

Reliability of Captive Deer and Cow In Vitro Digestion Values in Predicting Wild Deer Digestion Levels

HENRY CAMPA, III, DAVID K. WOODYARD, AND JONATHAN B. HAUFLE

Abstract

The in vitro dry matter digestibility (IVDMD) values of 9 forages were compared using rumen fluid collected from wild white-tailed deer (*Odocoileus virginianus*), captive white-tailed deer, and a Holstein cow. Five of the 9 forages analyzed displayed significantly different ($P < 0.05$) IVDMD between wild deer and captive deer inocula and between wild deer and cow inocula. Differences were attributed to the diet differences of the donor animals, which may have influenced the composition of microorganisms within the rumen. In vitro dry matter digestibility of all 5 forages considered together decreased significantly ($P < 0.05$) when rumen inocula were stored at various intervals. This study indicates that IVDMD values obtained using inoculum from captive and domestic animals on commercial diets should only be used by researchers to make comparisons of forages, not to predict actual digestibility levels by wild animals. In addition, in vitro samples should be inoculated as soon after collection as possible in order to obtain reliable data.

The quantity and quality of available forage is a major factor affecting the quality of rangelands for wildlife. Biologists have attempted to determine the nutritional quality of various forages and to evaluate digestibility by different animals. In order to determine how well selected wildlife species, such as ruminants, can utilize various forages, researchers have conducted digestibility experiments. Traditionally, digestion trials involving the feeding of animals were used. However, researchers encountered problems using this technique including expense, time, and difficulties in obtaining large amounts of forage (Palmer 1976). Because of such problems, in vitro procedures have been developed to estimate the degree of forage digestion by ruminants (Tilley and Terry 1963, Pearson 1970).

The advantage of using in vitro procedures is that they are relatively inexpensive and a large number of forage samples can be analyzed at one time using only a small amount of sample. Like conventional digestion methods, however, in vitro digestibility procedures also have their limitations in practice and application. Because it is often impractical to obtain inoculum from the wild ruminant under investigation, researchers have substituted, using inoculum from captive animals of the same species, or of a different species, in an attempt to quantify how well free ranging animals are using various forages (Palmer et al. 1976, Blankenship et al. 1982). However, because the composition of microorganisms within the rumen is dependent on the diet of an animal (Church 1969, Maynard et al. 1979), differences in diets may influence an animal's ability to digest a forage. Some researchers have documented that the use of inoculum from different ruminant species, on various diets, affected the IVDMD of various forages (Robbins et al. 1975, Horton et al. 1980). Welch et al. (1983), however, found no significant difference in the IVDMD of 25 forages among 6 ruminant species. Conflicting results may have been caused by differences in

the chemical composition of test forages or in differences in laboratory procedures.

This study investigated the feasibility of using cow and/or captive white-tailed deer (*Odocoileus virginianus*) as the inoculum source for estimating in vitro digestion in wild white-tailed deer. In addition, the effect that the time interval between collection of rumen fluid and inoculation of samples has on in vitro dry matter digestibility (IVDMD) levels was also investigated.

Methods

To investigate the effects various inoculum sources have on IVDMD of selected forages, rumen fluid was collected from wild white-tailed deer, captive white-tailed deer, and a nonlactating rumen-fistulated Holstein cow. Captive deer were fed commercial deer pellets. The Holstein cow was maintained on a diet of good quality alfalfa hay.

Three replicate trials were conducted using captive deer and cow inoculum and duplicate trials were run using wild deer rumen fluid. Rumen fluid for cow replicates was obtained from 1 animal on 3 different days in March, 1983. To obtain inoculum from captive deer, three 7-8 month old, male deer were sacrificed from the Houghton Lake Wildlife Experiment Station, Houghton Lake Heights, Mich., in February, 1983. Inoculum from wild deer was obtained by sacrificing 2 adult does: 1 in October, 1982, from an aspen (*Populus* spp.) clearcut, where vegetation samples were collected in Montmorency County Mich., and 1 in February, 1983, from an agricultural area in Ingham County Mich. The food habits of these 2 deer were not determined but, based on studies conducted in similar areas, their diets were assumed to be fairly diverse (Rogers et al. 1981).

