WESTERN SPRUCE BUDWORM CONSUMPTION - EFFECTS OF HOST SPECIES AND FOLIAGE CHEMISTRY

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Feeding efficiencies and growth rates of western spruce budworm larvae varied among hosts tested. Pupae attained normal size regardless of host species. Candidate defensive compounds (tannins and phenols) varied only slightly with the vigor of the host. The relationship between these defensive compounds and measures of larvae growth were not entirely consistent with current theories on the role of secondary plant compounds.

Introduction

Within a population of host plants, the intensity of insect attack and subsequent damage may vary considerably. It is not uncommon for two Douglas-fir, Pseudotsuga menziesii var. glauca (Beissn.) Franco, trees near each other to have quite different populations of western spruce budworm, Choristoneura occidentalis Freeman (McDonald 1981). Western spruce budworm (WSBW) frequently exhibits this preferential habit within and among its hosts in the Southwest.

Numerous explanations for preferential attack by insects have been suggested including differences in nutritional values, phenology, foliage toughness, host vigor, and plant defensive compounds. Foliage chemicals have been implicated as specific factors in the resistance-susceptibility characteristics of Douglas-fir (McMurray 1980). Literature on the important plant defensive compounds has been adequately reviewed by the following authors: tannins (Swain 1979), phenols (Levin 1971, Swain 1977), and nitrogen (Mattson 1980).

In this paper we present some preliminary results on differences in feeding by WSBW among the important hosts in the Southwest. We also report on correlations between WSBW feeding and concentrations of phenols, tannins, and proteins in white fir, Abies concolor (Gord. and Glend.) Lindl. ex Hildebr., and Douglas-fir foliage.

Methods

Tree Selection

Four hosts of WSBW occurring in northern Arizona were selected for the feeding experiments: Douglas-fir, white fir, corkbark fir, Abies lasiocarpa var. arizonica (Merriam) Lemm.; and Engelmann spruce, Picea engelmannii Parry ex Engelm. Trees were selected from mixed conifer stands approximately 16 km north of Flagstaff, Arizona, stand elevation approximately 2,440 meters. These stands had no recent history of WSBW outbreaks.

Trees suitable for site index determination were rated for vigor according to Waring et al. (1980). This technique uses the ratios of the basal area growth for one-year and five-year increments to total sapwood area. To obtain average measurements, four increment cores were taken at 90° angles from each of 25 trees of each host species. The increment cores were then stained with potassium iodide-iodine to aid in distinguishing the sapwood from the heartwood (Kutscha and Sachs 1962). Total radius, sapwood radius, and one- and five-year increment radii were measured. The ratios were calculated using this formula:

$$\frac{BA_x}{SA} = \frac{R^2 \pi - (R - r_x)^2 \pi}{R^2 \pi - (R - r_s)^2 \pi}$$

BA_x: basal area of growth increment for period x

SA : sapwood area

R : total radius

r_x : radius of increment growth

r_s : radius of sapwood

Fifteen trees were selected from the 25 which were cored for each species (five trees in each of three vigor categories). The five trees with the highest ratios were classified as high vigor trees, the five trees with the lowest ratios were classified as low vigor trees, and the five trees closest to the mean were classified as medium vigor trees. The theoretical range of BA/SA ratio is 1.00 to 0.00. The actual range of BA_5/SA observed for the four species was: Douglas-fir 0.49 to 0.14, white fir 0.59 to 0.20, corkbark fir 0.63 to 0.24, and Engelmann spruce 0.60 to 0.13.

Selection of Larvae

Second and third instar larvae were collected in early May from an ongoing WSBW outbreak within the Kaibab Naitonal Forest, Arizona. Budworm infested branch tips were collected from the four host species used in this study. Larvae were carefully segregated according to host species; this allowed the larvae to be fed foliage from the same host

species from which they were collected. The branch tips were laced in Aquapics® to maintain freshness and transported to the laboratory.

