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# Hemicellulose Monosaccharide Composition and in Vitro Disappearance of Orchard Grass and Alfalfa Hay

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Samples of alfalfa and orchard grass hays were incubated in rumen fluid for 0, 6, 12, 18, 24, 36, 48, and 72 h to compare monosaccharide composition and extent and rate of disappearance of the two forages. Hemicellulose monosaccharides were quantified in the neutral detergent fiber (NDF) residue of each sample. The xylose concentration of alfalfa increased more rapidly during in vitro fermentation than did that of orchard grass. Xylose to arabinose ratios increased with digestion time for both alfalfa and orchard grass, suggesting that the arabinose component of hemicellulose is more digestible than is the xylose component. Xylose to glucose ratios increased for both alfalfa and orchard grass, suggesting that the cellulose component is more rapidly degraded than is hemicellulose. The extent of NDF digestion was higher for orchard grass than for alfalfa. Uronic acids and glucose were the cell wall monosaccharides that were more digestible in orchard grass than in alfalfa.

The majority of feed and food energy in the world is in the form of plant material, which is not utilizable by humans. The study of plant cell wall components is important, not only for a basic understanding of structural integrity but also for accurate determination of nutrient availability to ruminants. Cellulose, hemicellulose, and pectin are the major structural polysaccharides found in plant material. Hemicellulose is the most complex of the fiber components of common forages (Bailey, 1973; Van Soest, 1982; Wilkie, 1979), and it needs better definition and characterization.

On grass diets, both ruminants and nonruminants digest more hemicellulose than cellulose (Daughtry et al., 1978; Keys et al., 1967; Van Soest, 1982). On legume diets, ruminants digest more cellulose than hemicellulose, but nonruminants still digest more hemicellulose than cellulose (Keys et al., 1967). Grasses show a wider range in hemicellulose digestibility values than does alfalfa (Van Soest, 1973). An increase in total structural polysaccharides fed to humans has been shown by Slavin and co-workers (1983) to decrease dry-matter digestibility of the diet. In a digestibility trial with sheep, Sullivan (1966) demonstrated that dry-matter (DM) digestibility and percent hemicellulose were significantly negatively correlated.

Ruminants have the capability to digest all structural polysaccharides. The majority of hemicellulose digestion takes place in the rumen and abomasum (Dehority, 1973). Mean rumen turnover time for fiber particles is 30–50 h (Van Soest, 1973). Since grasses usually take longer to completely break down than do legumes, their digestibilities tend to be lower than digestibilities of alfalfa (Van Soest, 1973).

The digestion of hemicellulose is subject to interference by lignin (Bailey, 1973; Bittner, 1983; Burdick and Sullivan, 1963; Van Soest, 1973, 1982). The digestibility of hemicellulose is closely related to that of cellulose and negatively related to lignification. Sullivan (1966) calculated a correlation coefficient of -0.83 between hemicellulose digestibility and lignin content in alfalfa.

Hemicellulose forms bonds with lignin (Van Soest, 1982; Wilkie, 1979). Use of <sup>13</sup>C nuclear magnetic resonance (NMR) indicates the presence of lignin-carbohydrate complexes (Barton et al., 1982). These lignin-carbohydrate complexes result in decreased recoveries when acid hydrolysis is used to extract hemicellulose (Barton et al.,

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1982). There is a certain amount of ester bonding between phenolic components of lignin and xylose, arabinose, and uronic acids of heteroxylans of hemicellulose (Barton et al., 1982; Harkin, 1973). The amount of bonding appears to increase with plant maturity (Bailey, 1973). Lignin inhibits digestion of hemicellulose by steric hindrance as well as by direct bonding to hemicellulose (Gaillard, 1962; Sullivan, 1966). No uniform effects can be found between grasses and legumes in terms of the effects of lignification (Van Soest, 1973). Barring interference from lignin, the heteroxylan of hemicellulose is variably digestible. Furanose linkages are easily hydrolyzed, but linkages involving uronic acids are resistant (Bailey, 1973). Xylose is most resistant of the chemical constituents of hemicellulose to acid hydrolysis (Burdick and Sullivan, 1963; Morris and Bacon, 1976).

Our purpose was to compare monosaccharide composition and in vitro digestibilities and rates of disappearance of hemicellulose monosaccharides of alfalfa and orchard grass hay.

