

AN ABSTRACT OF THE THESIS OF

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Title: The Effect of Broadcast Burning on the Quality of
Winter Forage for Elk, Western Oregon.

Abstract approval: *Redacted for Privacy*
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The Roosevelt elk (Cervus elephus roosevelti) is a National Forest management indicator species on the westside of the Cascade mountains, Western Oregon. A Habitat Effectiveness model is used by State and Federal agencies to evaluate elk habitat in the region. Concerns about the model's lack of differentiation between winter and summer ranges in the analyses and assumptions that burning will increase forage quality on winter range prompted this study. I investigated the effect of broadcast burning, plant association, and time since disturbance on the quality of trailing blackberry (Rubus ulna), red huckleberry (Vaccinium parviflorum), willow (Salix spp.), vine maple (Acer circinatum), salal (Gaultheria shallon), and red elderberry

(Sambucus racemosa). Crude protein, neutral detergent fiber, acid detergent fiber, acid insoluble ash, lignin, astringency, condensed tannin, and hydrolyzable tannin contents were measured.

No significant effects of burning, plant association, or age were observed for crude protein, hydrolyzable tannins, or neutral detergent fiber. Crude protein varied among taxa: trailing blackberry contained 9.65%, and the other taxa ranged from 5.21-7.24%. Neutral detergent fiber was highly variable: trailing blackberry contained 30.90%, and the other taxa ranged from 52.20%-65.06%. Acid detergent fiber content ranged from 44.88%-49.49% for all taxa except trailing blackberry (17.78%).

Lignin varied among taxa: trailing blackberry had the lowest content (6.37%) and salal had the highest (30.25%). Lignin content in salal was greater on recently disturbed sites.

Astringency ranged from 0.0015 mg protein precipitated per mg plant tissue in vine maple to 0.6737 in trailing blackberry. Salal and willow had intermediate astringencies: elderberry, huckleberry, and vine maple had the lowest. Hydrolyzable tannins were present in all species except red elderberry. Burning and plant association effected astringency and condensed tannin content in trailing blackberry and huckleberry. Samples from burned, very dry and resource-poor sites had higher astringencies than on similar unburned units and non-resource-limiting sites.

Condensed tannin contents increased with unit age in salal, huckleberry, and trailing blackberry, possibly accumulating during peak years of re-establishment after disturbance.

Vine maple and red huckleberry were the only taxa with positive digestible protein levels. Digestible protein content may be higher in winter forage on less severe sites.

Elk forage enhancement in winter range should be evaluated on a site-specific basis. Burning did not promote a detectable increase in quality for these forage taxa, and it decreased the quality of species sensitive to site conditions.

The Effect of Broadcast Burning on the Quality of
Winter Forage for Elk, Western Oregon

by

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This thesis is dedicated to the memory of my father, Donald William Friesen. He passed away before seeing his oldest daughter become a wildlife biologist. He showed a wide-eyed youngster the wonders of nature, and I like to believe he would be proud of my professional endeavors.

TABLE OF CONTENTS

INTRODUCTION

Literature Review	1
Study Objectives	4
Hypotheses to be Tested	5

STUDY AREA AND METHODS

Study Area	6
Methods	7
Winter Forage Species Determination	7
Sampling Design	11
Unit Selection Criteria	14
Plant Sampling	16
Laboratory Analyses	18
Statistical Analyses	19

RESULTS

Sampling Design	22
Forage Species	23
Effect of Treatment, Plant Association, and Age on Quality	24
Crude Protein, NDF, ADF, Lignin, and AIA	24
Condensed Tannin	27
Hydrolyzable Tannin	30
Astringency	30
Dry Matter Digestibility	34
Digestible Protein	37

DISCUSSION

Forage Species	42
Forage Quality	45
Crude Protein	45
Lignin	49
Condensed Tannin and Astringency	50
Condensed Tannin	54
Hydrolyzable Tannin	57
Total Nutrient Availability for Elk	58
Forage Summary and Management Recommendations	62

LITERATURE CITED	66
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APPENDICES	80
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LIST OF FIGURES

Figure

1. Transect layout used for measuring percent cover of plant species on cut-units. 17
2. Dry matter digestibility of forage species on the Willamette National Forest, Oregon (winter 1989/90). 34
3. Digestible protein of forage species on the Willamette National Forest, Oregon (winter 1989/90). 38
4. Effect of treatment on digestible protein of forage species on the Willamette National Forest (winter 1989/90). 41

LIST OF TABLES

Table

1.	Common names, scientific names, and codes of plants used as elk forage and in plant association descriptions on the Willamette National Forest, Oregon.	8
2.	Dominant understory species on plant association groups important on elk winter range.	10
3.	Preferred winter forage species for Roosevelt elk, Westside Oregon Cascades, as predicted by a cadre of wildlife biologists (1989), and available literature.	12
4.	Plant association groups on the Willamette National Forest, Oregon used to stratify forage sampling.	13
5.	Average crude protein, neutral detergent fiber, and acid detergent fiber for selected winter forage on the Willamette National Forest, Oregon.	25
6.	Average percent content of lignin and acid insoluble ash for selected forage species on the Willamette National Forest, Oregon.	26
7.	Average astringency, condensed tannin, and hydrolyzable tannin content of selected forage species on the Willamette National Forest.	28
8.	Average condensed tannin content of selected forage species by plant association and treatment on the Willamette National Forest, Oregon.	31
9.	Average astringencies of selected forage by treatment and plant association on the Willamette National Forest.	33
10.	Average dry matter digestibility of selected forage by plant association on the Willamette National Forest, Oregon.	36
11.	Results of correlation analyses of dry matter digestibility with lignin and astringency for winter forage species on the Willamette National Forest.	37
12.	Average digestible protein of selected forage by plant association on the Willamette National Forest, Oregon.	40

THE EFFECT OF BROADCAST BURNING
ON THE QUALITY OF WINTER FORAGE FOR ELK,
WESTERN OREGON

INTRODUCTION

LITERATURE REVIEW

Roosevelt elk (Cervus elaphus roosevelti) occur in Coastal and Cascade mountain areas of Oregon, Northern California, Washington, and Vancouver Island, British Columbia, Canada. Historically, their numbers have been low on the west slopes of the Cascades in Oregon (Bryant and Maser 1982). Successful reintroductions have increased the populations (Mace 1971). Some populations on the west slopes are migratory and some maintain the same range throughout the year (Ten Eyck and Ferry 1990). Winter range on the westside is considered a limiting habitat component, and has been defined as areas where < 0.7 m of snow is permanently present from December through April (Harshman and Jubber 1984).

Elk winter range on the westside is intensively managed for production of wood products. National Forest management considerations for elk and the ability to sustain quality habitat is being evaluated by State and Federal agencies (Thomas 1979). The Westside Elk Model (Wisdom et al. 1986) was developed to aid agencies in managing for elk on

roads. The Willamette National Forest has adopted this model to assess elk habitat effectiveness (U.S.D.A. 1990). Questions have been raised concerning the model's evaluation of forage quality. Differentiation between winter and summer ranges is not presently incorporated into the analyses. Two assumptions are made in assessing forage within the model: 1) forage quantity is not limiting in the westside Cascades at any time of the year, and 2) forage quality may be limiting. Broadcast burning, seeding, and fertilizing are assumed to increase forage quality in the model. The assumption that higher quality values should be assigned to all burned clearcuts on summer and winter ranges is a major point of contention in the Cascades. A modified Delphi Method (Dalkey 1969) was used to establish quality ratings for forage areas within the Westside Model. Validation of the experts' assumptions is still required.

More than any other factor, the digestibility and nutritional content of food within forage areas may limit elk productivity in Western Oregon (Trainer 1971, Mereszczak et al. 1981, Starkey et al. 1982). Big game body condition varies directly with changes in forage quality (Mautz 1978). As the nutritive content of forage species falls from spring through winter, physical maintenance through the critical winter months may be significantly effected by inadequate supplies of quality forage. Knowledge of food habits is

essential for elk habitat management planning and the evaluation of management practices. Relatively little is known about elk diet quality and nutritive requirements to sustain healthy, reproductive herds (Nelson and Leege 1982). Several studies have examined food habits of elk in Western Washington (Hanley 1980, Leslie et al. 1984, Merrill et al. 1987, Schoen 1977, Happe et al. 1990), Alaska (Batchelor 1965), British Columbia (Janz 1983, Youds et al. 1985), Southwestern Oregon (Harper 1985), and Northwestern California (Harper et al. 1967). Literature documenting the preferred forage species and their relative quality in the Western Oregon Cascades is inadequate.

Post-harvest treatments, including burning, fertilizing, and seeding increase the use of forage by elk on some cutover areas (Witmer et al. 1985). Use of fire is being increasingly scrutinized by those in the public concerned with air quality. Prescribed fire use is subjected to substantial regulation, and increasing air resource protection will demand clear justification of fire use. The influence of burning on the quality of winter forage in the westside Cascades is unknown.

Factors that effect forage quality are numerous. Interactions among factors complicate an attempt to isolate single influences. Dietary factors traditionally evaluated

in forage quality studies include crude protein and lignin content. These measurements are general indicators of potential nutrient availability to the animal. Further investigations including secondary compound content are needed to indicate true nutrient availability (Robbins et al. 1987a). The effect of fire on the secondary compound content of forage species is not known, and the potential influence needs to be evaluated.

Prediction of big game forage production may be possible from pre-harvest plant association classification (Hemstrom et al. 1987, Dyrness 1973). Plant associations have been evaluated for their production of big game forage before harvest (Hemstrom et al. 1987). Plant communities are indicators of soils, water, nutrient pools, and climatic conditions, and fire may differentially effect the quality of forage within these communities, but this has not been examined.

STUDY OBJECTIVES

My primary objective was to evaluate the effect of broadcast burning on the quality of selected elk winter forage species. Stratifying the effects of post-harvest broadcast burning by plant association could aid in site-

forage species. Stratifying the effects of post-harvest broadcast burning by plant association could aid in site-specific management plans containing alternatives prescribing fire. Investigating changes in quality over time since disturbance could provide short-term versus long-term cost/benefit information.

HYPOTHESES TO BE TESTED

The hypotheses to be tested include:

- Ho: Broadcast burning has no effect on forage quality, measured by astringency, lignin, crude protein, acid insoluble ash, and fiber content.
- Ho: Quality of selected browse species does not differ from one plant association to another.
- Ho: Quality of selected browse species does not change over a 10-year period.
- Ho: Quality of selected winter forage for elk is consistently effected by burning among plant associations and age classes.

STUDY AREA AND METHODS

STUDY AREA

The study area was the Willamette National Forest in West-central Oregon on the west slopes of the Cascade mountains. The Forest consists of approximately 688,260 ha (1.7 million acres): 608,502 ha (1.5 million acres) are forested. Elk winter range occupies 43% of the forested landbase (U.S.D.A. 1991). The Willamette National Forest is the highest timber-volume producing National Forest in the National Forest system (U.S.D.A. 1990), and its intensive harvesting history has created a highly fragmented landscape. Optimal thermal cover (old-growth) comprises 34% of the land base (U.S.D.A. 1989) and 24% is in managed 0- to 50-year-old stands (personal commun. Jim Mayo, Forest Timber Planner).

