.; Nolte, Dale L.; Griffin, Doreen L.; and Engeman, Richard M., "Effects of Thinning and Nitrogen Fertilization on Sugars and Terpenes in Douglas-Fir Vascular Tissues: Implications for Black Bear Foraging" (1998). *USDA National Wildlife Research Center - Staff Publications*. 178.

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Effects of Thinning and Nitrogen Fertilization on Sugars and Terpenes in Douglas-Fir Vascular Tissues: Implications for Black Bear Foraging

Bruce A. Kimball, Eric C. Turnblom, Dale L. Nolte, Doreen L. Griffin, and Richard M. Engeman

ABSTRACT: The chemical constituents of coniferous vascular tissues play a role in bear forage selection. In particular, bear foraging preferences are related to the concentrations of simple sugars (nonstructural carbohydrates) and terpenes in the forage. Analyses of vascular tissue samples from trees collected in test plots indicated that both thinning and fertilization caused the sugar concentration of vascular tissues in the lower bole to increase. However, these treatments had no effect on the concentrations of hydrocarbon monoterpenes, oxygenated monoterpenes, or sesquiterpenes. These results may explain the observations that black bears prefer to forage in thinned and fertilized stands. *For. Sci.* 44(4):599–602.

Additional Key Words: *Pseudotsuga menziesii*, *Ursus americanus*.

CONIFER VASCULAR TISSUES ARE staples in the spring diet of some bears (Noble 1993). Black bears (*Ursus americanus*) feed on cambial zone vascular tissues of conifers by removing the bark with their claws and scraping the vascular tissue with their incisors. This type of foraging is presumed to be limited to the spring until other forage items become available (Radwan 1969). An individual bear can peel the trunks of 50 to 70 trees per day (Schmidt and Gourley 1992).

In general, bears target trees ranging in age from 15 to 25 yr (Schmidt and Gourley 1992). Bears have been known to prefer the vascular tissues of Douglas-fir (*Pseudotsuga menziesii*) to western hemlock (*Tsuga heterophylla*), western red cedar (*Thuja plicata*), and red alder (*Alnus rubra*; Radwan 1969). Preference for lodgepole pine (*Pinus contorta*) over ponderosa pine (*Pinus ponderosa*) has also been reported (Barnes and Engeman 1995). We have observed that prefer-

ence may be related to tree ontogeny. For example, preference of western hemlock over Douglas-fir has been observed when western hemlock flushed before Douglas-fir.

The area of removed bark from a single tree may range from a few square centimeters to complete girdling of the lower portion of the main stem. Girdled trees are often found adjacent to trees that were ignored or minimally peeled. Tree selection by bears has also been observed at larger scales. Black bears prefer to forage vascular tissue in thinned stands versus higher density stands (Schmidt and Gourley 1992, Kanaskie et al. 1990, Mason and Adams 1989). Furthermore, preference for urea fertilized trees has been reported (Nelson 1989). Tree vigor has been implicated as a driving force for these observations.

Black bear foraging preference was recently shown to be related to the concentrations of the simple sugars (fructose, glucose, and sucrose), hydrocarbon monoterpenes, oxygen-

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Acknowledgments: The authors wish to thank the Stand Management Cooperative Policy Committee for their cooperation with this study. A special thanks to all land owners and their staff who provided access to the properties. Thanks also to John Beck, Jerry Guthrie, and Ed Yoshihara for their field and laboratory assistance. This work was partially funded through the Collaborative Research Team. The input and guidance of Dr. Frank R. Stermitz has been particularly helpful. The review comments of Connie Harrington and John Johnston were also gratefully appreciated. Mention of specific products does not constitute endorsement by either the United States Department of Agriculture or the University of Washington.

Manuscript received September 29, 1997. Accepted May 7, 1998.

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ated monoterpenes, and sesquiterpenes in the vascular tissue (Kimball et al. 1998). Douglas-fir trees that were foraged by bears were subjected to chemical analyses and preferences of free-ranging bears were tested with bioassays. In that study, bears were found to maximize sugar intake and minimize the intake of terpenes while foraging. Thus, for this study we hypothesized that bear preference for trees in thinned or fertilized stands was mediated by a higher concentration of vascular tissue sugars and/or lower terpene concentration versus trees in higher density or unfertilized stands. The study described here was designed to evaluate the impact of thinning and fertilization on the presence of sugars and terpenes in Douglas-fir vascular tissue.