## MATERIALS AND METHODS

Alfalfa (Medicago sativa L.) (IFN 1-00-063) and orchard grass (Dactylis glomerata L.) (IFN 1-03-438) were harvested as first-cutting baled hay at prebloom and at prehead, respectively. Forty random core samples of alfalfa and orchard grass were ground through a 1-mm screen and served as substrate for in vitro studies.

In Vitro Procedures. Samples of ground hay (0.5 g) were weighed in triplicate for removal times of 0, 6, 12, 18, 24, 36, 48 and 72 h. In vitro procedures are described in Goering and Van Soest (1970), Gaillard (1962), and Weller and Pilgrim (1974). The Kansas State buffer system was used (Marten and Barnes, 1979). Ground alfalfa and orchard grass samples were allowed to equilibrate with the buffer overnight at 39.5 °C in  $10 \times 175$  cm incubation tubes. Rumen fluid was collected from four ruminally cannulated steers, strained through four layers of cheesecloth, and pooled. Two steers were consuming all orchard grass diets, while the others were maintained on all alfalfa. Feed samples were inoculated with 30 mL of rumen fluid and placed in an incubator at 39.5 °C. Temperature of the rumen fluid was maintained during inoculation by the use of a water bath (39.5 °C). Carbon dioxide was bubbled through the rumen fluid to maintain anaerobiosis. Tubes were swept with  $CO_2$  and immediately stoppered after inoculation with rumen fluid. Upon removal, each sample was extracted with neutral detergent fiber (NDF) solution (Goering and Van Soest, 1970) and the residue saved for analysis of hemicellulosic sugars.

**Preparation of Monosaccharides.** Dry matter was determined by drying in a forced-air oven at 105 °C for 24 h, and nitrogen was quantitated by a micro-Kjeldahl procedure (AOAC, 1975). Neutral detergent fiber (NDF), acid detergent fiber (ADF), and acid detergent lignin (ADL) were determined by the methods of Goering and Van Soest (1970).

Neutral sugars of alfalfa and orchard grass were obtained by acid hydrolysis of the NDF fraction using the procedure of Blakeney and co-workers (1983). NDF determinations were made on three 0.5-g portions of each sample. All experimental values were determined in duplicate. A 20mg sample of the NDF residue was weighed into hydrolysis tubes. After a 45-min primary hydrolysis in 12 N H<sub>2</sub>SO<sub>4</sub>, the sample was diluted with 2.7 mL of distilled water to reduce its concentration to 1 M for secondary hydrolysis. A 3-h secondary hydrolysis was performed in a block heater at 100 °C. The hydrolysate was neutralized with 0.64 mL of 15 M ammonium hydroxide. Recoveries for acid hydrolysis were calculated by subtracting acid detergent lignin and acid detergent insoluble ash from NDF residues. Orchard grass and alfalfa recoveries of total sugars were 64.0% and 65.5%, respectively. myo-Inositol was added as an internal standard to correct for any sugar losses during subsequent acetylation (Collings and Yokoyama, 1979). A 100- $\mu$ L aliquot of a 2 g/100 mL solution of myo-inositol was added to give 0.5 mg/mL of myo-inositol in the solution. Sugar standards including rhamnose, arabinose, xylose, mannose, galactose, glucose, and myo-inositol were prepared, reduced, and acetylated. The gas chromatograph was calibrated on the basis of losses due to derivatization and acetylation of prepared standards.

A 200- $\mu$ L aliquot of the acid hydrolysate was taken, and sugars were reduced to their corresponding alditols by addition of 2 mL of sodium borohydride in dimethyl sulfoxide (2 g/100 mL). Reduction was done at 40 °C by means of a dry-heating block. A 200- $\mu$ L aliquot of glacial acetic acid was added to decompose excess sodium borohydride. 1-Methylimidazole (0.4 mL) was added as a catalyst for the acetylation reaction. Acetic anhydride (4 mL) was added to acetylate the reduced sugars to their corresponding alditol acetates. Water (10 mL) was added to decompose excess acetic anhydride. Dichloromethane (2 mL) was added to the acetylated sample to solubilize the alditol acetates. Samples were then centrifuged at 2000g to separate the phases. The bottom layer was pipetted into a septum cap vial prior to monosaccharide determination.

