Effect of Lichen and In Vitro Methodology on Digestibility of Winter Deer Diets in Maine

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In vitro fermentations using domestic cow and deer inocula were conducted to examine digestibilities of lichens and coniferous forages that are consumed by White-Tailed Deer (*Odocoileus virginianus*) in winter in Maine. Expected in vivo (converted from in vitro) dry matter digestibilities (DMD) of diets calculated from single species digestions were compared to 1) in vivo DMD of mixed diets and 2) apparent digestibilities that were obtained from previous research on captive White-Tailed Deer. Fermentations using cow inoculum were significantly lower than those using deer inoculum. No lichen-induced synergisms were found using either inocula. Apparent digestibilities from earlier studies were significantly higher than all in vitro or in vivo estimates of digestibility. Results suggest that analytical techniques and inocula source can underestimate digestibilities of some winter forages.

Key Words: digestibility, lichens, Maine, White-Tailed Deer, *Odocoileus virginianus*, winter diets.

Individual forages contribute different components to the nutritional quality of White-Tailed Deer (*Odocoileus virginianus*) diets in winter (Short 1971; Ullrey et al. 1971; Mautz et al. 1976). Browse species, which can constitute the bulk of winter diets, generally are of low digestibility (Mautz et al. 1976). Certain combinations of winter forages may be crucial to deer survival during extended periods of deep snow and low temperatures. Some winter forages may contain high levels of protein and/or carbohydrate that could increase digestibility of winter diets (Ullrey et al. 1971). Addition of cornstarch to in vitro digestions increased digestibility of five winter forages (McCullough 1979), indicating synergisms in the fermentation process.

Rochelle (1980) noted that a fruticose lichen, *Alectoria sarmentosa*, increased digestibility of diets consumed by Black-Tailed Deer (*O. hemionus columbianus*). Mixed diets that contained various proportions of lichen increased diet DMD by 5–15% above levels expected from combined digestibilities of component species. That suggested that lichen was acting as a carbohydrate source (Scotter 1965), which would enable deer to use recycled urea more efficiently (Orskov 1982:28). However, Person (1975) was unable to document such synergistic effects using in vitro digestions.

Published in vitro DMD of fruticose lichens show high variability. Hanley and McKendrick (1983) found extremely low digestibilities (21.1%) for *Usnea* spp., whereas Rochelle (1980) found high digestibilities (78.1%) for *Alectoria sarmentosa*. Both arboreal lichens are taxonomically similar (Family Usneacea) (Ahmadjian and Hale 1973) and are preferred by deer (Rochelle 1980; Hanley and McKendrick 1983; Hodgman and Bowyer 1985).

During winter in parts of Maine, White-Tailed Deer can have high proportions of conifers in their diet (Crawford 1982; Ludewig and Bowyer 1985); fruticose lichens also are consumed when available (Hodgman and Bowyer 1985). In our study, diets containing lichens (*Usnea* spp. and *Evernia mesomorpha*) and coniferous forages were digested in vitro to determine: 1) possible synergistic effects among forages, and 2) effects of inocula source on digestibility estimates. Also, average in vivo DMD values (converted from in vitro digestions) were compared to average apparent digestibilities of diets determined from captive White-Tailed Deer (Jenks 1986) to determine the utility of in vitro estimates in predicting in situ digestibilities.

Methods

White Cedar (*Thuja occidentalis*), Eastern Hemlock (*Tsuga canadensis*), Balsam Fir (*Abies balsamea*), mixed spruce (*Picea* spp.), and a naturally-occurring combination of fruticose lichens (*Usnea* spp. and *Evernia mesomorpha*) were collected in January 1985 in northern Maine (45°57′N; 69°10′W). Samples were frozen until prepared for digestion trials.

Four diets were formulated that contained equal portions of the four conifers and a lichen component of 0, 5, 15, and 25%. Forage samples were dried to constant weight at 50° C and ground in a Wiley Mill through a #20 mesh screen. Duplicate 0.3 g samples of each individual forage and the four experimental diets were digested using the two-stage digestion technique of Tilley and Terry (1963), as modified by Palmer et al. (1976) for use with deer forages. If duplicate samples differed by > 5% they were discarded. Forage standards that were obtained from W. L. Palmer (Pennsylvania State University) were digested to calculate specific regression formulas for each trial for conversion of in vitro to in vivo digestibilities (Palmer and Cowan 1980). Trial specific regressions were used to control between trial error (Milchunas and Baker 1982).

Rumen inocula were obtained from a fistulated cow (16% protein diet) and three White-Tailed Deer that were maintained on the four conifers and lichen for 25 days and died during or after digestion trials (Jenks 1986). Rumen contents were squeezed through two layers of cheese cloth. Availability of deer inoculum was fortuitous and therefore limited; only lichen diets and three of the four conifer species could be digested with deer inoculum.