Rumen fluid from all 3 sources was strained through 4 layers of cheesecloth into an Erlenmeyer flask and placed in a waterbath (39°C) for transportation to the laboratory. Inoculum was kept in a waterbath maintained at 39°C and flushed with CO₂ until inoculation. All samples, except those used in the inoculum storage test, were inoculated within 1 hr of obtaining rumen fluid. Analytical procedures used were described by Tilley and Terry (1963), modified by Michigan State University, Department of Dairy Science. Modifications included the use of a phosphate-carbonate buffer solution (Table 1) and using a ratio of 12 ml of rumen fluid to 10 ml buffer solution.

The plant samples used for digestion trials included orange-hawkweed (*Hieracium aurantiacum*), wild strawberry (*Fragaria virginiana*), panic grass (*Panicum virgatum*), and separate leaf and twig samples from 3 woody species: black cherry (*Prunus serotina*), trembling aspen (*Populus tremuloides*), and big-tooth aspen (*P. grandidentata*). Vegetation samples were collected in June, 1982, from a 10-year-old aspen clearcut. Tissues collected were restricted to the current annual growth of randomly selected individuals. These samples were dried at 60°C and ground in a Wiley mill to pass a 1.0-mm screen.

Authors are graduate students and assistant professor, Department of Fisheries and Wildlife, Michigan State University, East Lansing 48824.

This manuscript is Journal Article No. 10896 from the Michigan Agricultural Experiment Station.

Manuscript accepted January 9, 1984.

Table 1. Composition of in vitro buffer solution.

Ingredient	g/L
Na ₂ HPO ₄	8.72
KH ₂ PO ₄	4.08
MgSO ₄ •7H ₂ O	1.5
KCl	0.5
CaCl ₂	0.1
Na ₂ S•9H ₂ O	0.025
Na ₂ CO ₃ solution, 15.73g/100 ml	20(ml/L)
Urea solution, 8.0g/100 ml	10(ml/L)

Statistical analysis of the data was with analysis of variance and Duncan's new multiple-range test. Barlett's test was conducted on all data to test for homogeneity of variance. Data which had heterogeneous variances were subjected to an arc sine transformation (Steel and Torrie 1980).

Tests for effects of storage time of inoculum on IVDMD from 3 captive adult male deer from the Houghton Lake Wildlife Experiment Station and the Holstein cow were conducted. In vitro digestion trials were initiated 40 minutes after rumen fluid collection, with subsequent samples inoculated at 7, 20-minute intervals, with the final inoculation at 3 hr. Vegetation samples for this phase of the project consisted of cherry (*Prunus* spp.) leaves, cherry twigs, sedge (*Carex* spp.), orange-hawkweed, and panic grass collected from a 5-year-old jack pine (*Pinus banksiana*) clearcut. Subsamples of each of the 5 vegetation samples were used in all 8 time periods. Vegetation was dried, separated, and ground by methods stated earlier.

Regression equations were calculated to determine the relationship between IVDMD and storage time of the rumen fluid.

Results

Of the 6 samples from the 3 woody species analyzed, only 2 showed significantly different digestibility levels among inoculum sources (Table 2). Black cherry twigs and trembling aspen leaves had IVDMD levels which were significantly different between wild deer and captive deer and between wild deer and cow. Black cherry leaves were the only species which displayed significantly different IVDMD levels among all combinations of inoculum sources. Two of the 3 herbaceous species analyzed show significantly different IVDMD values between wild deer and captive deer inoculum sources and between wild deer and cow (Table 2). There were no significant differences in IVDMD between individual wild deer and individual captive deer.