**Determination of Monosaccharides.** A Hewlett-Packard 5890A gas chromatograph was used for separation of neutral sugars (rhamnitol, arabinitol, xylitol, mannitol, galactitol, glucitol). A flame ionization detector system was used with a 1.8-m length  $\times$  2-mm i.d. silanized glass column, packed with 3% SP-2330 on 100/120-mesh Supelcoport packing (Supelco, 1977). The SP-2330 is a cyanosilicone stationary phase, which gave relatively rapid separation of the alditol acetates quantitated in this study. Nitrogen gas was the carrier phase used at a flow rate of 30 mL/min. An aliquot of 1–2  $\mu$ L was injected into the column for each sample. Temperatures: 250 °C, injection port; 225 °C, oven; 275 °C, detector. A Hewlett-Packard 3392A integrator was used to quantify the alditol acetates.

Assay of Uronic Acids. The procedure for uronic acid quantification was outlined by Blumenkrantz and Asboe-Hanson (1973). A Shimadzu 120 spectrophotometer was used to measure uronic acids at a wavelength of 520 nm. A 200- $\mu$ L aliquot of the acid hydrolysate of NDF was diluted 1:10 with distilled water. Sodium tetraborate in concentrated sulfuric acid (2.4 mL of 4.76 g of NaB<sub>4</sub>O<sub>7</sub>/L of H<sub>2</sub>SO<sub>4</sub>) was added to 0.4 mL of the diluted sample. Samples were heated for 5 min at 100 °C in a dry block. Upon cooling, 40  $\mu$ L of color reagent (150 mg of 3phenylphenol diluted to a 100-mL volume with 0.125 M NaOH) was added. A blank was run for each sample to correct for any glucose interfering with absorbance. A standard curve was calculated with 0.005, 0.01, 0.02, 0.04, and 0.06 mg/mL of glucuronic acid.

**Calculations and Statistical Analysis.** First-order kinetics (Moore et al., 1985; Smith et al., 1972) were used to analyze the data for the in vitro studies. The extent of digestion was defined as the amount degraded at 72 h. In vitro degradation of cell walls approaches an asymptote in 72 h or less and can be considered complete (Smith et al., 1972). The amount of NDF digested at a given time was calculated as the difference between original concentration and residual amount and was expressed as a per-

Table I. Chemical Composition of Alfalfa and Orchard Grass Hay

component	alfalfa	orchard grass	SE
	Dry Matter,ª	%	
$hemicellulose^{b}$	16.1	29.4	0.86*
cellulose <sup>c</sup>	25.6	30.6	0.74*
acid detergent lignin	7.2	4.5	0.15*
crude protein	18.9	10.2	0.74*
Acid	l Hydrolysate,	<sup>d,e</sup> mg/g	
arabinose	38	79	3.8
xylose	96	180	3.2*
mannose	19	52	3.2
galactose	53	25	1.0*
glucose	595	563	9.2
uronic acids	40	62	4.7*

<sup>a</sup> Standard error of means for four observations. <sup>b</sup> Hemicellulose = neutral detergent fiber (NDF) minus acid detergent fiber (ADF). <sup>c</sup> Cellulose = ADF minus ADL minus acid detergent insoluble ash. <sup>d</sup> Acid hydrolysate of NDF residue expressed as milligrams/gram of NDF. <sup>e</sup> Standard error of means of six observations. Asterisk: P < 0.05.

centage. The residue at 72 h subtracted from the initial concentration represents the potentially digestible fraction (Moore et al., 1985). The rate constant was calculated as the slope of the line obtained by regressing the natural logarithm of the percent potentially digestible NDF against time. Alfalfa and orchard grass rate constants, lag times, and extents of digestion were compared by t test (Steel and Torrie, 1980).

Analyses of variance were performed on concentrations of monosaccharides and xylose to arabinose and xylose to glucose ratios at various fermentation times (Steel and Torrie, 1980). Those variables that demonstrated a significant effect of time on concentration were tested for linear and quadratic components by regression analysis. Differences in digestibility of individual monosaccharides between alfalfa and orchard grass were tested by t test (Steel and Torrie, 1980).

### RESULTS AND DISCUSSION

The average chemical compositions of alfalfa and orchard grass hays are listed in Table I. Alfalfa had a higher crude protein content than did orchard grass. The use of a grass and a legume allowed comparisons of two different sources of hemicellulose. Alfalfa had lower concentrations of hemicellulose and cellulose and higher lignin content than did orchard grass.