Annual precipitation in the area ranges from about 150 cm in local rain shadows to > 325 cm on some high ridges (U.S. Weather Bureau 1964). The precipitation patterns are highly influenced by topography. The south half of the Forest is drier than the north half, which significantly influences the distribution of plant associations. Four conifer series occur on the Forest (Hemstrom et al. 1987): Western Hemlock, Mountain Hemlock, Pacific Silver Fir and Grand Fir, and Douglas-Fir (see Table 1 for plant species

scientific names and codes). The Douglas-fir and Western hemlock series (Table 2) were important in this study because of their elevational distribution, shrub and forb components, and big game use. Warm, relatively dry slopes with thin, rocky soils are occupied by the Douglas-fir series. This series is rare north of the Mckenzie River, but fairly common in winter range on the southern end of the Forest. The more moist sites are occupied by the Western hemlock series, which includes the majority of the lower elevation forest.

Elevations on the Willamette range from 244 m to 3048 m. Snowpacks ranging from 150-250 cm are present throughout the winter above 1372 m. Elk winter range is generally considered to fall below 1066 m. Migratory animals use the higher elevations (> 1524 m) to the east during the summer months, migrating to lower elevations (< 1066 m) to the west prior to snowpack (Ten Eyck and Ferry 1990).

METHODS

Winter Forage Species Determination

This study was limited to selected winter forage species on the Willamette National Forest. A thorough investigation of seasonal diet compositions had not been completed for the

Table 1: Common names, scientific names, and codes of plants used as elk forage and in plant association descriptions on the Willamette National Forest, Oregon.

TREES

Douglas-fir	PSME	<i>Pseudotsuga menziesii</i>
Grand Fir	ABGR	<i>Abies grandis</i>
Mountain hemlock	TSME	<i>Tsuga mertensiana</i>
Pacific Silver Fir	ABAM	<i>Abies amabilis</i>
Western hemlock	TSHE	<i>Tsuga heterophylla</i>

SHRUBS

Baldhip Rose	ROGY	<i>Rosa gymnocarpa</i>
California Hazel	COCO2	<i>Corylus cornuta</i>
Chinquapin	CACH	<i>Castanopsis chrysophylla</i>
Dwarf Oregon grape	BENE	<i>Berberis nervosa</i>
Oceanspray	HODI	<i>Holodiscus discolor</i>
Oregon Boxwood	PAMY	<i>Pachistima myrsinites</i>
Poison oak	RHDI	<i>Rhus diversiloba</i>
Prince's Pine	CHUM	<i>Chimaphila umbellata</i>
Red huckleberry	VAPA	<i>Vaccinium parvifolium</i>
Rhododendron	RHMA	<i>Rhododendron macrophyllum</i>
Salal	GASH	<i>Gaultheria shallon</i>
Snowberry	SYAL	<i>Symphoricarpos albus</i>
Tall Oregon grape	BEAQ	<i>Berberis aquifilium</i>
Trailing blackberry	RUUR	<i>Rubus ursinus</i>
Vine Maple	ACCI	<i>Acer circinatum</i>
Whipple vine	WHMO	<i>Whipplea modesta</i>

Table 1 Continued

Herbaceous species

Beargrass	XETE	<i>Xerophyllum tenax</i>
Bluntleaf sandwort	ARMA3	<i>Arenaria macrophylla</i>
Bracken fern	PTAQ	<i>Pteridium aquilinum</i>
Coolwort foamflower	TITR	<i>Tiarella trifoliata</i>
Fairybells	DIHO	<i>Disporum hookeri</i>
False solomonseal	SMST	<i>Smilacina stellata</i>
Festuca species	FESTU	<i>Festuca</i>
Hairy hawkweed	HIAL	<i>Hieracium albiflorum</i>
Inside-out flower	VAHE	<i>Vancouveria hexandra</i>
Oregon Oxalis	OXOR	<i>Oxalis oregana</i>
Pacific trillium	TROV	<i>Trillium ovatum</i>
Pathfinder	ADBI	<i>Adenocaulon bicolor</i>
Queencup beadlilly	CLUN	<i>Clintonia uniflora</i>
Rattlesnake plantain	GOOB	<i>Goodyera oblongifolia</i>
Redwoods violet	WISE	<i>Viola sempervirens</i>
Smallflower nemophila	NEPA	<i>Nemophila paviflora</i>
Snow queen	SYRE	<i>Synthyris reniformis</i>
Strawberry	FRAGA	<i>Fragaria</i> sp.
Sweetscented bedstraw	GATR	<i>Galium triflorum</i>
Swordfern	POMU	<i>Polystichum munitum</i>
Three-leaved anemone	ADNE	<i>Anemone deltoidea</i>
Twinflower	LIBO2	<i>Linnaea borealis</i>
Vanilla leaf	ACTR	<i>Achlys triphylla</i>
Western starflower	TRLA2	<i>Trientalis latifolia</i>

TABLE 2: Dominant understory species on plant association groups important on elk winter range. From Hemstrom et al. 1987. (ordered by moisture regime from driest to wettest)

PLANT ASSOCIATION GROUP	DOMINANT UNDERSTORY SPECIES	
	SHRUBS	HERBACEOUS LAYER
DOUGLAS-FIR	Oregon grape California Hazel Oceanspray Baldhip rose Snowberry Whipple vine Poison oak Tall Oregon grape Vine maple Trailing blackberry	Swordfern Snowqueen Pathfinder Three-leaved anenome Hairy hawkweed Strawberry Western starflower <u>Festuca</u> species Bluntleaf sandwort Smallflower nemophila
DOUGLAS-FIR/ WESTERN HEMLOCK	Oregon grape Baldhip rose Snowberry Vine maple Oregon boxwood California hazel Whipple vine Salal Oceanspray	Vanilla leaf Pathfinder Swordfern Three-leaved anenome Western starflower Twinflower Redwoods violet Bracken fern
WESTERN HEMLOCK/ RHODODENDRON	Rhododendron Vine maple Salal Oregon grape Chinquapin Red huckleberry Prince's pine Trailing blackberry	Beargrass Twinflower Swordfern Pacific trillium Redwoods violet Rattlesnake plantain
WESTERN HEMLOCK	Oregon grape Vine maple Snowberry Salal Baldhip rose Red huckleberry Trailing blackberry	Vanilla leaf Coolwort foamflower False solomonseal Queencup beadlelilly Swordfern Pathfinder Three-leaved anenome Oxalis Fairybells Sweetscented bedstraw Pacific trillium Inside-out flower Twinflower Redwoods violet

the West slopes of the Cascades, so I surveyed 24 wildlife biologists with experience in elk management in the Pacific Northwest to determine which winter forage species might be selected by elk, and I reviewed 2 literature sources for applicable information. The 13 sources (biologists and literature) (Appendix A) suggested 34 species (Table 3), and 6 taxa were selected for sampling: trailing blackberry, salal, red huckleberry, willows, vine maple, and red elderberry. Biologists that did not respond to the survey usually declined because of a lack of first-hand knowledge.

Sampling Design

My objective was to evaluate the influence of broadcast burning, pre-harvest plant association, and time since disturbance on forage quality. There were 2 site-preparation treatments (broadcast burn post-harvest and no slash disposal); 4 plant association groups (Table 4) developed with the assistance of Dr. Miles Hemstrom, USFS Ecologist; and 3 age classes (1-2, 3-5, and 7-10 years) since clearcutting. We determined which plant associations fell within winter range and selected those that had shown big-game use during a previous investigation (Hemstrom et al. 1987).

Table 3: Preferred winter forage species for Roosevelt elk, Westside Oregon Cascades, as predicted by a cadre of wildlife biologists (1989), and available literature. (* indicates species chosen for sampling)

PLANT SPECIES

* Red Huckleberry
 * Trailing blackberry
 * Vine Maple
 * Willow
 Grasses
 * Salal
 * Red Elderberry
 Red Cedar
 Deerbrush Ceanothus
 Swordfern
 Sedge
 Snowbrush
 Madrone
 Pacific yew
 Oregon grape
 Salmonberry
 Deerfern
 Red alder
 Western Hemlock
 White Hairy Hawkweed
 Indian lettuce
 Woodrush Sedge
 Wedgeleaf Ceanothus
 Evergreen Huckleberry
 Lilac
 Baccharis
Ceanothus sp.
 Rose
 Miner's lettuce
 Thimbleberry
 Ginger
 Whipple vine
 Silk tassle

TABLE 4: Plant association groups on the Willamette National Forest, Oregon used to stratify forage sampling (from Hemstrom et al. 1987)

GROUP	COMMON CHARACTERISTICS
PSME/HODI/BENE /WHMO /SYMA /GRASS	:Very hot, dry, high moisture stress :Thin, shallow, skeletal soils, rocky or deep heavy clay :Low in coarse fragments and high in clay :Slash fires that consume duff may accelerate dry ravel of skeletal soils and reduce already low nitrogen levels
PSME-TSHE/BENE /GASH	:Dry and hot :Southerly aspects, often steep & rocky :Draughty, skeletal to relatively deep clay soils :Transitional to TSHE series :Slash fires that consume duff may accelerate dry ravel of the skeletal soils and reduce already low nitrogen levels
TSHE/RHMA-BENE /RHMA-GASH /RHMA-XETE	:Dry, exposed upper slopes & ridges :Soils well-drained and often rocky :Nitrogen limiting on some sites
TSHE/OXOR /POMU /ACTR /BENE /BENE-OXOR /BENE-ACTR	:Relatively deep soils, often with substantial coarse fragment content :Often moist into summer, though some drought on the driest sites :Soils should be generally resistant to impacts from slash fires, but those with rockier profiles may be affected by duff reduction

The study design included 3 replications of 24 unique site-types:

burned vs. unburned

3 time-since-disturbance classes

4 plant associations.

Seventy-two cut-units were required to meet the objectives.

Unit Selection Criteria

The following criteria were used to select cut-units. The units had to be located at < 1067 m. Units had to be 8-16 ha (20-40 acres) in size, have slopes < 50%, and have southerly aspects (characteristics that would enhance the chance of elk use) (Harper and Swanson 1970, Schoen 1977, Witmer 1981, Ten Eyck and Ferry 1990, Witmer et al. 1985, Wisdom et al. 1986). Elk use of cut-units for foraging predominantly occurs in the early years of vegetation establishment and diminishes as the unit grows into hiding cover (Witmer et al. 1985). I selected units \leq 10 years old that would be categorized as forage areas. Records were available to indicate the year and month the units were harvested. Only units treated with a broadcast burn (preferably spring) or no slash disposal were selected for the study. No units were selected with any other treatments (i.e., big-game repellents, browse cutbacks). It was also

desirable to have records of the dominant plant species prior to harvest of the unit.

A pool of possible units was obtained from the U.S. Forest Service's Timber Resource Information (TRI) database in Fort Collins, Colorado. Units that appeared to meet the above criteria were mapped and verified in the field. I found information relating to plant association types to be highly inaccurate, and a verification of plant association was conducted by keying stands adjacent to the cut-units using the Willamette National Forest's Plant Association Guide (Hemstrom et al. 1987). If an adjacent, non-harvested stand was not available, the cut-unit was not used for the study. Potential units were also determined by reading Environmental Assessments on sales in winter range areas, and through conversations with District timber and fire personnel. Finding unburned units was the most difficult task, and District personnel were used extensively to find these areas. Quite often information on the unburned units was only available by questioning personnel about units with broadcast burning in their prescription that had never been treated because of weather conditions.

Plant Sampling

A minimum of 20 plants of each taxa were clipped (where possible) in each unit to obtain 20 g of dry-weight material. Clippings included the new-year's growth (terminal bud plus approximately 2.5 cm of twig) on shrubs and the leaves of evergreens. In units that contained fewer than 20 plants of a given species, I collected clippings from every available plant. The 20 clippings were combined to form a composite sample to eliminate within-site variability. No plants were clipped within a 100-m of the adjacent stand to eliminate edge effects. The composite samples for each species from each unit were coded and stored in a freezer until dried for laboratory analyses.