Materials and Methods

Study Sites

This study was conducted on Stand Management Cooperative (SMC) installations located in western Washington and northwestern Oregon (Table 1). These sites were situated in well-established juvenile stands of Douglas-fir planted between 1974 and 1984. Within a site, 0.47 ha square plots were prepared. Each plot consisted of an interior 0.2 ha square and a surrounding buffer area.

Silvicultural Treatments

At each site, plots (interior and buffer areas) were randomly subjected to the appropriate treatment. Treatments were initiated between 1987 and 1992 to investigate the impacts of current silvicultural management practices on growth and wood production (Maguire et al. 1991, Stand Management Cooperative 1993).

Three fertilized and three unfertilized plots were examined at each installation. Urea (46-0-0) was hand delivered to each treated plot at a rate of 224 Kg/ha. Fertilization treatments were applied either 1, 2, 3, or 4 yr prior to sampling (Table 1).

Table 1. Location of test sites and time elapsed between fertilization treatment and sample collection.

Installation	Location	Years since fertilization
704	Cowlitz County, WA (46-10'N, 122-55'W)	1
705	King County, WA (47-10'N, 121-45'W)	1
708	Lewis County, WA (46-30'N, 122-5'W)	4
713	Skagit County, WA (48-30'N, 121-40'W)	4
718	Linn County, OR (44-40'N, 122-40'W)	3
722	Marion County, OR (44-50'N, 122-30'W)	3
725	Jefferson County, WA (47-50'N, 122-50'W)	2
726	Lincoln County, OR (44-45'N, 123-50'W)	2
736	King County, WA (47-35'N, 121-45'W)	4

Precommercial thinning was systematically employed to yield three density treatment levels. Two plots (one fertilized and one unfertilized) were not thinned and maintained at high density (850 to 1400 stems/ha—sph), two plots were precommercially thinned to a mid-density level (400 to 700 sph) and two plots were thinned to a low density level (250 to 325 sph). Not all density levels were represented at three of the sites. Pretreatment tree diameter data was evaluated to determine if mean plot diameter varied by density level prior to thinning.

Sample Collection

Each plot was sampled once. Samples were collected over the period of May 17 to July 1, 1996. Eight trees were sampled in the buffer area of each plot. Cambial zone vascular tissue (phloem and oleoresin located immediately below the cork cambium) was collected by removing an 80 cm (high) × 10 cm (wide) patch of bark on the east side of each tree with steel cutting tools resembling meat cleavers. The top of the sample area was 1.0 m off the ground. The vascular tissue was scraped from the tree into a tared plastic freezer bag with a laboratory spatula. The diameter at breast height (dbh) of each tree sampled was measured.

The freezer bag and contents were immediately placed in liquid nitrogen for 2 to 5 min. After complete freezing, the samples were kept on dry ice until placed in a laboratory freezer at -24°C. Vascular tissue mass was determined as the mass of sample obtained from the 800 cm² sample area. Frozen samples were homogenized by pounding with a mallet and divided into two portions for chemical analyses. One portion remained frozen for terpene analyses while the other portion was lyophilized prior to sugar analyses.

Chemical Analyses

Terpene analyses were performed in duplicate according to the method of Kimball et al. (1995) except that only a single mixed standard was prepared. The concentrations of 24 individual terpenes were determined in each sample. Duplicate sugar analyses of the lyophilized vascular tissue samples were performed according to the methods described in Kimball et al. (1998). Fructose, glucose, and sucrose were quantified in each sample.

Chemical Variables

The mean values of the eight samples taken from within the experimental unit (plot) were the variables subjected to statistical analyses. As in a previous study, three terpene variables were calculated as the sum of terpenes in each unique structural group: hydrocarbon monoterpenes (HC), oxygenated monoterpenes (OXY), or sesquiterpenes (SES, Kimball et al. 1998). The total sugar concentration was defined as the sum of glucose, fructose, and sucrose.

Growth Variables

The two measures were diameter growth (dbh) and vascular tissue mass (mass of cambial zone vascular tissue per 800 cm² sample area). Diameter and mass values used in statistical analyses were means based on the eight trees sampled in each plot.