The monosaccharide compositions of acid hydrolysates of neutral detergent fiber are found in Table I. Glucose, xylose, and uronic acids are the major sugars (85%) of alfalfa hemicellulose (Ben-Ghedalia and Miron, 1984). Similar concentrations of xylose and uronic acids were noted in this study, 89% for alfalfa and 85% for orchard grass. The xylose content of orchard grass (180 mg/g) was significantly (P < 0.05) higher than that in alfalfa (96 mg/g). Hungate and co-workers (1983) noted that xylose comprised 10% of total alfalfa cell wall monosaccharides. Xylose content is indicative of the amount of hemicellulose in the forage (Morris and Bacon, 1976). A lower xylose concentration indicates proportionately less hemicellulose and more cellulose in the cell wall. Galactose content in the cell wall was significantly higher in alfalfa than in orchard grass. Orchard grass contained a higher uronic acid content than did alfalfa. Rhamnose was present in small amounts in alfalfa but was not detected in orchard grass.

In comparing cellulose and hemicellulose content, as determined by the Van Soest detergent system, with the total amount of hemicellulosic and cellulosic sugars, the

 Table II. Neutral Detergent Fiber Digestion Kinetics of

 Alfalfa and Orchard Grass Hay in Vitro

	alfalfa	orchard grass	SEª
rate constant, h <sup>-1</sup>	0.056	0.049	0.0028
lag time, h	3.2	4.1	0.60
extent at 72 h, %	54.4	61.6	0.72*

<sup>a</sup>Standard error of means of 24 observations. Asterisk: P < 0.05.



**Figure 1.** Disappearance of alfalfa and orchard grass neutral detergent fiber during in vitro digestion with rumen microorganisms.

Van Soest system consistently overestimated hemicellulose and underestimated cellulose. By using the Van Soest system, hemicellulose should have comprised 329 and 456 mg/g of the NDF residue for alfalfa and orchard grass, respectively. Values obtained by totalling hemicellulosic sugars were 246 and 398 mg/g for alfalfa and orchard grass, respectively. Alfalfa and orchard grass cellulose values were 522 and 472 mg/g, respectively, for Van Soest's detergent system and 595 and 563 mg/g, respectively, for cell wall monosaccharide totals. One explanation for this difference is that all glucose isolated was attributed to the cellulose fraction. Hemicellulose has glucose linked to the xylan backbone of the polymer (Bailey, 1973). As a result, cellulose values would be inflated, while hemicellulose would be underestimated when using individual monosaccharides to calculate their relative proportions on the forage.

In vitro NDF digestion kinetics of alfalfa and orchard grass appear in Table II. Utilizing NDF extraction of in vitro residues vs. dry matter for digestion kinetics is advantageous because neutral detergent solution removes all microbial matter and leaves a residue of undigested cell walls. A measure of true digestibility is obtained by removing bacterial debris (Van Soest, 1982). The rates of NDF disappearance in vitro were not significantly different for alfalfa and orchard grass hays. Lag is a substrate-related phenomenon that has an effect that increases at an exponential rate, indicating it is an extremely important variable in digestion (Mertens, 1977). Lag time was not significantly different between alfalfa and orchard grass hay. Extent of NDF digestion was significantly higher for orchard grass than for alfalfa. Coefficients of determination for the natural logarithm of the potentially digestible NDF disappearance vs. time were 0.97 and 0.96 for orchard grass and alfalfa, respectively. Alfalfa and orchard grass NDF disappearance plotted against time in vitro are shown in Figure 1. Alfalfa contained less NDF than orchard grass

Table III. A	Alfalfa and (	<b>Orchard Grass</b>	Monosaccharide	<b>Composition</b> at	Various in	Vitro Fermentation Times
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	acid hydrolysate," mg/g						
time, h	arabinose	xylose	mannose	galactose	glucose	uronic acids	
			Alfalfa				
0	29.5	80.6	23.9	31.6	697.7	34.7	
6	24.1	99.9	15.7	24.4	611.0	22.8	
12	9.7	66.3	16.2	12.0	515.3	26.7	
18	16.3	80.6	24.2	9.4	411.0	26.3	
24	5.7	69.2	14.6	7.1	372.0	20.7	
36	4.1	60.6	10.1	5.9	279.7	13.6	
48	1.9	55.7	9.3	4.3	265.3	15.2	
72	2.2	53.2	5.1	2.4	229.5	13.9	
$\mathbf{SE}^{b}$	4.48*	17.68*	4.33*	2.59*	17.00*	4.10*	
			Orchard Grass				
0	42.0	152.0	7.3	35.0	681.7	41.7	
6	20.0	187.0	5.0	20.7	592.7	33.3	
12	15.3	197.0	1.0	14.0	473.0	28.3	
18	14.7	147.3		11.7	476.7	18.0	
24	10.9	161.7	1.7	10.3	374.3	17.2	
36	14.7	145.7		11.5	362.7	13.8	
48	9.7	108.0	1.3	9.3	234.0	10.9	
72	7.5	88.3	1.0	6.4	167.0	10.5	
$\mathbf{SE}^{b}$	6.07*	21.05*	2.05*	4.64*	33.91*	3.48*	