All plants observed on the units were recorded, and a field notes on the preferred species abundance and distribution were maintained. The plants were recorded as either distributed throughout the unit randomly or as occurring in isolated clumps consisting of a few plants.

I used transects to determine the percent cover of all species located on the cut-units. The center of the cut-unit was determined and 6 45-m transects were placed in a S-shaped pattern (Figure 1). All plant species that were intercepted by the transect line were recorded, and their intercept

length was measured (Daubenmire 1959, Floyd and Anderson 1987, Hanley 1978). The placement of the transects varied with the size of the cut-unit: for units < 16 ha, I used the designed layout exactly. In units > 16 ha, or of odd sizes, I inserted 15-30 m of "empty transect" between sampled areas. This ensured broader distribution and reduced concentration of sampling in the center of the unit.

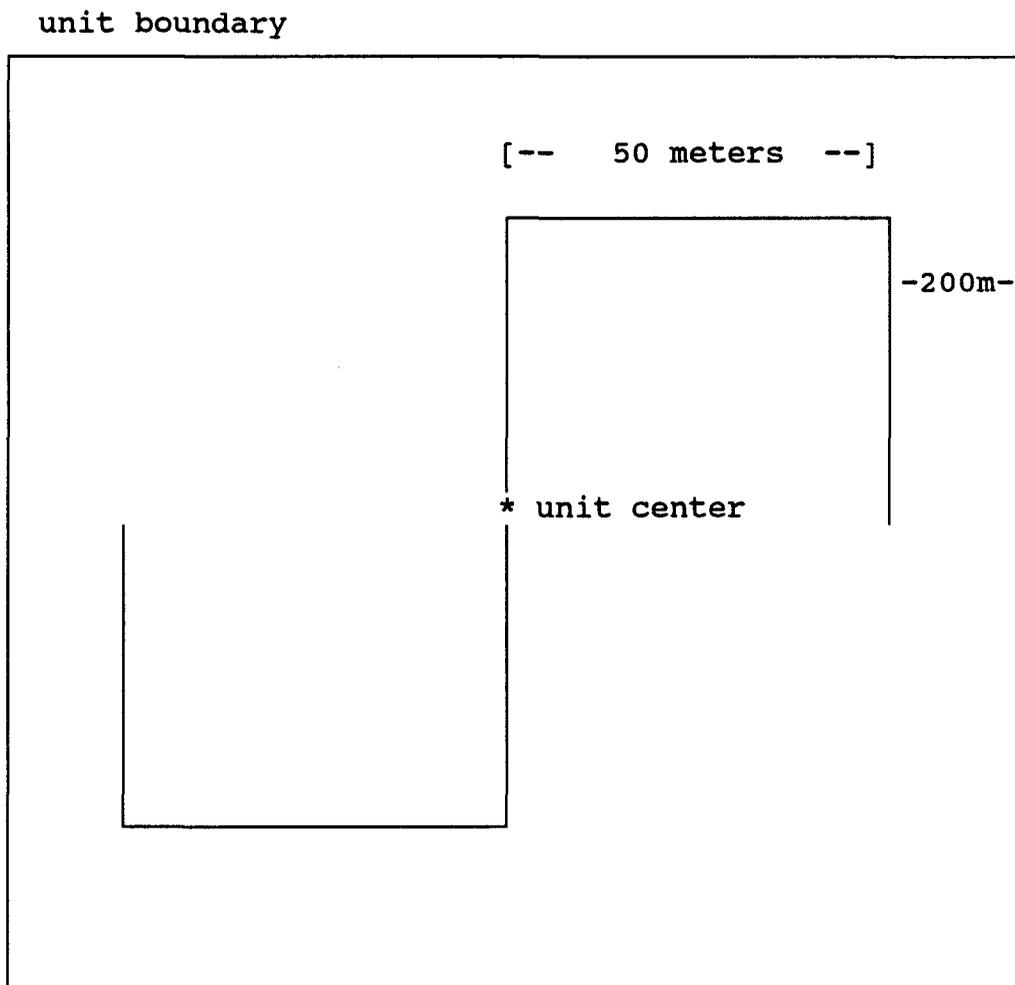


Figure 1: Transect layout used for measuring percent cover of plant species on cut-units.

Forage species were clipped during this part of the study primarily along the transect lines, though further searches of the unit were often required to find 20 plants of each species.

Use of the transects was abandoned after the first replication because of heavy snow cover. During the second and third replications, the field notes were maintained on the observed abundance and distribution of the forage species, and all plant species observed on the site were recorded. Plants were selected for clipping in a random fashion: we divided the unit into halves and walked a zig-zag pattern from one end of the cut-unit to the other. Often an intensive search was required to find rare and isolated plants, involving searching most of the unit.

Laboratory Analyses

Initial analyses of the plant samples was conducted by Washington State University's Habitat Laboratory, Pullman. The frozen samples were air-dried at 40 degrees celcius. Crude protein (CP) was determined using the Kjeldahl procedure and multiplying the resultant percent of nitrogen by 6.25. Neutral Detergent Fiber (NDF) and Acid Detergent Fiber (ADF) analyses methods were used to determine the

percent of cell wall total fiber and lignocellulose, respectively (Goering and Van Soest 1970, Van Soest and Wine 1967). The referenced procedures were used with one exception: the sodium sulfite reagent was not used in the analysis (Van Soest 1983, 1963). The percent-content of Acid Insoluble Ash (AIA) was determined sequentially from the Acid Detergent Fiber analyses. By ashing the ADF residue, which consists of cellulose, lignin, cutin, and acid-insoluble ash, the crude lignin fraction was determined (Goering and Van Soest 1970).

Additional laboratory analyses were conducted at Oregon State University's Range Laboratory, Corvallis. Each plant sample was analyzed to determine: astringency, which is the amount of protein that the plant tannins could precipitate (Asquith and Butler 1985, Happe 1990); the amount of condensed and hydrolyzable tannins (Happe 1990); and whether tannins were involved in protein extraction using the protein precipitating phenolic assay (Hagerman and Butler 1978, Happe 1990). (See Appendix B for complete laboratory method description)

Statistical Analyses

Raw data from the 8 laboratory analyses described above

(crude protein, lignin, ADF, NDF, AIA, astringency, condensed tannins, and hydrolyzable tannins) were analyzed using SAS General Linear Model (GLM) analysis (Ray 1982) for each species. The model was designed to detect differences between treatments and among ages and plant associations and all possible interactions at both $p < 0.05$ and $p < 0.10$ levels. Gibbons (1976) recommended increasing the level of significance when multiple comparisons are used. The small sample sizes involved with the study warrant a larger p -value. Increasing the power of the analyses was also accomplished by increasing the p -value. The assumption of normality necessary for valid interpretation of Analysis of Variance (ANOVA) output was evaluated by plotting the residuals from the GLM and evaluating the W and $p(W)$ values (Zar 1984). Generally, if $W < 0.90$, the data were transformed using recommendations by Sabin and Stafford (1990). If transformed data were non-normal, the data were ranked prior to ANOVA. The data in either the raw, log transformation, or ranked form were used in all subsequent statistical analyses. The Tukey Test (Zar 1984) was used for each dependent variable to reveal any significant differences between burned and non-burned sites ($p < 0.05$ and $p < 0.10$).

GLM output that indicated any significant differences between independent variables were further analyzed using Least Square Means (LSMEANS) (Ray 1982). The interacting

independent variables were built into the LSMEANS model to indicate exactly which plant associations, ages, and treatments resulted in the differences at the 0.05 and 0.10 levels.

Data from the forage quality analyses were used to compute digestible protein (DP) (Robbins et al. 1987a):

$$DP = [-3.87 + 0.9283X - (11.82 * ASTRINGENCY)]$$

(note: X = crude protein in percent)

and dry matter digestibility (DMD) (Robbins et al. 1987b):

$$DMD = [0.9231e^{-0.0481x} - 0.03(AIA)] [NDF] \\ + [(-16.03 + 1.02NDS) - 2.8(ASTRINGENCY)]$$

The percentage of cutin plus lignin equalled "x" in the DMD equation. Mean cutin contents for winter forage diets (from Robbins et al. 1987b) was used in the equations. Cutin contents are usually small (< 5%) in winter forage, and the difference in DMD calculated using a mean versus no cutin content resulted in an average difference of 3.5%. DMD and DP were also evaluated using the above described statistical analyses, with the addition to the model of species as an independent variable. Significant differences among species were also determined.

RESULTS

SAMPLING DESIGN

A total of 50 cut-units were used for plant sampling in this study. The availability of 7- to 10-year-old unburned sites was inadequate to meet my sampling needs, so this age and treatment block was not represented in the data. Age comparisons between burned and unburned units were only tested for the 1- to 2- and 3- to 5-year age-classes. For within-treatments and among-species analyses, the 10-year age class was included in the data set.

The availability of units meeting my criteria within the Douglas-fir/Western hemlock plant association was limited. Data from sampling the available units in this plant association are discussed only where at least 3 replicates were obtained.

Not all of the forage species were found on every unit sampled, resulting in varying sample sizes. The sample size for each analyses should be taken into consideration for all interpretations.

FORAGE SPECIES

Vine maple was found in clumps on 60% of the units. It occurred on 100% of the unburned sites and 91% of the burned sites. Salal occurred in clumps on burned and unburned sites. Elderberry was rarely observed on any of the sampled sites. Only 3 burned sites contained this species: 7- to 10-year-old Douglas-fir and Western hemlock sites and 3- to 5-year-old Western hemlock sites. The plants were isolated, and there were usually only 2-3 plants within the entire unit. This species' low availability made it impossible to obtain a large enough sample size for comparisons of quality between treatments and among plant associations. Trailing blackberry appeared to have random distribution, and it occurred on burned and unburned sites. Red huckleberry was found in clumps on some plant associations and randomly distributed on others. It occurred on 73% of the burned sites and 88% of the unburned sites. Willows were found in isolated clumps within the cut-units. They occurred on 73% of the burned sites and 88% of the unburned sites.

EFFECT OF TREATMENT, PLANT ASSOCIATION, AND AGE ON QUALITY

Crude Protein, NDF, ADF, Lignin, and AIA

I did not detect a significant effect of treatment, plant association, or age on CP or NDF (Table 5). CP varied among species, with trailing blackberry having the highest percent content (9.65%) and the other species falling in the 5.21-7.24% range. Neutral detergent fiber was highly variable, with means ranging from 30.90% in trailing blackberry to 65.06% in vine maple (Table 5).

Acid detergent fiber content ranged from 44.18-49.49% for all species except trailing blackberry (17.78%) (Table 5). Burned sites produced trailing blackberry with significantly higher percent content of ADF than unburned sites ($p < 0.05$).

Lignin varied among species, with trailing blackberry having the lowest content (6.37%) and salal having the highest content (30.25%) (Table 6). Lignin in salal occurred in significantly higher percentages on 1- to 2-year-old burned and unburned sites (33.57%, 33.07%) than 3- to 5-year-old unburned sites (25.10%) ($p < 0.10$, $DF=4$, $F=2.07$).

TABLE 5: Average (SE) Crude Protein (CP), Neutral Detergent Fiber (NDF), and Acid Detergent Fiber (ADF) for selected winter forage on the Willamette National Forest, Oregon (winter 1989/90). Averages with the same small-case letter are significantly different.