The field collected larvae were stored at 2-4°C to slow development until five to seven days before the experiments were to begin. The larvae were then placed at room temperature until they advanced to the fourth instar. Larvae were determined to be in the fourth stadium by head capsule measurements. Bean and Blatzer (1957) and Wagg (1958) were used as guidelines.

Foliage Collection

To standardize the host phenological stage, feeding experiments were begun when the host foliage was determined to be at the "brush" stage (bud cap gone, needles even but no shoot growth so needles appear to arise from one spot) (Shepard unpublished).

We collected paired foliage samples from the midcrown of the host trees at the four cardinal directions. Paired twigs were selected for uniformity in size and phenology. One twig was fed to a budworm, the other twig was used to estimate the average dry weight per needle of the feeding twig. This average dry weight was later used to estimate the dry weight of the foliage consumed by the budworm larva.

Feeding Procedure

The paired twigs were saturated in a 100% relative humidity environment overnight. The following morning the mature foliage was trimmed from the feeding twig. While being held under water, the twig was excised and placed in a water-filled Aquapic®. The prepared foliage was then placed in a paper-lined 150 x 25 mm petri dish.

Fourth instar larvae were weighed fresh and placed on the prepared foliage to feed. Initial dry weight of each larva was estimated using an average percent dry weight of fourth instar larvae. The petri dishes were placed in controlled environmental chambers for the duration of the experiments. Temperature in the chambers ranged from 24 to 26°C; the photoperiod was 16 hours of light and 8 hours of darkness, as recommended by Robertson (1979).

Following each 72 hour feeding period the feeding twigs were removed and replaced with fresh foliage. The number of damaged needles was recorded and the damaged needles still attached to the twig and wasted needles were collected and oven dried at 60°C until no further weight loss occurred. Both wasted and remaining foliage were then weighed on a balance accurate to 0.1 mg. The amount of foliage ingested by each larva was estimated using the following formula:

$$I = (DW \times N) - (W + R)$$

I : foliage ingested

DW: mean dry weight per needle

N : number of needles damaged

W : foliage wasted (clipped but not consumed)

R : foliage remains (damaged but not clipped from the twig)

Each replicate was terminated within 24 hours of pupation. Total foliage ingested was calculated; pupae were sexed and weighed fresh. Feces and pupae were then oven dried at 60°C and weighed.

Nutritional indices were calculated on a dry weight basis according to Waldbauer (1968). The following nutritional indices are used in this study: foliage ingested (I), relative consumption rate (RCR), relative growth rate (RGR), approximate digestibility (AD), and efficiency of conversion of ingested food to body weight (ECI).

Chemical Analysis

Foliage used in the chemical analyses was clipped from the host trees selected for the feeding experiment and immediately frozen for 18 hours at -20°C. Samples were then freeze-dried at -50°C and 50 millitorr in a freeze dryer. Samples were stored at -20°C under dessication until analyzed.

Proteins were analyzed using the Coomassie Brilliant Blue Dye technique (Bradford 1976). Total phenols were determined using the Folin-Denis technique according to Swain and Hillis (1959) and Ribereau-Gayon (1972). Tannins were analyzed using the vanillin reaction assay (Price et al. 1978) as modified by Zucker (unpublished).

Results and Discussion

Comparison of Rearing Methods

The effect of three commonly used rearing procedures on fresh and dry pupal weights was determined for WSBW feeding on Douglas-fir and corkbark fir. Fresh pupal weights were significantly different among rearing methods for corkbark fir but not for Douglas-fir (Table 1). For corkbark fir, fresh pupal weights were significantly higher for insects bagged on foliage in the field than for insects fed excised foliage in the lab. Excised foliage and bagged foliage tests were conducted on the same trees and crown positions. Fresh pupal weights were not significantly different between insects reared on excised foliage and insects that developed under normal field conditions for either host species. Dry weights of pupae were not significantly different between rearing methods (Table 2).