Statistical Design

The data were analyzed as a four-factor mixed-model design. The design was unbalanced because the density of 5 of the 54 plots did not fall in the specified density levels. Tests for fixed effects: time since fertilization (1, 2, 3, or 4 yr), fertilization (yes or no), stand density (high, mid, or low); as well as all factor level interactions were conducted using the mixed procedure in SAS (Littell et al. 1996). The random effects were site (nested in time since fertilization) and the interactions of both fertilization and density with site (time). Multiple comparisons of least square means were made using the pdiff option in SAS (SAS 1990).

Univariate analyses were performed for each of the following responses: total sugars, HC, OXY, SES, mass of cambial zone vascular tissue, and tree diameter. The relationships between the responses were investigated using correlational analysis (proc corr, SAS 1990). Pretreatment diameter data were evaluated by generating 95% confidence intervals for each density level from the mean tree diameter of each plot.

Results

Growth Responses

Tree diameter, a measure of cumulative growth, was greater at low stand densities as compared to the mid and high densities ($P = 0.007$; Table 2). Fertilization also had a positive effect on tree diameter ($P = 0.034$). Fertilized trees had a mean dbh of 19.7 cm as compared to 18.6 cm for unfertilized trees.

Pretreatment diameter data indicated that dbh did not vary by density level prior to thinning. Confidence intervals for the low, mid, and high density plots were 7.58–8.70 cm, 7.68–8.80 cm, and 7.43–8.55 cm, respectively.

Vascular tissue mass, a measure of current year's growth, was significantly impacted by the interaction of fertilization and density ($P = 0.01$). Thus, fertilization had a positive effect on vascular tissue mass in high density stands, while having no effect at mid or low density levels (Figure 1).

Chemical Variables

Total sugar concentration was greater in low density stands than in mid or high density stands ($P = 0.02$; Table 2). Total sugars were also subject to the interaction of fertilization and the time elapsed since the fertilization treatment ($P = 0.05$; Figure 2). Fertilization had a positive effect on total sugars in plots that were fertilized 1 yr prior to measurement, but had no effect on plots fertilized 2, 3, or 4 yr earlier. No terpene variable was subject to any fixed effect.

Table 2. Differences in tree diameter ($P = 0.007$) and vascular tissue sugar concentration ($P = 0.02$) by tree density level (sph = stems per hectare; values with different letters within a column are significantly different).

Density level	Tree diameter (cm)	Total sugars (%)
Low (250–325 sph)	20.4a	3.19a
Mid (400–700 sph)	18.9b	3.03b
High (850–1,400 sph)	18.1b	2.98b

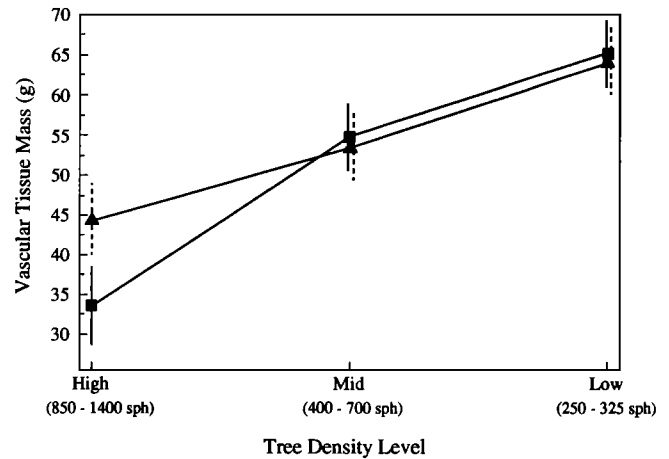


Figure 1. Effects of fertilization and tree density on cambial zone vascular tissue mass ($P = 0.01$; sph = stems per hectare). ■ Not fertilized, ▲ Fertilized.

Relationships Among Variables

Total sugar concentration was positively correlated with mass of vascular tissue ($r = 0.400$; $P = 0.004$) and tree diameter ($r = 0.605$; $P < 0.0001$). Vascular tissue mass and tree diameter were also positively correlated ($r = 0.315$; $P = 0.03$).