SPECIES	TRT ¹	N	% Crude Protein	% Neutral Detergent Fiber	% Acid Detergent Fiber
RUUR	UB	19	9.80 (1.99)	30.82 (3.08)	17.00 (1.68)a
	BB	29	9.56 (0.99)	30.95 (6.31)	18.29 (2.77)a
VAPA	UB	15	6.88 (0.80)	61.68 (5.99)	45.91 (3.26)
	BB	16	6.41 (1.77)	58.82 (4.55)	43.92 (3.82)
SALIX	UB	6	6.77 (0.77)	57.89 (4.87)	45.93 (5.48)
	BB	14	7.45 (0.96)	55.48 (7.43)	43.42 (6.24)
ACER	UB	16	6.30 (0.65)	64.40 (3.32)	47.63 (2.40)
	BB	27	6.36 (1.63)	65.44 (3.26)	47.71 (2.34)
GASH	UB	16	6.29 (1.70)	56.73 (4.65)	48.34 (6.51)
	BB	27	5.86 (0.88)	56.88 (4.44)	50.17 (3.44)
SARA	UB	0			
	BB	3	5.21 (1.53)	57.07 (23.76)	45.58 (18.52)

1 -- Treatment: BB = burned UB = unburned

Lignin in trailing blackberry differed between burned and unburned sites, but these differences were inconsistent among plant associations.

TABLE 6: Average (SE) percent content of Lignin and Acid Insoluble Ash (AIA) for selected forage species on the Willamette National Forest, Oregon (winter 1989/90).

SPECIES	TRT ¹	N	LIGNIN		ACID INSOLUBLE ASH	
RUUR	UB	19	6.48	(3.43)	0.10	(0.08)
	BB	29	6.29	(3.82)	0.18	(0.33)
VAPA	UB	15	23.09	(5.09)	0.14	(0.30)
	BB	16	23.87	(8.29)	0.09	(0.06)
SALIX	UB	6	21.79	(3.64)	0.10	(0.09)
	BB	14	24.93	(7.87)	0.22	(0.43)
ACCI	UB	16	16.68	(6.87)	0.13	(0.15)
	BB	27	15.60	(4.76)	0.08	(0.05)
GASH	UB	16	29.33	(7.90)	0.33	(0.58)
	BB	27	30.80	(6.82)	1.10	(0.10)
SARA	UB	0				
	BB	3	22.71	(10.32)	0.07	(0.04)

1 -- Treatment: BB = burned UB = unburned

Acid insoluble ash content ranged from 0.07% in elderberry to 0.19% in salal (Table 6). Acid insoluble ash in vine maple differed among age classes on burned and unburned sites ($p < 0.10$, $DF=4$, $F=1.94$). Vine maple on 1- to

2-year-old unburned sites had significantly higher percentages of acid insoluble ash than any other site (0.176%).

Condensed Tannin

Condensed tannin contents were effected by treatment in 3 of the 5 species. Condensed tannin content ranged from 0 mg tannin/mg plant tissue in vine maple to 0.1806 in red elderberry. The greatest concentrations were observed in red elderberry, willow, and salal. Huckleberry and trailing blackberry had comparatively moderate amounts (Table 7).

Condensed tannin content in huckleberry was effected by treatment and age ($p < 0.02$, $DF=4$, $F=1.83$). Huckleberry had higher condensed tannin levels on unburned 3- to 5-year-old sites (0.0506) than unburned 1- to 2-year-old sites (0.0023) or burned 3-5 and 7-to 10-year-old sites (0.0030, 0.0116). Condensed tannin content in huckleberry was also effected by treatment and plant association ($p < 0.05$, $DF=7$, $F=.81$). Huckleberry had more condensed tannin content on burned Douglas-fir sites than on burned Western hemlock-rhododendron sites (Table 8). Huckleberry on unburned Douglas-fir/Western hemlock sites had more condensed tannin than on burned Western hemlock-rhododendron sites.

TABLE 7: Average (SE) Astringency, Condensed Tannin, and Hydrolyzable Tannin content of selected forage species on the Willamette National Forest (winter 1989/1990).

SPECIES	N	ASTRINGENCY ¹	CONDENSED TANNIN ²	HYDROLYZABLE TANNIN ³
RUUR	49	0.6737 (0.1071)	0.0290 (0.0255)	0.2349 (0.0286)
VAPA	32	0.0434 (0.0260)	0.0224 (0.0421)	0.0024 (0.0047)
SALIX	22	0.2204 (0.1469)	0.1525 (0.0991)	0.0062 (0.0068)
ACCI	44	0.0015 (0.0152)	0.0000 (0.0049)	0.0013 (0.0059)
GASH	43	0.1498 (0.0799)	0.0746 (0.0501)	0.0041 (0.0063)
SARA	3	0.0981 (0.0668)	0.1806 (0.1162)	0.0000 (0.0000)

1 - Astringency = mg of Bovine Serum Albumen precipitated per mg of plant tissue

2 - Condensed Tannin = mg of condensed tannin per mg of plant tissue

3 - Hydrolyzable Tannin = mg of hydrolyzable tannin per mg of plant tissue

Condensed tannin content in salal differed among age classes ($p < 0.06$, $DF=4$, $F=1.32$). Condensed tannin in salal on 3- to 5-year-old burned sites was higher (0.1056) than on 7- to 10-year-old burned sites (0.0607).

Trailing blackberry had higher condensed tannin content on burned 3- to 5-year-old sites (0.0453) than burned or unburned 1- to 2-year-old sites (0.0177, 0.0267) ($p < 0.03$, $DF=7$, $F=2.64$).

Condensed tannin content in trailing blackberry was effected by treatment ($p < 0.05$, $DF = 18$, $F = 3.43$), and the effects were inconsistent among plant associations and ages. The significant differences include:

- Burned 1- to 2-year-old Western hemlock sites produced trailing blackberry with higher condensed tannin content than similar unburned sites.
- Burned 3- to 5-year-old Douglas-fir sites produced trailing blackberry with higher condensed tannin content than burned 1- to 2-year-old Douglas-fir sites.
- Unburned 3- to 5-year-old Western hemlock sites produced trailing blackberry with higher condensed tannin content than similar unburned 1- to 2-year-old sites.

Condensed tannin in trailing blackberry was effected by treatment (Table 8), but this effect was inconsistent among plant associations ($p < 0.02$, $DF = 7$, $F = 1.59$):

- Burned Western hemlock sites produced trailing blackberry with higher condensed tannin content than unburned Western hemlock sites.
- Burned Douglas-fir sites produced trailing blackberry with higher condensed tannin content than on burned or unburned Western hemlock-rhododendron sites.
- Burned Western hemlock sites produced trailing blackberry with higher condensed tannin content than burned or

unburned Western hemlock-rhododendron sites.

- Unburned Western hemlock sites produced trailing blackberry with higher condensed tannin content than burned Western hemlock-rhododendron sites.
- Unburned Douglas-fir sites produced trailing blackberry with higher condensed tannin content than Western hemlock-rhododendron sites.
- Unburned Douglas-fir sites produced trailing blackberry with higher condensed tannin content than unburned Western hemlock sites.

Hydrolyzable Tannin

I did not detect a significant effect of treatment, plant association, or age on hydrolyzable tannin content. Hydrolyzable tannins (measured in mg of tannic acid per mg of plant tissue) were present in all species except red elderberry. Trailing blackberry contained comparatively high hydrolyzable tannin content (0.2353 mg/mg) (Table 7).

Astringency

Astringency ranged from 0.0015 mg protein precipitated per mg plant tissue in vine maple to 0.6737 in trailing

TABLE 8: Average (SE) condensed tannin content of selected forage species by plant association and treatment. on the Willamette National Forest, Oregon (winter 1989/1990). Averages with the same small-case letter are significantly different.

SPECIES	TRTMNT ¹	PLANT ASSC. ²	N	CONDENSED TANNIN ³		
RUUR	UB	PSME	6	0.0488	ijk	(0.0138)
	BB	PSME	9	0.0391	cd	(0.0218)
	UB	PSME/TSHE	4	0.0197		(0.0183)
	BB	PSME/TSHE	2	0.0220		(0.0220)
	UB	THSE-RHMA	4	0.0148	dfk	(0.0104)
	BB	TSHE-RHMA	9	0.0147	cegjm	(0.0120)
	UB	TSHE	6	0.0245	agi	(0.0219)
	BB	TSHE	9	0.0423	aef	(0.0128)
VAPA	UB	PSME	3	0.0293		(0.0316)
	BB	PSME	2	0.0586		(0.0293)
	UB	PSME/TSHE	4	0.0439	o	(0.0344)
	BB	PSME/TSHE	1	0		(0)
	UB	TSHE-RHMA	4	0.0367		(0.0553)
	BB	TSHE-RHMA	8	0.0104	o	(0.0255)
	UB	TSHE	4	0.0330		(0.0365)
	BB	TSHE	6	0.0268		(0.0260)
SALIX	UB	(POOLED DATA)	7	0.1807		(0.0904)
	BB	(POOLED DATA ⁴)	15	0.1621		(0.0903)
ACCI	UB	(POOLED DATA)	17	0		(0)
	BB	(POOLED DATA)	27	0 ⁵		(0.0028)
GASH	UB	(POOLED DATA)	16	0.0632		(0.0301)
	BB	(POOLED DATA)	27	0.0836		(0.0534)

1 - TRTMNT -- BB = Broadcast Burned UB = Unburned

2 - PLANT ASSC. = Plant Association Group (See Table 4)

3 - CONDENSED TANNIN = Measured in mg condensed tannin/
mg plant tissue

4 - POOLED DATA -- Data were pooled for species where no
significant differences were indicated
among plant associations.

5 - Negative condensed tannin concentrations were obtained
for this species.

blackberry. Salal and willow had intermediate astringencies compared with the other species. Elderberry, huckleberry, and vine maple had the lowest astringencies of those sampled (Table 7).

Astringency in red huckleberry and trailing blackberry differed among sites and between treatments (Table 9). Differences among plant associations (red huckleberry $p < 0.05$, $DF=7$, $F=1.79$; trailing blackberry $p < 0.06$, $DF=7$, $F=1.59$) include:

- Red huckleberry on burned Douglas-fir sites had higher astringencies than on unburned Douglas-fir sites.
- Red huckleberry on burned Douglas-fir sites had higher astringencies than on unburned Western hemlock sites or burned and unburned Western hemlock-rhododendron sites.
- Trailing blackberry on burned Douglas-fir sites had higher astringencies than on burned Western hemlock sites.
- Trailing blackberry on burned Douglas-fir sites had higher astringencies than on burned or unburned Western hemlock-rhododendron sites.
- Trailing blackberry on burned Douglas-fir sites had higher astringencies than on burned Western hemlock sites.

TABLE 9: Average (SE) astringencies of selected forage by treatment and plant association on the Willamette National Forest (winter 1989/90). Astringencies with the same letter are significantly different.