Table 1. Fresh pupal weights of female western spruce budworm by rearing method and host species. a/

REARING METHOD	DOUGLAS-FIR	CORKBARK FIR
EXCISED FOLIAGE	70.6 (5) b/	69.0 (5) A c/
FIELD BAGGED	91.6 (4)	83.6 (6) B
FIELD COLLECTED	70.5 (77)	59.8 (51) A
F-PROB.	0.1390	0.0014

a/ weights in milligrams.
b/ numbers in parentheses are number of cases.
c/ oneway ANOVA, LSD multiple range test, $\alpha = 0.10$, values followed by different letters are significantly different.

Table 2. Oven-dry pupal weights of female western spruce budworm by rearing method and host species. a/

REARING METHOD	DOUGLAS-FIR	CORKBARK FIR
EXCISED FOLIAGE	20.8 (5) b/	19.2 (5)
FIELD BAGGED	16.4 (4)	18.7 (6)
FIELD COLLECTED	———	———
F-PROB.	0.4705	0.8974

a/ oneway ANOVA, $\alpha = 0.10$, LSD multiple range test, values followed by different letters are significantly different.
b/ numbers in parentheses are number of cases.

The technique of excising foliage for feeding experiments may lead to misleading results if inducible defensive mechanisms exist, because excised branches may not be able to respond to insect feeding by increasing concentrations of defensive compounds as would a normal branch. However, our study shows that feeding on excised foliage in the laboratory is probably a good approximation of feeding as it occurs under natural conditions. Bagging of insects on trees (white muslin bags) tends to increase pupal weights, probably as a result of reduced harassment from predators and parasites and possibly because of a modified microclimate.

Influence of Host Species

A commonly held belief among foresters is that some host species are more resistant to both WSBW and eastern spruce budworm, Choristoneura fumiferana Clemens, than are other hosts. We compared the relative suitability of three of the important hosts in Arizona (Table 3). Pupal weights were not significantly different among the three hosts, which indicates that all hosts are a suitable substrate for insect development. Feeding efficiency (ECI) and growth rate (RGR) were significantly higher on Douglas-fir than on either white fir or corkbark fir. However, budworm larvae can apparently compensate for the lower quality of the foliage, as reflected in low ECI and RGR, by increasing their consumption rate (I) and probably the duration of the feeding period. This ability of insects to compensate for lower food quality by increasing consumption is often overlooked in studies of host plant-herbivore interaction. Too few replicates of Engelmann spruce were available to include in this analysis; however, qualitatively it appeared as if WSBW did quite well on spruce foliage when the host growth and the herbivore requirements were synchronized.

Table 3. Nutritional indices and pupal weights for western spruce budworm reared on three host species in Arizona.

HOST	N	I (mg)	RCR	RGR	ECI	PUPAL WT. a/ (mg)
DOUGLAS-FIR	13	185.	1.7	0.12 A b/	7.8 A	58.6
WHITE FIR	20	226.	2.0	0.10 B	5.4 B	60.8
CORKBARK FIR	10	223.	1.7	0.09 B	5.8 B	60.0
F-PROB.		0.341	0.183	0.012	0.003	0.957

a/ pupal weight on fresh basis, all other values based on oven-dry weights.
b/ values followed by different letters are statistically significant, oneway ANOVA, LSD multiple range test, $\alpha = 0.10$.

Chemical Content of WSBW Host Foliage

Current year's foliage of selected host trees was collected seven to 10 days after the beginning of the feeding studies. Foliage was collected from the same trees and crown position as those used in the feeding studies. Tannin concentration did not vary significantly between host vigor categories for any of the species tested (Table 4). Engelmann spruce had approximately twice the tannin content of any other host. Total phenols were not different among vigor categories for Douglas-fir and white fir (Table 5). However, medium vigor corkbark fir trees had lower total phenols than did the high or low vigor trees. Low vigor Engelmann spruce trees had lower total phenols than high and medium vigor trees. Protein content of foliage tended to decrease with host vigor for all host species tested (Table 6). However, this trend was statistically significant only for Engelmann spruce.