Results of inoculum storage time trials showed that IVDMD of all species combined significantly decreased ($P<0.05$) over 3 hr using both captive deer and cow inoculum (Fig. 1). The IVDMD of

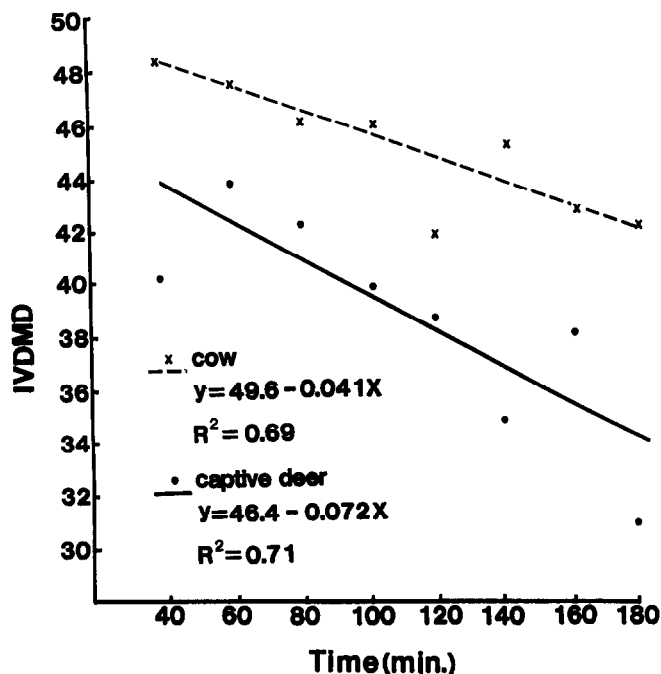


Fig. 1. Effects of time prior to inoculation on the in vitro dry matter digestibility of forages (mean values of 5 forages at each time) using cow and captive deer inoculum.

individual forage species did not show a significant decline over time using captive deer inoculum. Using cow inoculum, however, cherry leaves, panic grass, and orange-hawkweed all showed a significant decrease in IVDMD ($P<0.05$) (Fig. 2).

Discussion

Results indicate that of the forages used for the in vitro digestion comparison trials, 5 (56%) had significantly different IVDMD values between wild deer and captive deer and between wild deer and cow. These data suggest that in vitro digestion of some forages by wild deer differed significantly from both captive deer and cow. This dissimilar capability to efficiently utilize the selected forages may have been caused by differences in the diet composition of the respective donor animals. Since the captive deer and cow were fed a homogeneous ration of good nutritional quality, the diversity of microorganisms which inhabit the rumen may have been limited. Robbins (1983) stated that the inoculation of bacteria into the

Table 2. Mean IVDMD of forage species from three inoculum sources. Number of animals used for each inoculum source is indicated in parentheses.

Forage species	Inoculum sources		
	Wild deer (2)	Captive deer (3)	Cow (3)
<i>Fragaria virginiana</i> (wild strawberry)	68.8a ¹	30.9b	45.4b
<i>Hieracium aurantiacum</i> (orange-hawkweed)	63.6	56.3	57.8
<i>Panicum virgatum</i> (panic grass)	54.5a	31.8b	38.9b
<i>Prunus serotina</i> (black cherry) leaves	34.2a	44.5b	51.3c
<i>P. serotina</i> (black cherry) twigs	52.7a	26.9b	33.1b
<i>Populus tremuloides</i> (trembling aspen) leaves	50.3a	26.7b	28.9b
<i>P. tremuloides</i> (trembling aspen) twigs	47.6	39.7	41.9
<i>Populus grandidentata</i> (big tooth aspen) leaves	41.9	47.9	49.3
<i>P. grandidentata</i> (big tooth aspen) twigs	56.3	43.2	53.2

¹Any two means within a row with different letters indicate a significant difference ($P<0.05$).