SPECIES	TRTMNT ¹	PLANASSC. ²	(N)	ASTRINGENCY ³	SE
RUUR	UB	PSME	6	0.6989	(0.1091)
	BB	PSME	9	0.7417 abc	(0.1044)
	UB	PSME/TSHE	4	0.7513 def	(0.0891)
	BB	PSME/TSHE	2	0.6410	(0.1262)
	UB	TSHE-RHMA	4	0.6071 cf	(0.1097)
	BB	TSHE-RHMA	9	0.6428 be	(0.0696)
	UB	TSHE	6	0.6698	(0.0711)
	BB	TSHE	9	0.6247 ad	(0.0982)
VAPA	UB	PSME	3	0.0262 g	(0.0178)
	BB	PSME	3	0.0857 gjkl	(0.0242)
	UB	PSME/TSHE	4	0.0571	(0.0257)
	BB	PSME/TSHE	1	0.0045	(0)
	UB	TSHE-RHMA	4	0.0333 l	(0.0156)
	BB	TSHE-RHMA	8	0.0378 k	(0.0280)
	UB	TSHE	4	0.0377 j	(0.0162)
	BB	TSHE	6	0.0547	(0.0151)
SALIX SP	UB	(POOLED)	7	0.2301	(0.1589)
	BB	(POOLED ⁴)	15	0.2159	(0.1407)
ACCI	UB	(POOLED)	17	0.0024	(0.0104)
	BB	(POOLED)	27	0.0009	(0.0175)
GASH	UB	(POOLED)	16	0.1172	(0.0412)
	BB	(POOLED)	27	0.1691	(0.0902)

1 - TREATMENT -- BB = Broadcast burned UB = Unburned

2 - PLANASSC. -- Plant Association Group (see Table 4)

3 - ASTRINGENCY = mg of Bovine Serum Albumen precipitated/
mg of plant tissue

4 - POOLED DATA = Data were pooled for species where
a significant difference was not
detected among plant associations

- Trailing blackberry on burned Douglas-fir sites had higher astringencies than on burned or unburned Western hemlock-rhododendron sites.
- Trailing blackberry on unburned Douglas-fir/Western hemlock sites had higher astringencies than on burned or unburned Western hemlock-rhododendron sites.
- Trailing blackberry on unburned Douglas-fir/Western hemlock sites had higher astringencies than on burned Western hemlock sites.

Dry Matter Digestibility

Dry matter digestibility (DMD) was highest in trailing blackberry (70.61%) and lowest in salal (38.85%). The other 4 species were in the 41-47% range (Fig. 2).

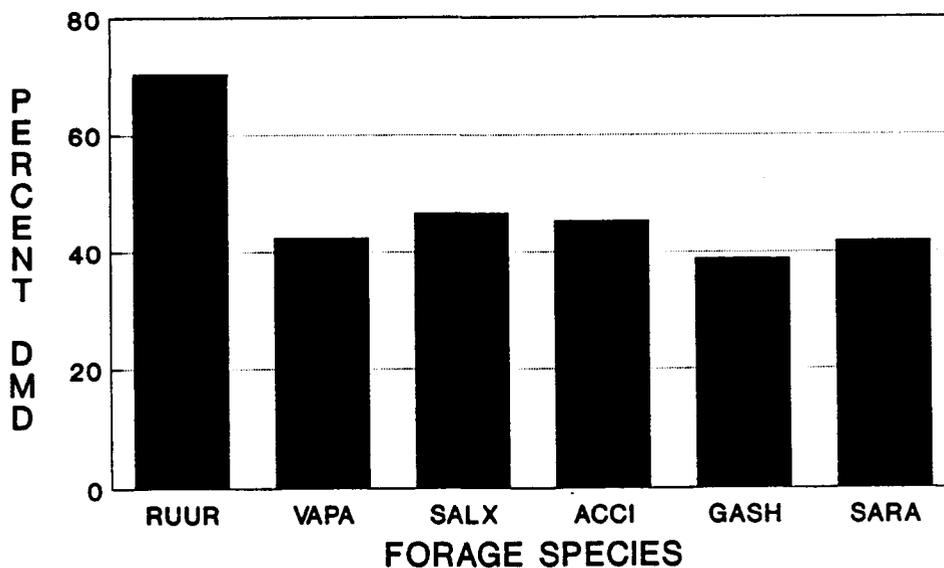


Figure 2: Dry matter digestibility of forage species on the Willamette National Forest, Oregon (winter 1989/90)

DMD differed among plant associations ($p < 0.06$, $DF = 3$, $F = 2.54$). Differences between burned and unburned sites were inconsistent among species and plant associations ($p < 0.03$, $DF = 27$, $F = 1.65$). Trailing blackberry had higher DMD on all plant associations than the other 5 species. Salal had significantly lower DMD than vine maple, trailing blackberry, and willow.

Differences in DMD occurring among plant associations include (Table 10):

- Willow on Douglas-fir sites had lower DMD than on Western hemlock-rhododendron sites.
- Red huckleberry on Western hemlock sites had lower DMD than those on Western hemlock-rhododendron sites.

DMD was correlated with lignin and neutral detergent fiber for all species except red elderberry (Table 11).

TABLE 10: Average (SE) Dry Matter Digestibility (%) of selected forage by plant association on the Willamette National Forest, Oregon (winter 1989/90). DMD's with the same letter are significantly different.

SPECIES	PLANT ASSC. ¹	N	DRY MATTER DIGESTIBILITY	
RUUR	(POOLED DATA ²)	49	70.61	(4.31)
VAPA	PSME	5	43.36	(4.39)
	PSME/TSHE	5	33.39	(7.76)
	TSHE-RHMA	12	47.29 a	(12.94)
	TSHE	10	40.19 a	(4.77)
SALIX SP	PSME	7	42.13 b	(5.09)
	TSHE-RHMA	7	48.61 b	(16.74)
	TSHE	7	49.47	(17.48)
ACCI	(POOLED DATA)	44	45.33	(8.42)
GASH	(POOLED DATA)	43	38.85	(6.03)
SARA	(POOLED DATA)	3	41.78	(22.34)

1 - PLANT ASSC. = Plant Association Group (See Table 4)

2 - POOLED DATA -- Data were pooled for species where a significant difference was not detected among plant associations

Table 11: Results of correlation analyses of Dry Matter Digestibility with lignin and astringency ($p < 0.05$) for winter forage species on the Willamette National Forest (winter 1989/90).

SPECIES	DRY MATTER DIGESTIBILITY
RUUR	L ($p=0.0001$, $r=0.70$) N ($p=0.0001$, $r=0.60$)
VAPA	L ($p=0.001$, $r=0.63$) N ($p=0.001$, $r=0.82$)
SALX	L ($p=0.001$, $r=0.79$) N ($p=0.001$, $r=0.94$)
ACCI	L ($p=0.001$, $r=0.69$) N ($p=0.0001$, $r=0.60$)
GASH	L ($p=0.001$, $r=0.66$) N ($p=0.001$, $r=0.71$)
SARA	A ($p=0.025$, $r=0.99$)

L = Correlation with Lignin

A = Correlation with Astringency

N = Correlation with Neutral Detergent Fiber

Digestible Protein

Digestible protein ranged from -2.84 g/100 g feed in trailing blackberry to 2.13 g/100 g feed in vine maple. Trailing blackberry, willow, salal, and elderberry had negative digestible protein content. Only vine maple and

huckleberry indicated positive digestible protein levels (Fig. 3). DP differed among species ($p < 0.0001$), $DF=5$, $F=113.32$), and DP was effected by plant association ($p < 0.10$, $DF=16$, $F=1.52$), and treatment ($p < 0.03$, $DF=5$, $F=2.57$).

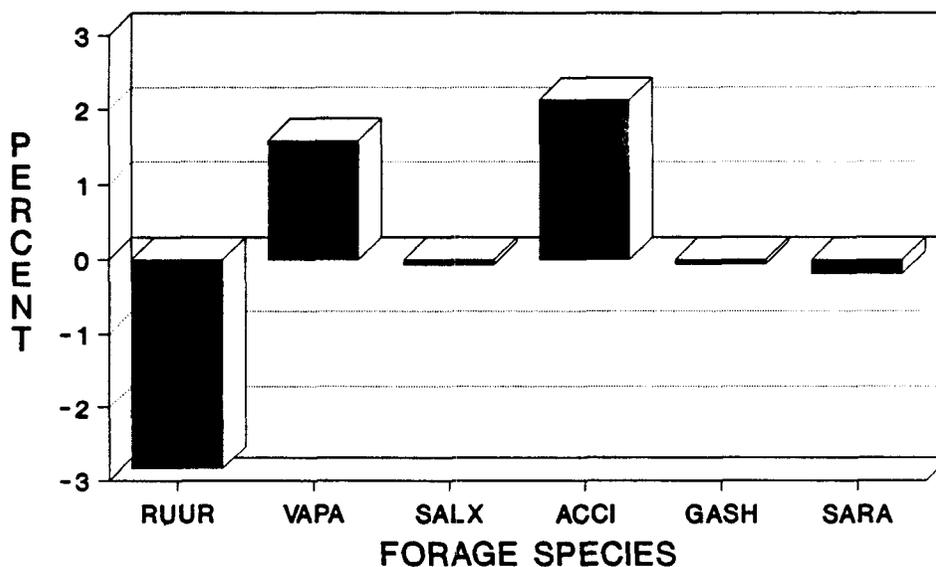


Figure 3: Digestible protein of forage species on the Willamette National Forest, Oregon. (Winter 1989/90)

Vine maple had significantly higher DP indices than salal, trailing blackberry, and willow. Huckleberry had significantly greater DP content than salal, willow, or trailing blackberry. Trailing blackberry had significantly lower DP content than all species, with the exception of elderberry.

Trailing blackberry had lower DP content on Douglas-fir/Western hemlock sites than on Western hemlock sites. Salal on Douglas-fir sites had lower DP content than on Western hemlock-rhododendron sites. Vine maple had higher DP content on Western hemlock-rhododendron sites than Western hemlock or Douglas-fir sites. Huckleberry on Douglas-fir sites had greater DP than on Western hemlock or Western hemlock-rhododendron sites (Table 12).

Salal and red huckleberry had lower DP levels on burned sites than unburned sites. Willow had higher DP content on burned sites (Fig. 4).

TABLE 12: Average (SE) Digestible Protein (%) of selected forage by plant association on the Willamette National Forest, Oregon (winter 1989/90). Digestible proteins with the same letter are significantly different.

SPECIES	PLANT ASSC. ¹	N	DIGESTIBLE PROTEIN	
RUUR	PSME	15	-3.29 b	(1.70)
	PSME/TSHE	6	-3.69 a	(1.77)
	TSHE-RHMA	13	-2.83	(1.71)
	TSHE	15	-2.07 ab	(1.98)
VAPA	PSME	5	3.12 cd	(1.95)
	PSME/TSHE	5	1.59	(0.82)
	TSHE-RHMA	12	1.17 d	(1.80)
	TSHE	10	1.30 c	(0.65)
SALIX	(POOLED DATA ²)	22	-0.07	(2.04)
ACCI	PSME	13	1.74 e	(0.67)
	PSME/TSHE	4	1.96	(0.81)
	THSE-RHMA	13	2.51 ef	(1.06)
	TSHE	14	1.65 f	(0.63)
GASH	PSME	12	-0.67 g	(1.44)
	PSME/TSHE	4	0.04	(0.41)
	TSHE-RHMA	13	0.58 g	(2.12)
	TSHE	14	-0.14	(0.93)
SARA	(POOLED DATA)	(3)	-0.19	(0.82)

1 - PLANT ASSC. = Plant Association Group (See Table 4)

2 - POOLED DATA -- Data were pooled for species where a significant difference was not detected among plant associations

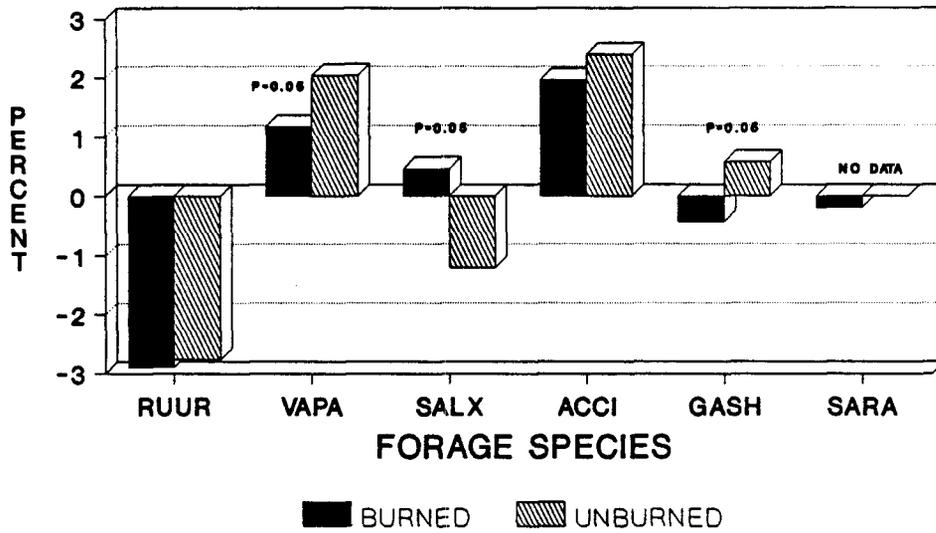


Figure 4: Effect of treatment on digestible protein of forage species on the Willamette National Forest.