Table 4. Percent dry weight of tannins in western spruce budworm host foliage by host vigor class. a/

VIGOR	DOUGLAS FIR	WHITE FIR	CORKBARK FIR	ENGELMANN SPRUCE
HIGH	4.78 A	5.64 A	4.51 A	9.12 A
MEDIUM	3.91 A	4.58 A	4.98 A	9.71 A
LOW	4.98 A	3.79 A	4.40 A	8.86 A
$\bar{X}$	4.49	4.66	4.63	9.28

a/ values followed by different letters are significantly different, oneway ANOVA, LSD multiple range test, $\alpha = 0.05$.

Table 5. Total phenol content of western spruce budworm host foliage by host vigor class. a/ b/

VIGOR	DOUGLAS FIR	WHITE FIR	CORKBARK FIR	ENGELMANN SPRUCE
HIGH	0.11 A	0.07 A	0.19 A	0.12 A
MEDIUM	0.10 A	0.08 A	0.13 B	0.13 A
LOW	0.09 A	0.06 A	0.18 A	0.08 B
$\bar{X}$	0.10	0.06	0.17	0.11

a/ total phenols expressed in terms of absorbance per mg.
b/ values followed by different letters are significantly different, oneway ANOVA, LSD multiple range test, $\alpha = 0.05$.

Table 6. Percent dry weight of protein in western spruce budworm host foliage by host vigor class. a/

VIGOR	DOUGLAS FIR	WHITE FIR	CORKBARK FIR	ENGELMANN SPRUCE
HIGH	14.62 A	21.17 A	22.76 A	23.68 A
MEDIUM	12.41 A	20.85 A	19.08 A	22.90 A
LOW	11.69 A	17.47 A	19.66 A	16.77 B
$\bar{X}$	12.66	19.93	20.57	21.36

a/ values followed by different letters are significantly different, oneway ANOVA, LSD multiple range test, $\alpha = 0.05$.

Relationship Between Foliage Chemistry and Insect Feeding

Pearson's correlation coefficients and a simple linear regression were used to evaluate the relationship between foliage chemistry and WSBW feeding. In each case foliage samples for chemical analysis and feeding were collected from the same aspect and crown position of the same tree. Only Douglas-fir and white fir are discussed here due to insufficient sample size for the other species.

Tannins in Douglas-fir were positively correlated with both foliage ingested and approximate digestibility (Table 7). These results were quite surprising considering that the theory of plant apparency (Feeny 1976, Rhoades and Cates 1976) would predict that the quality of the foliage and subsequent consumption should decrease as tannins increase. Phenols are not significantly correlated with any of the larval growth parameters studied. Protein is positively correlated with pupal weight. Adjusted R^2 values presented in Table 8 indicate that leaf tannin content can be used to predict the amount of Douglas-fir foliage ingested and that protein content is a useful predictor of pupal weight.

Table 7. Correlation coefficients indicating the relationship between Douglas-fir foliage chemistry and western spruce budworm larval growth parameters (n = 7).

FOLIAGE CHEMICALS	LARVAL GROWTH PARAMETERS					
	INGESTED	RCR	RGR	AD	ECI	PUPAL WT.
TANNINS	0.72 a/	0.55	0.62	0.76 b/	-0.25	0.65
PHENOLS	0.26	-0.24	-0.37	-0.46	0.06	-0.38
PROTEINS	0.64	0.01	0.62	0.41	0.26	0.70 a/

a/ $P \leq 0.10$.
b/ $P \leq 0.05$.

The relationships between foliage chemicals and larval growth parameters for white fir are different from those observed for Douglas-fir. In this case tannins are negatively correlated with growth rate and efficiency (Table 9). This is consistent with the plant apparency theory. Adjusted R^2 values indicate foliage tannin content can be used to predict growth rate and efficiency (Table 10). Neither phenols nor proteins are correlated with any measure of larval growth for WSBW feeding on white fir foliage.