<sup>a</sup> Acid hydrolysate of neutral detergent fiber (NDF) residue expressed as milligrams/gram of NDF. <sup>b</sup> Standard error of means for 24 observations. Asterisk: P < 0.05.

at time zero. However, extent of NDF digestion was significantly higher for orchard grass than for alfalfa. Similar rates and extents of digestion of cell wall in alfalfa and orchard grass have been observed (Smith et al., 1971).

The alfalfa and orchard grass monosaccharide compositions of the NDF hydrolysate at various in vitro fermentation times are presented in Table III+ Monosaccharide composition of alfalfa was affected by fermentation time for arabinose (P < 0.0001), xylose (P < 0.015), mannose (P < 0.0001), galactose (P < 0.0001), glucose (P < 0.0001), and uronic acids (P < 0.0001). Orchard grass monosaccharide composition was affected by fermentation time for arabinose (P < 0.0001), xylose (P < 0.0001), mannose (P < 0.003), galactose (P < 0.0001), glucose (P < 0.0001), mannose (P < 0.003), galactose (P < 0.0001), glucose (P < 0.0001), mannose (P < 0.003), galactose (P < 0.0001), glucose (P < 0.0001), and uronic acids (P < 0.0001).

Changes in arabinose concentrations with time of fermentation had both linear (P < 0.0001) and quadratic (P < 0.0001)< 0.009, alfalfa; P < 0.0010, orchard grass) components. Coefficients of determination were 0.83 for alfalfa and 0.69 for orchard grass. There was no significant linear or quadratic effect for change in xylose concentration in alfalfa or orchard grass. Orchard grass mannose disappearance showed both linear (P < 0.002) and quadratic (P < 0.0017) trends with fermentation time. The change in mannose content of alfalfa had neither linear nor quadratic effects. Galactose content changed linearly (P < 0.0001)and quadratically (P < 0.0001, alfalfa; P < 0.004, orchard grass) with fermentation time. Coefficients of determination were 0.94 and 0.75 for alfalfa and orchard grass, respectively. Glucose changes were both linear (P <0.0001) and quadratic (P < 0.001) for alfalfa and orchard grass. Coefficients of determination were 0.99 for alfalfa and 0.96 for orchard grass. Uronic acid concentration showed significant linear (P < 0.03, alfalfa; P < 0.001,orchard grass) trends. Uronic acid concentration also exhibited a quadratic effect (P < 0.0001) for orchard grass, but alfalfa showed no quadratic effect. Coefficients of determination were 0.73 and 0.91 for alfalfa and orchard grass, respectively.

The ratios of xylose to arabinose and xylose to glucose for alfalfa and orchard grass at various in vitro fermentation times are shown in Table IV. The ratio of xylose to arabinose increased quadratically (P < 0.02) with time

Table IV. Xylose to Arabinose and Xylose to Glucose Ratios in Alfalfa and Orchard Grass at Various in Vitro Fermentation Times

	xylose	to arabinose	xylose to glucose		
time, h	alfalfa	orchard grass	alfalfa	orchard grass	
0	2.7	3.6	0.12	0.22	
6	4.0	9.4	0.16	0.32	
12	6.9	12.8	0.13	0.42	
18	4.9	10.1	0.20	0.31	
24	12.2	15.1	0.19	0.43	
36	15.2	10.0	0.22	0.40	
48	31.5	11.4	0.22	0.46	
72	35.0	11.8	0.23	0.54	
SEª	7.56*	2.81*	0.034*	0.060*	

 $^a$  Standard error of means for 24 observations. A sterisk: P < 0.05.