DISCUSSION

FORAGE SPECIES

The plant community composition before harvest influences the vegetation development after harvest (Dyrness 1973, Franklin and Dyrness 1984), and an evaluation of the pre-harvest plant association must be balanced with objectives for forage production and availability over time. On sites where providing available winter forage is a management objective, broadcast burning after harvest may not produce the desired results because of changes in abundance of selected species.

None of the plant associations in this study were predicted to contain all of the selected forage species (Hemstrom et al. 1987), so variation in occurrence among plant associations was expected. Huckleberry, trailing blackberry, salal, and vine maple were found in all plant associations, though their occurrence varied among ages and treatments. These species are considered residuals, usually persisting in cut-units after disturbance (Franklin and Dyrness 1984, Dyrness 1973). I believe these species were in all of the sampled stands prior to harvest. On the Willamette National Forest plant associations are not always distinct. Transitional zones occur that have mixtures of

plant species not typically associated with each other (personal comm., Miles Hemstrom).

Elderberry and willow are not indicative of specific plant associations. Willows are considered invading species that seed into areas of high disturbance, such as along Class 4 stream headwall areas (Franklin and Dyrness 1984, Daubenmire 1968). I found willows in these isolated areas. Elderberry is also an invader, inhabiting very wet, disturbed sites. This species was only found on Western hemlock sites, the wettest of the plant associations sampled. The few isolated patches of elderberry I found indicate that it may be generally unavailable to elk as winter forage. Willow would also be available to elk only in isolated patches. Harper (1971) found winter diets of elk consisted of 0.5% willow and 0.3% elderberry. The other 4 selected winter forage species appear to be moderately to highly available across elk winter range on the Willamette National Forest.

I can not suggest with my data that burning effects abundance of these forage species. Other research on succession in the west central Cascades does indicate that burning can decrease the abundance of some species I sampled. Broadcast burning has been observed to diminish the shrub component of the plant community for the first 5 years after harvest, with recovery of the residuals occurring by year 10

(Franklin and Dyrness 1984, Dyrness 1965 and 1973, Isaac 1940, Steen 1966, Morris 1958). Unburned sites usually have higher percent cover of shrub species (Morris 1958, Steen 1966, Harper 1985, Dyrness 1973, Edgerton 1972), and shrub regrowth is usually slower on burned areas when compared to unburned areas (Bigley and Henderson 1990). I observed that vine maple, red huckleberry, and trailing blackberry seemed more abundant on unburned than burned sites. Morris (1958) and Steen (1966) also found red huckleberry more commonly on unburned than burned sites. Red huckleberry reproduces by rhizomes, which can decrease the adverse affect of burning on survival (Bigley and Henderson 1990). However, it also has a juvenile, vine-like stage for its first 4-5 years (Camp 1942) that may be effected by burning. Lower abundance of trailing blackberry on burned sites is in agreement with findings by Harper (1985). Even low-intensity fires will consume the above-ground stems. Harper (1985), Steen (1966) and Dyrness (1965) found vine maple, huckleberry, and salal cover diminished on burned sites. Dyrness (1965) found vine maple to be very sensitive to disturbance. Abundance of some residual species used as winter forage may be diminished with broadcast burning.

FORAGE QUALITY

Crude Protein

CP content of species I sampled ranged from 5-10%. Trailing blackberry contained the highest percentage (10%), with the rest falling in the 6-7% range.

A plant's phenology greatly influences its quality as forage. Crude protein levels were expected to be low in the sampled plants because of winter dormancy (Nelson and Leege 1982); my data reflected this. Plant protein is found primarily within the cytoplasm with small amounts associated with the cell wall (Lyttleton 1973, Preston 1974, Albersheim 1975). Plant protein content increases during early growth or regeneration and decreases with vegetative maturity as plants increase emphasis on producing nitrogen-free structural components (Greenwood and Barnes 1978).

Differences in CP among age classes would be expected, but age class of the unit was not necessarily indicative of the age of a plant. Composite samples in this study usually included 20 individual plants, ranging in age from seedling to mature.

Harper (1985) found similar CP levels in trailing blackberry in southern Oregon. CP levels in my vine maple and red huckleberry samples (both 6.4%) were higher than Happe et al. (1990) who found 2.7% crude protein in vine maple stems and 4.5% CP in huckleberry. Leslie et al. (1984) found 5.8% CP in red huckleberry. I also found higher CP in willow (7.43%) compared to Leslie et al. (1984) (5.2%). Huckleberry and willow samples in my study were often composed of stems and leaves that had not undergone abscission. Higher CP in my samples would be expected because of the additional leaf-material, which has higher CP content than stems. I was interested in sampling what was available to the animals in winter, so composite samples were not separated by plant part.

Diets with CP ranging from 8-14% were fed to cow elk in a study by Thorne et al. (1976), and animals were able to maintain or slightly increase their body weights. Nelson and Leege (1982) stated that elk required a minimum of 5.5-6% CP for maintenance. A majority of the selected forage species investigated in this study would fall in the maintenance zone. Crude protein measurements do not take into consideration potential adverse effects of secondary compounds, which may reduce the availability of protein to the animal. Maintenance levels of CP may be further diminished by factors that reduce protein assimilation.

I did not detect differences in CP in plants sampled on different sites regardless of treatment. Inconsistent effects of burning on CP have been observed (Cannon et al. 1987, Swank 1956, Einarsen 1946, Gilliam 1988, DeByle et al. 1989, Arnett 1990). The hypothesis that burning promotes a flush of available nitrogen on some sites, which could potentially increase the nitrogen concentration within the species occupying the site, was not confirmed in my study. The soils of the Willamette National Forest have been determined to be nitrogen limited (Gessel et al. 1972, Miller et al. 1976). McNabb et al. (1986) found that only 1-2% of the nitrogen pool on a site is actually available. Nitrogen becomes available to a plant from cycling through soil organisms, oxidation by fire, precipitation, or nitrogen fertilizer. Organic matter in the soil contains the majority of the nitrogen (Kraemer and Hermann 1979), and the loss of this organic matter is seen as the most significant consequence of fire. Studies have documented a decrease or no observable change in available nitrogen following burning, (Dyrness et al. 1989; Gilliam 1988), but consistent measures of increased availability of nitrogen following burning have not been obtained (Van Cleve and Dyrness 1983). McNabb and Cromack (1990) felt that burning is most likely to increase nitrogen availability in colder forest types with slow decomposition rates. The Pacific Northwest does not fall within that category.

CP reflects the concentration of nitrogen within the plant. Plants may have higher total nitrogen content on sites with higher nitrogen availability, but they often redistribute the available nitrogen into new tissue formation (Sollins et al. 1980). Because of the plant's growth, concentration in the tissues may not change. I found no evidence in my data to indicate that burning will increase the nitrogen concentration in the selected winter forage species thereby increasing the crude protein concentration.

The availability of early-successional, high quality plant species that invade the site after burning may be increased (Dyrness 1965 and 1973, Morris 1958, Steen 1966, Schoonmaker and McKee 1988), but that was outside the scope of this study. The primary invaders on a site following broadcast burning are annuals that may not be important in winter forage diets of elk (Leslie et al. 1984, Harper 1985). The quick establishment of these invading populations may out-compete the residual browse species for available nutrients, resulting in limited response by the residuals to a short flush of nitrogen (Harper 1985, Daubenmire 1968). Burning may increase the availability of winter forage species by increasing the growth of some residuals, but fire generally decreased the abundance of most of the selected forage species. Increased growth in residual individuals may not make up for the decrease in abundance.

Lignin

I observed greater percentages of lignin in salal on 1- to 2-year-old burned and unburned sites. Lignin is an aromatic monosaccharide polymer. Harkin (1973) felt its primary use by the plant is to add rigidity to the cell wall, though lignin may also be used by plants as a defense mechanism (Stafford 1988). It is produced from the same biosynthetic pathway as tannins (Jung and Fahey 1983). Lignin concentrations increase with advancing maturity (Allison and Osbourn 1970, Lindgren et al. 1980, Morrison 1980). It is fairly indigestible because of its resistance to enzymatic and acid hydrolysis and gastrointestinal fermentation. Lignin production is related to the carbon fixation rate on a site. Low light levels produce plants with low lignification. Salal may have higher lignin content on recently disturbed sites because of the reduction in shrubs and subsequent increase in light intensity. Increased lignification in salal may be a reflection of increased temperatures, a relationship observed in some temperate grasses (Ford et al. 1979). Salal may also be producing higher concentrations of lignin during recovery on disturbed units to ensure re-establishment. Bigley and Henderson (1990) indicated that salal was slow to recover from broadcast burning. Mechanical wounding generally leads to an increase in phenolic compounds such as lignin. The disturbance of

logging activity itself on the sites may have caused this residual plant to respond with increased lignification (Rittenger et al. 1987).

Condensed Tannin and Astringency

I found that increased condensed tannin and astringencies may be expected in species sensitive to increases in stress caused by burning on resource-limiting sites. Tannin production is an important defense mechanism that plants have evolved to provide protection from herbivores and disease (Rhoades and Cates 1978, Swain 1979). Their presence or absence has been attributed to a variety of factors including light intensity (Larsson et al. 1986); available nitrogen content in the soil (Barry and Foss 1983) available nutrients (Tiarks et al. 1988, Waterman and Mole 1989, Barry and Manley 1986, Larsson et al. 1986, Owen-Smith and Cooper 1987); available water (Fales 1984); mechanical injury (Chalker-Scott and Krahmer 1988); and evolutionary strategies of the plant involving growth rates and consumption by herbivores (Coley et al. 1985, Rhoades and Cates 1978, Feeney 1975). Considering the wide variety of factors influencing tannin content, variability was expected within and between the species I sampled on varying sites.

The effect of broadcast burning on forage tannin content has not been investigated. Numerous studies have investigated the effect of fire on plant nutrition, but measurements of quality have been primarily limited to CP and fiber contents.

The plant associations I sampled covered a range from very dry (Douglas-fir) to intermediate (Western hemlock-rhododendron) to wet (Western hemlock) sites. The dry sites are also nitrogen limited, often causing seedlings to appear chlorotic (Hemstrom et al. 1987). The differences in resource availability alone on these sites could have led to variation in tannin content. The disturbance caused by logging and additional stress from broadcast burning could effect tannin production.

Astringency and condensed tannin content produced the most variability within my data among plant associations and between treatments. Astringency was greatest for trailing blackberry, followed by willow, salal, elderberry, huckleberry, and vine maple. Happe et al. (1990) found similar astringency for vine maple (0) and red huckleberry (0.06). Species with slow growth generally invest in the relatively costly production of secondary compounds to reduce the effects of herbivory, while fast-growers tolerate higher rates of damage from herbivores and produce lower

concentrations of anti-herbivory compounds (Coley et al. 1985). My data did not consistently reflect this observation.