Table 8. Adjusted R^2 values for simple regression equations using Douglas-fir foliage chemicals to predict western spruce budworm larval growth (n = 7). d/

FOLIAGE CHEMICALS (x)	LARVAL GROWTH PARAMETERS (Y)					
	INGESTED	RCR	RGR	AD	ECI	PUPAL WT.
TANNINS	0.40 a/	NS c/	NS	NS	NS	NS
PHENOLS	NS	NS	NS	NS	NS	NS
PROTEINS	NS	NS	NS	NS	NS	0.57 b/

a/ $P \leq 0.10$.
b/ $P \leq 0.05$.
c/ NS; not significant.
d/ data transformed where appropriate to best fit the form of the relationship.

Table 9. Correlation coefficients indicating the relationship between white fir foliage chemistry and western spruce budworm larval growth parameters (n = 8).

FOLIAGE CHEMICALS	LARVAL GROWTH PARAMETERS					
	INGESTED	RCR	RGR	AD	ECI	PUPAL WT.
TANNINS	0.44	0.42	-0.68 a/	0.45	-0.85 b/	-0.22
PHENOLS	-0.28	-0.33	-0.38	-0.17	-0.37	-0.27
PROTEINS	0.18	0.14	0.15	0.12	0.13	0.25

a/ $P \leq 0.10$.
b/ $P \leq 0.05$.

Table 10. Adjusted R^2 values for simple regression equations using white fir foliage chemicals to predict western spruce budworm larval growth (n = 8). d/

FOLIAGE CHEMICALS (x)	LARVAL GROWTH PARAMETERS (Y)					
	INGESTED	RCR	RGR	AD	ECI	PUPAL WT.
TANNINS	NS a/	NS	0.38 b/	NS	0.64 c/	NS
PHENOLS	NS	NS	NS	NS	NS	NS
PROTEINS	NS	NS	NS	NS	NS	NS

a/ NS; not significant.
b/ $P \leq 0.10$.
c/ $P \leq 0.05$.
d/ data transformed where appropriate to best fit the form of the relationship.

Douglas-fir, corkbark fir, white fir, and probably Engelmann spruce are all suitable host material for WSBW when current foliage flush is synchronized with the larval feeding period. Douglas-fir is of slightly better quality than the other species tested. However, because of the ability of the budworm to compensate for lower food quality by consuming more foliage, larvae can grow and develop quite well on lower foliage quality hosts. It is possible that white fir and corkbark fir could actually sustain more defoliation damage than Douglas-fir because their foliage is of lower quality. This is important to keep in mind when considering low foliage quality as a criterion for selecting resistant species or genotypes. In northern Arizona, Douglas-fir and true firs typically sustain more damage than Engelmann spruce, but this may be primarily because spruce is not phenologically synchronized with the feeding cycle of the WSBW.

There are suprisingly few differences in the tannins and phenols tested among vigor categories. Our study fails to show any evidence that high vigor trees maintain higher levels of defensive chemicals than do low vigor trees. Nor is there any evidence to suggest that by excising foliage, and presumably preventing induction of defensive compounds, insects can perform better than if they feed on a host which could actively defend itself. There may be differences in protein content between vigor categories, but even in the worst case (foliage with only 11% protein) there appears to be adequate protein to sustain insect development.

Differences in tannins, phenols, and proteins among species are more striking. Tannins are almost twice as abundant in Engelmann spruce as in the other species. But, considering the different relationship between tannins in white fir and Douglas-fir and feeding parameters, it is uncertain whether high tannin content has a negative or positive effect. There are differences in phenols between species but, here again, there is no apparent relationship to feeding parameters. Douglas-fir, which based on efficiency and growth rate of larvae is the best host, actually had the lowest average protein content of the host species tested.

Phenols do not appear to influence feeding by WSBW even though the phenolic content in budworm hosts is within the concentration range that has been reported to influence host selection by aphids (Zucker 1982).