Discussion

We observed the impacts that thinning and fertilization were designed to produce on tree growth. Thinning had a significant impact on tree diameter as did fertilization. In addition to increasing diameter growth, thinning also impacted the vascular tissue sugar concentration. The total sugar concentration was significantly higher in the trees growing in low density stands. Correlational analysis also identified the link between growth and vascular tissue sugars. Total sugars were positively correlated with both diameter growth and vascular tissue mass. Sugar levels in the cambial zone are generally tied to growth (Kramer and Kozlowski 1979).

Total sugars were only increased by fertilization 1 yr following treatment (Figure 2). While no fertilized plots were sampled in the year of treatment, it is likely that fertilization

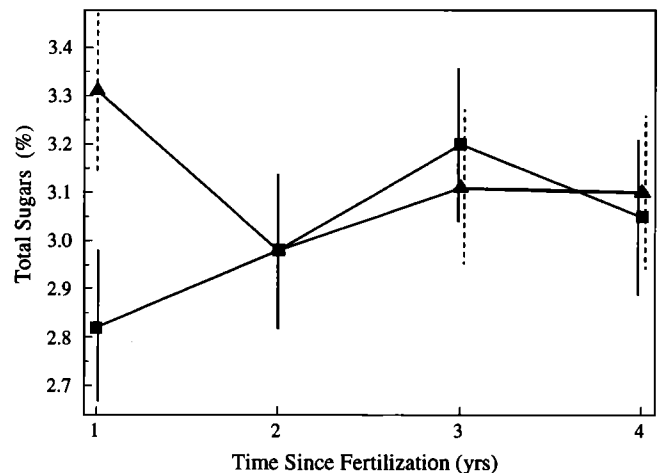


Figure 2. Effect of fertilization and time elapsed since fertilization on total sugar concentration ($P = 0.05$). ■ Not fertilized, ▲ Fertilized.

effects could have been apparent in the same year of treatment. Carlyle (1995) found that 51% of nitrogen uptake by Monterey pine (*Pinus radiata*) occurred in the first 20 wk following fertilization.

Thinning had no impact on vascular tissue terpenes (HC, OXY, or SES). This is consistent with previous work which demonstrated that loblolly pine (*Pinus taeda*) phloem levels of α -pinene, β -pinene, myrcene, and limonene were not affected by thinning (Matson et al. 1987). Fertilization also had no impact on any of the vascular tissue terpene variables. Previous studies have similarly demonstrated that nitrogen fertilization at a rate of 224 kg/ha had no impact on foliar terpenes in grand fir (*Abies grandis*) while fertilization at 448 kg/ha reduced the foliar concentration of only a few individual monoterpenes (Muzika et al. 1989).

A previous study demonstrated that free ranging black bears respond to changes in terpene and sugar concentrations when foraging (Kimball et al. 1998). When given a choice, bears preferred higher sugar-containing diets and were deterred when terpenes increased. The current study demonstrated that thinning and fertilization significantly impacted the concentrations of some of these same vascular tissue chemicals. The observation that bears prefer to forage in thinned stands may be partially attributed to the higher vascular tissue sugar concentrations of trees in thinned stands. Furthermore, exposure to terpenes would not be significantly impacted by foraging in thinned stands. Bears may additionally benefit by foraging in thinned stands because the available mass (per 800 cm²) of cambial forage was greatest in trees from low density stands.

Low density stands with open canopies are visually different from high density stands with closed canopies. In particular, the structure of the understory vegetation is significantly affected by the amount of light transmitted through the canopy. The flavor and feedbacks of foraging in low density stands may be associated with the visual properties of the stand including the amount of light on the forest floor and the understory vegetation. Cognitive processes allow foraging animals to recognize preferred diets without having to taste them (Provenza et al. 1992). Therefore, black bear preference of vascular tissue in low density stands may be mediated by the chemical constituents of the forage and discerned by visual association.

The observation that bears prefer to forage in fertilized stands may also be explained in part by the chemical constituents of the vascular tissues. Although plots were not inspected yearly, the work of Nelson (1989) indicated that foraging activity in fertilized stands of Douglas-fir was greatest in years immediately following fertilization. Our results indicate that preference for urea fertilized trees may be limited to the first year following fertilization. Vascular

tissue sugar concentrations were found to be higher in fertilized versus unfertilized trees one year following treatment, but not in subsequent years (Figure 2). In addition, vascular tissue terpenes were not impacted by fertilization.

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