Coley et al. (1985) has hypothesized that production of anti-herbivory defenses within a plant are directly related to the potential growth rate of the plant. If potential growth rate decreases because of resource limitations, and herbivory does not decrease in a similar manner, replacement of resources lost to the herbivores will become costly. The relatively high production costs associated with defensive chemicals would become justified. I observed a relationship between resource limitation with astringency and condensed tannin content in some species.

Red huckleberry appeared to be sensitive to the stress of burning on Douglas-fir sites. Huckleberry from burned Douglas-fir sites, which are very dry and nitrogen poor, had higher astringencies than on similar unburned units. These sites also produced red huckleberry with greater astringencies than unburned sites in the Western hemlock association, which is not moisture- or nutrient-limiting, and the Western hemlock-rhododendron association, which is moderately resource-limiting. Red huckleberry is a species that is slow to mature even on resource-rich sites, so the benefit of reduced herbivory on sites that do not promote

rapid growth may compensate for expenditures needed to produce the tannins. Barry and Manley (1986) observed that increases in soil nutrient and climatic stress in Lotus species caused increases in production of condensed tannin. Condensed tannin was highly correlated with astringency for red huckleberry.

Trailing blackberry showed the same type of sensitivity on Douglas-fir sites. Astringencies in trailing blackberry were greater on the burned Douglas-fir sites than on burned Western hemlock or burned/unburned Western hemlock-rhododendron sites. A combination of moisture and nutrient-stress with increased light intensities seems to influence tannin production. The increased light intensity on a burned site known to be moisture- and nutrient-limited, such as the Douglas-fir sites, could account for the higher astringencies seen in trailing blackberry. This relationship was detected on the 2 species with the largest sample size, indicating that to detect differences in astringency concentrations a large sample size should be obtained.

Because of the apparent sensitivity of these 2 species to stress, burning of dry sites should be carefully scrutinized because of the subsequent increases in astringency.

Condensed Tannins

My data indicate that higher condensed tannin content could be expected in plants produced on the resource-limiting plant associations (Douglas-fir and Douglas-fir/Western hemlock), and that burning of these sites could increase already elevated tannin concentrations for some species. Burning of resource-rich sites (Western hemlock) can significantly increase tannin content to concentrations above those found on the poorer sites. I found condensed tannin contents increased with age in some species, reaching a peak at 3-5 years.

Condensed tannins are the most widely distributed tannin in vascular plants (Bernays et al. 1989). Their effect on nutrition is related to their ability to form strong bonds with proteins (Blytt et al. 1988). Cooper and Owen-Smith (1985) have suggested that condensed tannins inhibit microbial breakdown of plant cell walls. Condensed tannin presence generally results in diminished weight gains and poor feed efficiency (Blair and Mitaru 1983). Blytt et al. (1988) showed that condensed tannins in a purified form inhibited digestive enzymes. The presence or absence of condensed tannins is effected by the same factors described above for astringency, and I observed some of the same relationships between condensed tannin concentration and

resource-limitation. Units 3- to 5-years-old had higher condensed tannin contents in salal, huckleberry, and trailing blackberry than 1- to 2-year-old unburned units and 7- to 10-year-old burned units. However, condensed tannin in trailing blackberry did not decline on the 7- to 10-year-old sites. It appears that condensed tannins in salal and red huckleberry are reaching a peak concentration at 3-5 years. Condensed tannin in trailing blackberry continued to increase into year 10. As physiological maturity of a plant advances, tannin contents have been observed to increase (Theander et al. 1981). These species may be accumulating condensed tannins during the peak years of re-establishment after disturbance. Higher tannin concentrations produced during re-establishment could be used as a defense against herbivory as the residual species compete for nutrients with the invader plant populations.

Burning of dry sites can decrease infiltration rates, increasing the potential for moisture-stress for the plant community, and increasing the tannin content in species sensitive to this stress. Burning appeared to effect the condensed tannin concentration in trailing blackberry. The very dry, burned plant associations produced trailing blackberry with higher condensed tannin content than burned/unburned, moderately-wet Western hemlock-rhododendron sites; a pattern similar to that seen with astringencies. The

increased light intensities, increased climatic stress, and low nutrient levels probably accounted for the increased tannin production in trailing blackberry on burned sites. The effect of site on condensed tannin content was indicated on unburned, resource-limiting sites, which produced trailing blackberry with greater condensed tannin content than any other unburned site. Even without the increased stress caused by burning, the resource-limiting sites produced trailing blackberry with elevated condensed tannin content. This indicates the sensitivity of this species to site-specific conditions.

Burning of wet sites (Western hemlock) appeared to increase the tannin content in trailing blackberry above concentrations found in similar unburned sites and above the content found in intermediate burned and unburned sites. I do not know why trailing blackberry would produce comparatively high tannin contents on burned wet sites, but the significant effect should be a management consideration.

Condensed tannin in red huckleberry did not differ between burned and unburned sites within the same plant association, but the effect of resource-limitation on tannin content was evident. Burned Western hemlock-rhododendron sites (intermediate nutrient and moisture regime) produced huckleberry with significantly less tannin content than

burned resource-limiting sites (Douglas-fir) or unburned Douglas-fir/Western hemlock sites. Astringency content of huckleberry was affected by burning.

Hydrolyzable Tannins

Hydrolyzable tannin concentrations were very low or absent in all species sampled with the exceptions of trailing blackberry and willow. Trailing blackberry and willow had high concentrations of hydrolyzable tannins on all sites. Hydrolyzable tannins are restricted to the dicotyledons, and very little is known about the physiological responses of animals to ingestion. Adverse nutritional effects are not specifically known, but hydrolyzable tannins are capable of precipitating proteins, and they may cause physiological effects in mammals through absorption directly into the metabolic pathway. The response to this absorption is not known, but it may present a deterrent to feeding at some level (Robbins et al. 1987a, McCleod 1974, Mould and Robbins 1982). Hydrolyzable tannin in trailing blackberry was highly correlated with astringency. Astringency was also positively correlated with condensed tannin in this species. Additional investigation of the effects of this tannin are necessary, especially as they occur in big game forage species. Elk have been observed to eat trailing blackberry throughout the

year at moderate levels. Harper (1971, 1985) observed that trailing blackberry constituted 17.5% of an elk winter diet. The success of hydrolyzable tannin as a deterrent to herbivory depends on the elk associating an astringent taste with metabolic aberrations when ingested (Rhoades 1979). At what feeding level this association occurs is not known.

Total Nutrient Availability for Elk

Vine maple and red huckleberry were the only species with positive digestible protein levels, indicating that of the species sampled, they would provide the greatest available protein to the elk. I found that digestible protein content may be expected to be higher in winter browse on unburned sites that are not resource-limited than on burned, resource-limited sites.

Food selection by free-ranging animals is based on perceptions of cost-benefit constraints within the nutritional environment (Arnold and Hill 1972). Digestion and metabolic processes that enable absorption, distribution, and utilization of specific chemicals within ingested foods provide feedback to animals so that they can constantly evaluate their nutrient environment (Robbins 1983).

A multitude of factors effect the digestibility of forage, and the influence of the factors differs among animal species and individuals. Measurements that use multiple factors to determine overall digestibility and available protein of forage were developed by Robbins et al. (1987ab). Available protein is a function of plant CP content, non-digestible fiber-bound protein, and the protein precipitating capacity of the plant tissue (Robbins et al. 1987b). The indices of digestible protein (DP) and dry matter digestibility (DMD) indicate the total available protein and digestibility of forage while accounting for potential adverse effects tannins may have on the nutrient availability and the animals' ability to counteract the effects.

The digestible protein equation was developed by feeding black-tailed and white-tailed deer forages with a range of tannin contents. CP content and in vitro digestibilities of the forages were determined, and their difference (digestible protein reduction) was plotted against astringency to develop the DP index. Physiological processes vary between species as well as individuals, and this must be considered in the application of this equation to other ruminants. The DP equation may provide a more realistic measurement of nutrient availability to the animals than CP, fiber, and lignin content alone.

DP is reduced proportionally to the astringency of the plant material. Forage species with high astringencies (trailing blackberry, salal, and willow) will provide reduced available protein. Shrub leaves had astringencies high enough to reduce DP in Robbins' study (1987a). Salal and trailing blackberry samples I tested consisted primarily of leaves, and huckleberry and willow plants often still held leaves that had not undergone abscission in the late fall. The presence of leaf material in these samples may reflect in their high astringency compared to samples consisting of twigs. I found salal, trailing blackberry and willow to have the highest astringencies of those sampled and subsequently, the lowest DP indices.

Robbins et al. (1987b) found low astringencies in twigs and stems of winter browse. In my study vine maple twigs had the lowest astringency of any sampled species, and therefore high DP. High DP indices were found for red huckleberry, which also had low astringency.

Burning sites appeared to increase the DP levels for willow, while huckleberry and salal had higher DP levels on unburned sites. The increased DP on the unburned sites for red huckleberry followed the relationship observed of lower astringencies on unburned sites for this species. Nutrient- and moisture-limiting sites had lower DP content in trailing

blackberry, salal, vine maple, and red huckleberry than non-limiting sites because of increased astringencies on these sites. This indicates that forage quality may be limiting on these sites for elk.

DMD is calculated using fiber, solubles, and astringency information. Lignin and NDF are negatively correlated with DMD. Astringency does not heavily influence the forage DMD, adversely effecting only the neutral detergent soluble component. Trailing blackberry had the highest DMD levels because of its relatively low lignin and fiber contents, even with its high astringency. Salal had the lowest DMD of all species because of its high lignin content, low neutral detergent soluble component, and moderate astringency.

DMD did not differ by treatment or site for any forage species in my study. I did not observe effects of treatment or plant association on any of the variables used for calculating DMD (with the exception of astringency). I believe that DP is a sensitive index for comparing effects of site conditions on protein availability. Robbins et al. (1987b) stated that summative equations such as DMD would model nutritional availability in forage with high astringencies, but I did not find a correlation between astringency and DMD using my data.

The dry matter digestibility contents I calculated are higher than in-vitro dry matter digestibility (IVDMD) values found by Happe et al. (1990) and Leslie et al. (1984) for the same species. IVDMD is a artificial digestion procedure conducted in a laboratory using digestion flasks and rumen liquor, usually from cattle. Digestibility values obtained from IVDMD do not involve plant astringency and the animal's ability to counteract adverse effects of tannins bonding to proteins and starch. Higher digestibility values would be obtained through calculations such as DMD that factor compensatory abilities for astringency effects.

Forage Summary and Management Recommendations

I did not detect a positive influence of broadcast burning on the quality of selected winter browse. I would suggest that a model for evaluating elk habitat effectiveness should include a differentiation between summer and winter forage area treatments with considerations for site-specific plant associations. A forage area in winter range that is expected to produce high quality, highly available browse through the winter may not occur on burned units or on resource-limited sites. Burning does generally increase the availability of high quality herbaceous species, and the objectives of the area based on season of use must be

evaluated before burning is prescribed. The removal of slash to facilitate big-game movement through units is an issue that also must be considered. Fire intensities can be varied, and plant response to light-burns applied to remove small-diameter slash needs to be investigated.