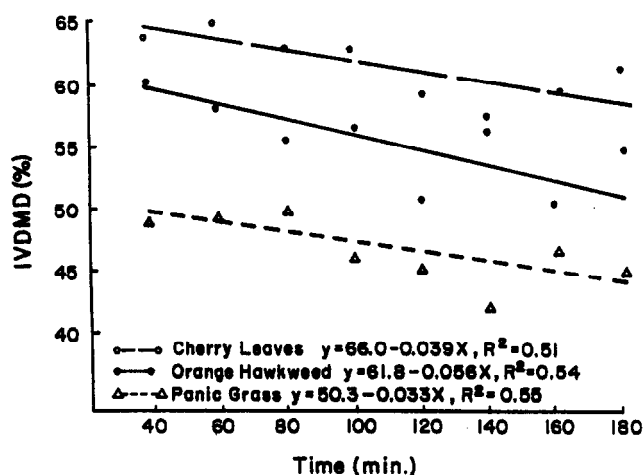


Fig. 2. Effects of time prior to inoculation on the in vitro dry matter digestibility of 3 forages using cow inoculum.

neonate rumen is often dependent on such factors as the feed handled by the mother or maternal feces touched. Therefore, if wild deer have the opportunity to feed on a variety of forages, it is more likely they are in contact with a greater diversity of microorganisms and therefore may be more capable of utilizing a greater diversity of forages than domestic or penned individuals (Blankenship et al. 1982). Good populations of microorganisms can also be influenced by the nutritional quality of the diet of an animal (Church 1969), so poor quality diets could also influence IVDMD, regardless of the likely diversity of microorganisms present. However, all individuals used in this study were in excellent condition and assumed to be on a diet of good nutritional quality. Therefore, differences in rumen microorganisms appear to be responsible for the differences in IVDMD for some forages. The forage species which exhibited nonsignificantly different IVDMD between inoculum sources could be attributed to a possible dissimilarity in the chemical composition of these forages to the diets of one or more of the donor animals.

The results from the time trials for both captive deer and cow inoculum paralleled the results by Schwartz and Nagy (1972) using mule deer (*Odocoileus hemionus*). In vitro dry matter digestibility significantly decreased after approximately 2 hr of storage. In addition, IVDMD levels became more variable at times beyond 2 hr.

The nonsignificant results obtained for time trials when considering individual forage species and using captive deer inoculum may be attributed to the diet composition of donor animals. Because captive deer were fed a high protein pelleted ration, instead of a forage fed to the cow, appropriate microorganisms may not have been present to digest forages adequately enough to detect changes in digestibility of individual species over time, given the variance associated with digestion trials of one sample at each time. Using cow inoculum, however, significant decreases in IVDMD were detected with cherry leaves, panic grass, and orange-hawkweed. Those results may be attributed to possible similarities, such as nutrient content and/or stage of maturity, between the 3 forage species and the alfalfa hay the cow was fed. Previous analysis conducted on the 3 forage species indicated crude protein contents ranged from 17 to 23% while lignin content was 5.6 to 8.5% (Campa 1982). These crude protein values are similar for the protein content (18.1%) of second cutting alfalfa hay reported by Crampton and Harris (1969). According to Van Soest (1975) lignin must compose at least 40% of the total cell-wall constituents for the

digestion of the cell-wall constituents to cease. Lignin content for these 3 species was below this level (only 11 to 27%), therefore cell-wall constituents were able to be partially digested. Cherry twigs and sedge did not show the significant decreases in IVDMD over time using cow inoculum, possibly due to the low protein and high fiber contents of these species. The cell-wall constituent values for these 2 species were 66 and 72% respectively, while crude protein contents for the 2 species were 9 and 12%, respectively (Campa 1982). These cell-wall values are considerably higher than crude fiber content (31.2%) of alfalfa hay reported by Crampton and Harris (1969) and crude protein values are considerably lower. Because lignin may hinder the utilization of vegetation by ruminants (Van Soest 1965, Milchunas and Baker 1982) by binding to other fiber constituents, sedge and cherry twigs may not have been digested sufficiently to detect changes in IVDMD over time.