Following Westoby (1974), expected dietary digestibilities were calculated from single species digestions (in vivo converted DMD using deer and cow inoculum) and were compared using t-tests (Sokal and Rohlf 1981). Digestibilities of single species summations (t-tests) and mixed diets (ANOVA) also were compared to diet apparent digestibilities (Robbins 1983: 279). Apparent digestibilities of the four diets were determined in four trials (four deer/trial) by randomly assigning deer to diets for a 9–14 day pretrial period after which all feces were collected for 5–7 days (Jenks 1986).

Results

Converted in vivo DMD averaged 36.6% (SD = 8.41) for all plant species digested with cow inoculum (Table 1). In vivo DMD of mixed diets was not different from single species summations of DMD ($t = 0.048$, df = 31, $p = 0.96$) [Table 2]. Therefore, no synergisms in the mixed diets were observed with cow inoculum.

Converted in vivo DMD for forages digested with deer inoculum averaged 50.3% (SD = 9.94) [Table 1]. The four diets and a sample of hemlock were not digested with deer inoculum. Hemlock digestibility was estimated by regressing the 2 estimates of in vivo DMD (x = cow; y = deer

TABLE 1. Mean in vitro and in vivo[a] dry matter digestibilities (DMD) of plant species digested with cow and deer inoculum.

	Cow Inoculum		Deer Inoculum	
Species	In Vitro	In Vivo	In Vitro	In Vivo
Abies balsamea	37.5	33.4	33.9	46.2
SD	(0.19)	(1.91)	—	—
N[b]	3	3	1	1
Picea spp.	27.7	24.5	34.6	41.4
SD	(0.94)	(1.82)	—	—
N	3	3	1	1
Thuja occidentalis	47.1	43.3	37.9	47.4
SD	(1.18)	(2.34)	—	—
N	3	3	1	1
Tsuga canadensis	48.8	45.6	—	49.0[c]
SD	(0.79)	(1.66)	—	—
N	3	3	—	1
Usnea spp. / *Evernia mesomorpha*	39.1	36.1	43.9	67.3
SD	(6.76)	(5.83)	(8.36)	(2.33)
N	9	9	2	2

[a]Converted from in vitro estimates using procedure of Palmer and Cowan (1980).
[b]Average of duplicate estimates not exceeding a difference of 5%.
[c]Estimated using regression analysis (see text).

TABLE 2. Mean in vivo dry matter digestibilities (DMD)[a] and apparent digestibilities for diets containing a lichen component.

Percent Lichen		Dietary digestibility: Single Species Summations — Cow Inoculum	Single Species Summations — Deer Inoculum	Mixed Diets — Cow Inoculum	Mixed Diets — Apparent[b] Digestibility
0		36.7	46.0	35.4	54.3
	SD	—	—	1.6	5.8
	N	1	1	6	3
5		36.7	47.0	37.0	55.0
	SD	—	—	1.7	3.0
	N	1	1	8	3
15		36.6	49.2	35.6	56.2
	SD	—	—	2.4	5.9
	N	1	1	7	4
25		36.6	51.3	37.2	57.5
	SD	—	—	4.6	1.0
	N	1	1	8	4

[a]Converted from in vitro digestibility following Palmer and Cowan (1980).
[b]Determined from digestibility trials (Jenks 1986).

inocula) for the remaining three conifers and predicting in vivo DMD for hemlock with its in vivo estimate (x) and the regression formula ($Y = 33.595 + 0.338X$; $r^2 = 0.93$) [Table 1]. Expected in vivo DMD estimates for diets were determined with those single species estimates of digestibility (Table 2).

A two-factor analysis of variance comparing converted (cow inoculum) and apparent digestibility for the four lichen diets (method × percent dietary lichen) was significant ($F_{4,38} = 17.76$, $p < 0.001$) [Table 2]. No differences were found among diets containing a lichen component ($F_{3,38} = 0.19$, $p > 0.10$); however, apparent digestibilities were significantly higher than converted in vivo DMD estimates (cow inoculum ($F_{1,38} = 70.49$, $p < 0.001$). Expected in vivo DMD estimates using deer inoculum also were higher than those determined with cow inoculum ($t = 9.92$, df = 6, $p < 0.001$) but lower than apparent digestibilities from digestion trials ($t = 3.51$, df = 16, $p = 0.003$) [Table 2; Figure 1].

Converted in vivo DMD of lichen in deer inoculum (67.3%) [Table 1] was nearly twice that of cow inoculum estimates (36.1%). As a result, any increase in lichen proportion in simulated deer diets increased expected dietary DMD (Figure 1). The same effect was not noted for cow inoculum because DMD estimates of lichen were similar to those of conifers (Table 1).

Statistical test scores presented here do not agree with those in Jenks (1986). Errors in computation did not affect interpretation and were corrected during editorial review.