Because of the low astringencies and abundance of red huckleberry and vine maple, these species would provide the highest available, quality forage of the species that I sampled. The quality of red huckleberry is decreased on burned and resource-limited sites, however, and that should be taken into consideration. Willow had high digestibility, but because of its high astringency and low abundance on the Forest it is not available in quantity or quality for elk. Trailing blackberry has high digestibility, but because of its high astringency it may not be high quality forage for elk. It appeared to be sensitive to the effect of resource-limitation and burning. Deer are known to eat considerable amounts of trailing blackberry, but they also produce proline-rich saliva that can bind with tannins before they reach the digestive tract (Robbins et al. 1987b). Parotid salivary glands in ruminants are 3 times larger in browsers than grazers (Hofman 1973, Kay et al. 1980), and browse has been observed to constitute as high as 70% elk diets (Harper 1971). The ability of elk to produce proline-rich saliva to bind with tannins is not known. Robbins' equations factor

an ability to counteract adverse effects of tannins based on studies with black-tailed deer, but how well this actually applies to elk is unknown. If elk do not have this ability, the quality of these forage species would be lower than indicated.

Salal had the lowest DMD and negative DP. It is abundant on the Forest, but it appears to be sensitive to burning, with lower DP on burned sites. The elderberry sample was too small to draw conclusions from, but its rarity indicates that it is not widely available to elk.

The influence of tannin content on forage quality must be evaluated for other elk forage species. It was apparent in the few species I sampled that burning and resource-limitation on sites can significantly effect the tannin content in some forage species. An increased understanding of specific phenolic chemical effects on animals and species-specific adaptations to ingestion of phenolics is needed to comprehend the nutritional needs of elk. Further investigations on the digestibility of other forage species on the westside Cascades is necessary. Information on the ability of elk to negate the adverse effects of condensed tannin on nutrient assimilation is also needed.

The extent that elk on the Willamette National Forest

consume shrub species in winter needs to be validated. The very low quality of the selected forage species in this study indicate that elk may be obtaining other types of forage to sustain themselves through the winter.

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APPENDICES

APPENDIX A

Preferred winter browse species for Roosevelt elk, Westside Oregon Cascades, as predicted by a cadre of wildlife biologists (1989), and available literature. (* indicates species chosen for sampling)

<u>PLANT SPECIES</u>	<u>PREDICTED BY</u>
* Red Huckleberry (<u>Vaccinium parvifolium</u>)	Smith, Gangle, Morris, Steele, Everett. Wolfer, Heintz, Zahn, Leslie
* Trailing blackberry (<u>Rubus ursinus</u>)	Smith, Morris, Everett, Hall, Wolfer, Zahn, Heintz, Leslie,
* Vine Maple (<u>Acer circinatum</u>)	Hall, Gangle, Everett, Leslie, Zahn, Heintz, Nelson & Leege, Steele
* Willow (<u>Salix</u> sp.)	Morris, Everett, Gangle, Zahn, Hastick, Heintz, Steele, Nelson & Leege
Grasses	Smith, Wolfer, Everett, Heintz, Hastick, Nelson & Leege, Leslie
* Salal (<u>Gaultheria shallon</u>)	Sharrow, Wolfer, Everett, Zahn, Heintz, Leslie, Nelson & Leege
* Red Elderberry (<u>Sambucus racemosa</u>)	Gangle, Heintz, Morris, Steele, Everett
Red Cedar (<u>Thuja plicata</u>)	Sharrow, Wolfer, Heintz, Leslie
Deerbrush Ceanothus (<u>Ceanothus integerrimus</u>)	Hall, Steele, Wolfer, Hastick
Swordfern (<u>Polystichum munitum</u>)	Sharow, Everett, Leslie, Nelson & Leege
Sedge (<u>Carex</u> sp.)	Zahn, Leslie, Smith, Nelson & Leege
Snowbrush (<u>Ceanothus velutinus</u>)	Wolfer, Hastick, Hall
Madrone (<u>Arbutus menziesii</u>)	Hall, Gangle

APPENDIX A CONTINUED

<u>SPECIES</u>	<u>PREDICTED BY</u>
Pacific yew (<u>Taxus brevifolia</u>)	Gangle, Wolfer
Oregon grape (<u>Berberis nervosa</u>)	Sharrow, Everett
Salmonberry (<u>Rubus spectabilis</u>)	Zahn, Morris
Deerfern (<u>Blechnum spicant</u>)	Leslie, Nelson & Leege
Red alder (<u>Alnus rubra</u>)	Zahn, Nelson & Leege
Western Hemlock (<u>Tsuga heterophylla</u>)	Leslie, Nelson & Leege
White Hairy Hawkweed (<u>Hieracium albiflorum</u>)	Smith
Indian lettuce (<u>Montia linearis</u>)	Smith
Woodrush Sedge (<u>Carex luzulina</u>)	Smith
Wedgeleaf Ceanothus (<u>Ceanothus cuneatus</u>)	Hall
Evergreen Huckleberry (<u>Vaccinium ovatum</u>)	Hall
Lilac (<u>Ceanothus</u> sp.)	Sharrow
Baccharis (<u>Baccharis</u> sp.)	Sharrow
<u>Ceanothus</u> sp.	Heintz
Forbs (general)	Heintz
Rose (<u>Rosa</u> sp.)	Heintz

APPENDIX A CONTINUED

<u>SPECIES</u>	<u>PREDICTED BY</u>
Miner's lettuce (<u>Montia siberica</u>)	Morris
Thimbleberry (<u>Rubus parviflorus</u>)	Morris
Ginger (<u>Asarum caudatum</u>)	Morris
Whipple vine (<u>Whipplea modesta</u>)	Everett
Silk tassel (<u>Garrya fremontii</u>)	Steele

SURVEY SOURCES

Everett, Evelyn: U.S. Forest Service Botanist, Willamette NF,
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Zahn, Max: Washington Department of Wildlife, Olympia,
Washington

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Leslie, D.M., Jr, E.E. Starkey, and M. Vavra. 1984. Elk and deer diets in old-growth forests in western Washington. *J. Wildl. Manage.* 48:762-775.

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APPENDIX B

LABORATORY PROCEDURE FOR DETERMINATION OF ASTRINGENCY,
CONDENSED TANNIN, AND HYDROLYZABLE TANNIN CONTENTProtein Precipitating Capacity of Plant Samples

Blue-dyed bovine serum albumen (BBSA) solution was prepared as described by Asquith and Butler (1985). Protein concentration of the BBSA was determined by the Lowry method (Peterson 1983). The Lowry assay indicated that the protein concentrations of the BBSA solutions met desired specifications.

Plant samples weighing 0.2700 g (+/- 0.005) were mixed with small quantities of 50% v/v methanol. The solution was boiled at 90 degrees celcius for 8 minutes then centrifuged at 3000 g for 15 minutes. The solution was brought to a final concentration of 10 mg plant tissue/ml solution.

Species were sorted by replicates representing each unique site (i.e. a combination of the independent variables: treatment, plant association, and age-class). Each sample was tested with BBSA concentrations ranging from 1.0 to 12.0 mg/ml to determine equivalence point (Hagerman and Robbins 1987). This protein level was used to determine the protein precipitating capacity for the remaining replicates. Two duplicates were tested within each group of samples extracted

to check the consistency of the procedure.

Protein precipitation capacity was determined by adding: 0.5 ml of extract, 0.5 ml methanol, and 4.0 ml of BBSA. Samples were then vortexed. Blanks of 1.0 ml 50% methanol and 4.0 ml of each BBSA concentration were prepared. After an incubation of 17-24 hours at 4 degrees celsius, the samples were centrifuged for 15 minutes at 3000 g. The supernatant was removed and the remaining fluid aspirated. Pellets were resuspended with 3.5 ml of BBSA solution B (1% sodium dodecyl sulfate, 5% trithanolamine, and 20% isopropanol). The tubes were vortexed, and the absorbance at 590 nm was determined in quantitative mode. Equivalence points were identified by the peak absorbance.

Standards for the BBSA solution were prepared in duplicate with 0.5 ml of each protein level (1.0 mg/ml to 12.0 mg/ml) and 3.0 ml of BBSA solution B. Standards and the samples were read against a blank of 0.5 ml methanol and 3.0 ml BBSA solution B.

TANNIN INVOLVEMENT IN PROTEIN PRECIPITATION

For each sample, the amount of tannin involved in protein extraction was determined using the protein

precipitating phenolic assay (Hagerman and Butler 1987). Protein precipitating phenolics (PPP) were determined for the same sample groups as the BBSA. A range of 1.0-8.0 mg/ml protein was used to identify the equivalence points. A 0.5 ml sample of tannin extract was placed in a test tube: 2.0 ml of PPP solution (Bovine serum and an acetate buffer) was added, vortexed, and the solution was incubated at 4 degrees celsius along with the BBSA test samples for 17-24 hours. Following incubation, samples were centrifuged at 3000 g for 15 minutes. The supernatant was poured off, and the remaining fluid was aspirated. Pellets were resuspended with 4.0 ml of 1% sodium dodecyl sulfate and 5% triethanolamine (PPP solution B), vortexed, and 1.0 ml of 0.01 M FeCl/0.01 M HCl (PPP solution C) was added. The extracts were incubated at room temperature for 15-30 minutes. Samples were read against a blank solutions of 4.0 ml PPP-B and 1.0 ml PPP-C in photometric mode at 510 nm.

CONDENSED TANNIN CONTENT

Condensed tannin reagents included:

- vanellin/ 4% in methanol: made from stock of 1% vanellin w/v (10 g vanellin/ 1.0 L methanol) and 8% HCl v/v (60.0 mls HCl/1.0 L 50% methanol)

- 4% HCl in methanol: made from stock (equal volumes of 8% HCl and 50% methanol)
- catechin standard (0.4 g catechin/100 ml 50% methanol - final catechin concentration 4 mg/ml)

Reagents were warmed in a water bath to 30 degrees celcius. A 0.25 ml portion of the extract and 2.5 ml vanellin/HCl were placed in a test tube and incubated 20 minutes in the water bath. Sample blanks were prepared with 0.25 ml of each extract and 2.5 ml 4% HCl in methanol and set aside for 20 minutes.

Catechin standards were prepared in duplicate by adding 0.0, 0.1, 0.2, 0.3, 0.4, and 0.5 ml catechin, brought to a volume of 0.5 ml with 50% methanol, and vanellin/4% HCl (5.0 ml) was added to these solutions. Standards were incubated along with samples. The extracts and blanks were read against a reagent blank of 0.5 ml methanol in 5.0 ml vanellin/4% HCl. Absorbance was read at 500 nm in quantitative mode.

HYDROLYZABLE TANNIN CONTENT

To determine hydrolyzable tannin content 0.25 ml of the extract and 0.75 ml 50% methanol were placed in plastic test

tubes. The tubes were capped and placed in a 40 degree celsius water bath for 30 minutes. Sample blanks were prepared with 0.25 ml of each extract and 1.75 ml 50% methanol placed in micro-cuvettes and set aside for 30 minutes. A solution of seven percent w/v (7.0 g/ 100 ml water) potassium iodate was warmed in the water bath.

Standards were prepared in duplicate with 0.0, 0.25, 0.5, 0.75, and 1.0 ml tannic acid (tannic acid concentration = 1.0 mg /ml, 0.5 g tannic acid/500 ml 50% methanol) brought to 1.0 ml total volume with 50% methanol. The standards were incubated along with the samples. Potassium iodate (1.0 ml) was added to each sample, and the absorbance was determined after 15 seconds at 550 nm in quantitative mode. Standards were read against a reagent blank of 1.0 ml 50% methanol and 1.0 ml potassium iodate.

Samples were read against micro-cuvette blanks. Samples with 0.0 or negative concentrations following BBSA determination were not included in the condensed or hydrolyzable tannin content testing.

CALCULATIONS

A median absorbance for each standard was determined

after concentrations for all samples had been obtained. The median was used to recalculate each concentration to standardize the data for comparisons.