From these results it appears that inoculum from captive deer and cows cannot be used to accurately estimate IVDMD in wild deer if animals are fed dissimilar diets. However, the relative values from in vitro digestibility trials using captive deer or cows as the inoculum source can still provide useful comparisons of the utilization of forages.

The length of storage time between collection of rumen fluid and the inoculation of samples is a variable researchers should consider before conducting any in vitro digestion trial. If laboratory facilities are not in close proximity to donor animals, one may obtain depressed values and variable data. Therefore, researchers should attempt to minimize inoculum storage time by inoculating samples as soon after collection as possible.

Literature Cited

- Blankenship, L.H., L.W. Varner, and G.W. Lynch. 1982. In vitro digestibility of South Texas range plants using inoculum from four ruminants. *J. Range Manage.* 35:664-66.
- Campa, H., III. 1982. Nutritional response of wildlife forages to municipal sludge application. M.S. Thesis, Michigan State Univ., East Lansing.
- Church, D.C. 1969. Digestive physiology and nutrition of ruminants. Vol. 1. Oregon State University Book Stores, Inc., Corvallis.
- Crampton, E.W. and L.E. Harris. 1969. Applied animal nutrition. W.H. Freeman and Company. San Francisco, Calif.
- Horton, A.M., D.A. Christensen, and G.M. Steacy. 1980. In vitro fermentation of forages with inoculum from cattle and sheep fed different diets. *J. Agron.* 72:601-605.
- Maynard, L.A., J.K. Loosli, H.F. Hintz, and R.G. Warner. 1979. Animal nutrition. McGraw-Hill Book Co., New York.
- Milchunas, D.G., and D.L. Baker. 1982. In vitro digestion-sources of within- and between-trial variability. *J. Range Manage.* 35:199-203.
- Palmer, W.L., R.L. Cowan, and A.P. Ammann. 1976. Effect of inoculum on in vitro digestion of deer foods. *J. Wildl. Manage.* 40:301-307.
- Pearson, H.A. 1970. Digestibility trials: in vitro techniques. p. 85-92. In: H.A. Paulsen, E.H. Reid, and K.W. Parker, eds. Range and wildlife habitat evaluation-A research symposium. USDA Forest Serv. Misc. Publ. 1147.
- Robbins, C.T. 1983. Wildlife feeding and nutrition. Academic Press, Inc. New York, N. Y.
- Robbins, C.T., P.J. Van Soest, W.W. Mautz, and A.N. Moen. 1975. Feed analysis and digestion with reference to white-tailed deer. *J. Wildl. Manage.* 39:67-79.
- Rogers, L.L., J.J. Moaty, and D. Dawson. 1981. Foods of white-tailed deer in the Upper Great Lakes Region-a review. USDA Forest Serv. Gen. Tech. Rept. NC-65.
- Schwartz, C.C., and J.G. Nagy. 1972. Maintaining deer rumen fluid for in vitro digestion studies. *J. Wildl. Manage.* 36:1341-1343.
- Steel, R.G.D., and J.H. Torrie. 1980. Principles and procedures of statistics, a biometrical approach. McGraw-Hill Book Co., New York.
- Tilley, J.M.A., and R.A. Terry. 1963. A two-staged technique for the in vitro digestion of forage crops. *J. Brit. Grassland Soc.* 18:104-111.
- Van Soest, P.J. 1975. Physio-chemical aspects of fiber digestion. p. 351-365. In: E.W. McDonald and A.O.I. Warner, eds. Digestion and metabolisms in the ruminant. Proc. IV Int. Symp. on Ruminant Physiology. Univ. New Engl. Publ. Unit. Armidale, NSW 2351 Australia.
- Welch, B.L., J.C. Pederson, and W.P. Clary. 1983. Ability of different rumen inocula to digest range forages. *J. Wildl. Manage.* 47:873-877.