Discussion

Thomas and Kroeger (1981) and Thomas et al. (1984) suggested that low in vitro DMD of lichens commonly ingested by Caribou (*Rangifer tarandus*) resulted because of low nitrogen or temporal conditions that limited digestibility. A 60 hr fermentation stage increased digestibility of lichens (Thomas and Kroeger 1981; Thomas et al. 1984); however, no increase in digestibility of browse was observed. Milchunas and Baker (1982) found no relationship for between trial differences in forage digestibility and nitrogen concentration of inoculum. We digested the lichen combination for 60 hr in cow inoculum (in vivo DMD = 36.3%) but observed no increase in digestibility above the regular 48 hr fermentation (in vivo DMD = 36.1%) [Table 1].

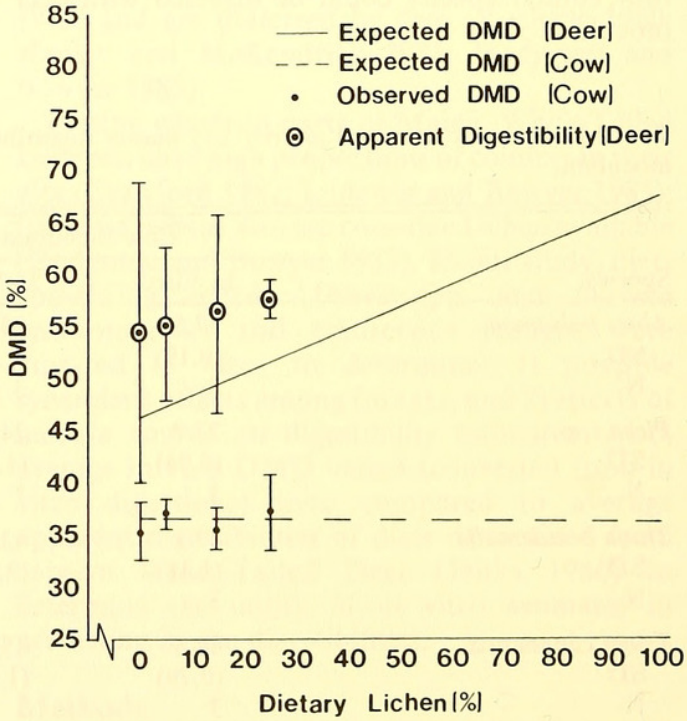

FIGURE 1. Relationship between percent lichen for four diets (0, 5, 15, and 25% lichen) containing equal portion of four conifer species and in vivo dry matter digestibility (DMD) (converted from in vitro digestions with cow and deer inocula) and apparent digestibilities determined from captive White-Tailed Deer (Jenks 1986). Bars indicate confidence intervals.

Overall digestibility for winter forages was low when digested with cow inoculum; digestibility of all forages increased when digested with deer inoculum (Table 1). Similarly, Blankenship et al. (1982) found an overall increase in digestibilities for forages fermented with deer inoculum compared to those from cow, goat, and sheep inocula. Campa et al. (1984) also found differences between forage digestibilities determined with cow and wild deer inocula, as well as a difference in forage digestibility when captive, fistulated deer were maintained on the diet being digested. Our study suggested that two inocula can produce disparate estimates of digestibility. Conversely, other researchers have found little variation in digestibility due to inoculum donor (Welch et al. 1983; Crawford and Hankinson 1984).

Differences in in vivo DMD of diets between inoculum sources approached 20% (Figure 1). Robbins et al. (1975) noted an 4.4% difference in digestibilities obtained using cow and deer inocula; cow inoculum overestimated browse digestibilities. Digestibilities of conifers obtained using wild deer inoculum (Rochelle 1980) were higher than those from cow inoculum (Leslie 1982). These disparities suggest that inoculum source and/or donor diet may significantly affect in vitro results.

Differences between expected in vivo DMD estimates (deer inoculum) and apparent digestibility values indicated that dietary digestibilities from summations of single species digestions can give inaccurate results. These differences may be greatest when diets contain forages of varying solubility (Milchunas and Baker 1982). In such instances, in vitro techniques may provide relative relationships among forages but not accurate in vivo digestibilities (Campa et al. 1984).

No lichen induced synergisms were found that enhanced digestibility of winter diets (digested with cow inoculum) as noted by Rochelle (1980); however, lichen diets could not be digested with deer inoculum. Calculated dietary digestibilities determined from single species summations (deer inoculum) increased as the more digestible lichen increased; in vivo DMD increased to levels above 50% with a minimum of 20% dietary lichen (Figure 1). Ammann et al. (1973) observed a positive energy balance in deer when diets were above 50% digestible dry matter. Available lichen may enhance energy balance during winter when poorly digested browse species make up the bulk of winter diets. Lichens also may act as carbohydrate sources to increase efficiency of urea cycling (Orskov 1982: 28).

Although lichen induced synergisms could not be demonstrated in our study, availability of lichen in winter could have important implications for deer management in boreal habitats. When lichen is not available and deer consume browse of low digestibility ($< 50\%$), digestible energy may be limiting. Ingestion of some lichen species may increase fermentative efficiency and overall digestibility of the diet. In areas where lichens are available, dietary digestibility would be increased through an additive effect of increased dietary lichen (Figure 1).

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