of fermentation for alfalfa. The xylose to arabinose ratio for orchard grass had significant linear (P < 0.004) and quadratic (P < 0.013) components. Coefficients of determination were 0.73 and 0.38 for alfalfa and orchard grass, respectively. Xylose to arabinose ratios are indicative of the degree of linearity or branching of hemicellulose (Bittner, 1983). A high xylose to arabinose ratio would indicate a high degree of polymerization with little bonding with other monosaccharide constituents and would be characteristic of feeds with low digestibility. A low xylose to arabinose ratio suggests a short-chain polymer with a large amount of branching with other monosaccharides and a high digestibility. The extent of orchard grass NDF digestion was greater than that for alfalfa (Table II). Alfalfa has lower total cell walls, lower hemicellulose and higher cell solubles, and higher lignin contents than does orchard grass (Smith et al., 1972). In an in vitro study done by Smith et al. (1972), the in vitro cell wall indigestibilities for alfalfa and orchard grass were 45% and 12%, respectively. Corresponding results from this experiment were 45.6% and 38.4% indigestibility for alfalfa and orchard grass, respectively. Even though alfalfa tends to be more digestible than does orchard grass, the cell wall portion of alfalfa is more indigestible. Because of the relative ease of hydrolysis of furanose linkages, arabinose side chains would be expected to be digested

Table V. Extent of in Vitro Disappearance of Alfalfa and Orchard Grass Monosaccharides at 72 h

	disappearance, %						
	arabinose	xylose	mannose	galactose	glucose	uronic acids	
alfalfa	89.3	33.0	79.6	92.4	67.3	59.9	
orchard grass	82.8	41.4	85.8	81.6	75.5	74.4	
$SE^a$	3.92	4.48	10.16	4.62	0.22*	1.07*	

 $^a$  Standard error of means for six observations. A sterisk: P < 0.05.

more readily than the  $\beta$ -1 $\rightarrow$ 4-linked xylan backbone (Bailey, 1973), and xylose to arabinose ratios would be expected to increase.

Xylose to glucose ratios changed with various in vitro fermentation times (Table IV). Glucose provides a measure of cellulose content, and xylose is indicative of hemicellulose content (Morris and Bacon, 1976). An increase in the xylose to glucose ratio would indicate a more rapid digestion of cellulose than hemicellulose. Xylose to glucose ratios were higher for alfalfa at 72 h than at 0 h (P < 0.001), but no significant linear or quadratic trends were evident. Orchard grass xylose to glucose ratios showed a significant linear trend (P < 0.006) with increasing fermentation time.

The extents of in vitro disappearance of alfalfa and orchard grass monosaccharides are found in Table V. There were no significant differences in arabinose, xylose, mannose, and galactose disappearance between alfalfa and orchard grass. However, uronic acids were less digestible in alfalfa than in orchard grass. The depressed digestibility of uronic acids in alfalfa would indicate a great amount of hemicellulose bonding with lignin. Uronic acids are proposed to be one of the primary monosaccharide constituents linking heteroxylans to phenolic components of lignin (Harkin, 1973). The glucose fraction of orchard grass was more digestible than that of alfalfa, indicating a higher amount of cellulose disappearance in orchard grass than in alfalfa.

Hemicellulose digestion was calculated by multiplying the fraction of arabinose, xylose, mannose, galactose, and uronic acid disappearance by their relative proportion as a component of hemicellulose and adding fractional disappearances. Cellulose disappearance was assumed to be equivalent to that for the glucose fraction of the monosaccharides. Digestibilities calculated for hemicellulose were 62.5% and 63.1% for alfalfa and orchard grass, respectively. Cellulose disappearances were 67.3% and 75.5% for alfalfa and orchard grass, respectively. Calculated digestibilities show a tendency toward lower disappearance of hemicellulose than cellulose. The increases in xylose to glucose ratios with fermentation time (Table IV) indicate that the hemicellulose component of both alfalfa and orchard grass is less digestible than the cellulose component. Assuming the lignin fraction was totally indigestible, the expected NDF disappearances based on calculated hemicellulose and cellulose disappearances for alfalfa and orchard grass were 55.8% and 64.2%, respectively. These figures are in close agreement with the extents of NDF digestion found in Table II (54.4% and

61.6% for alfalfa and orchard grass, respectively). The lower uronic acid and decreased glucose disappearances found in alfalfa appear to be primarily responsible for the decreased extent of cell wall disappearance found in alfalfa relative to orchard grass.

**Registry No.** Hemicellulose, 9034-32-6; D-xylose, 58-86-6; arabinose, 147-81-9; D-glucose, 50-99-7; cellulose, 9004-34-6; D-galactose, 59-23-4; D-mannose, 3458-28-4.

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