The relationship between foliage tannins and some of the larval growth parameters is intriguing. It is likely that the tannins are not identical in Douglas-fir and white fir. But, the levels of tannins are well within the range found by Feeny and Bostock (1968) who argued that higher tannin content is equated with lower food quality. Bernays (1981) suggests that tannins may actually be used as a nutrient source by grasshoppers. In Douglas-fir, tannins were positively correlated with digestibility. In white fir, tannins were negatively correlated with growth and

efficiency. It is difficult to explain these apparently opposite results, except that tannins and proteins are intercorrelated in Douglas-fir which may result in a random correlation. Clearly we know little about the role of foliage tannins in larval feeding and growth in the WSBW.

Literature Cited

Bean, J.L. and H.O. Blatzer. Mean head width for spruce budworm larval instars in Minnesota and associated data. Journal of Economic Entomology. 50:499; 1957.

Bernays, E.A. Plant tannins and insect herbivores: an appraisal. Ecological Entomology. 6:353-360; 1981.

Bradford, M.M. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Analytical Biochemistry. 72:248-254; 1976.

Feeny, P.P. Plant apparency and plant defense. In: Biochemical Interactions Between Plant and Insects J. Wallace and R. Mansell, eds. Recent Advances in Phytochemistry. 10:1-40; 1976.

Feeny, P.P. and H. Bostock. Seasonal changes in the tannin content of oak leaves. Phytochemistry 7:871-880; 1968.

Kutscha, Norman P. and Irving B. Sachs. Color tests for differentiating heartwood and sapwood in certain softwood tree species. Report No. 2246. U.S. Department of Agriculture, Forest Products Laboratory; 1962. 14 pp.

Levin, D.A. Plant phenolics: an ecological perspective. The American Naturalist. 105(942):157-181; 1971.

Mattson, W.J. Jr. Herbivory in relation to plant nitrogen content. Annual Review of Ecology and Systematics. 11:119-161; 1980.

McDonald, G.I. Differential defoliation of neighboring Douglas-fir trees by western spruce budworm. Research Note INT-306. USDA Forest Service; 1981.

McMurray, T.I. Effects of drought stress induced changes in host foliage quality on the growth of two forest insect pests. MS Thesis, University of New Mexico; 1980. 56 pp.

Price, M.L., S. VanScoyoc and L.G. Butler. A critical evaluation of the vanillin reaction as an assay for tannin in sorghum grain. Journal of Agricultural Chemistry. 26(5):1214-1218; 1978.

Rhoades, D.F. and R.C. Cates. Toward a general theory of plant and anti-herbivore chemistry. In: Biochemical Interactions Between Plants and Insects J. Wallace and R. Mansell eds. Recent Advances in Phytochemistry. 10:168-213; 1976.

Ribereau-Gayon, P. Plant Phenolics Hafner, New York; 1972. p 47.

Robertson, J.L. Rearing the western spruce budworm. Canada/US. Spruce Budworm Program Publication; 1979.

Swain, T. Secondary compounds as protective agents. Annual Review of Plant Physiology. 28:479-501; 1977.

Swain, T. and W.E. Hillis. The phenolic constituents of Prunus domestica I. The quantative analysis of phenolic constituents. Journal of the Science of Food and Agriculture. 10:63-68; 1959.

Swain, T. Tannins and lignins. In: Herbivores: Their Interactions With Secondary Plant Metabolites. G.A. Rosenthal and D.H. Janzen eds. Academic Press, New York; 1979. 744 pp.

Wagg, J.W. Bruce. Environmental factors affecting spruce budworm growth. Forest Lands Research Center, Corvallis OR. 1958. Res. Bull. 11: 27 pp.

Waldbauer, G.P. The consumption and utilization of food by insects. Advances in Insect Physiology. 5:229-288; 1968.

Waring, R.H., W.G. Thies and D. Muscato. Stem growth per unit of leaf area: a measure of tree vigor. Forest Science. 26(1):112-117; 1980.

Zucker, W.V. How aphids choose leaves: the roles of phenolics in host selection by a galling aphid. Ecology. 63(4):972-981; 1982.