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Effect of Ozone and Sodium Hydroxide Treatments on the Composition and in Vitro Digestion of Screened Manure Fiber from Different Sources

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Screened manure fiber (SMF) was separated from manure of heifers fed complete diets consisting of a concentrate + either vetch hay (VH) or oat hay (OH) at 30 and 70% of the ration's dry matter (DM). The four SMF sources were subjected to sodium hydroxide and ozone treatments. Chemical treatment was the major factor affecting the neutral detergent fiber (NDF) content of SMF. Accordingly, three distinct groups of NDF values with no overlap were obtained: untreated > NaOH-treated > ozone-treated SMF. Sodium hydroxide had a slight solubilizing effect on the NDF of SMF (≥10%), whereas ozone was far more effective (~38%). Lignin content in SMF was in the range 17–22%. The effect of 6% NaOH on the content of lignin was in the direction of a slight increase; ozone, however, degraded up to two-thirds of the lignin of SMF. As with the NDF fraction, three distinct groups of hemicellulose values, distributed according to chemical treatments, were clearly noticeable: untreated > NaOH-treated > ozone-treated SMF. Ozone exerted the most effective solubilizing action upon the matrix polysaccharides. In vitro organic matter digestibility of untreated SMF was very low (12–20%). Ozone increased the IVOMD to the range 70–75%; the effect of 6% NaOH was more modest, upgrading the SMF to the range 36–53%.

Digestion studies at the monomer level have shown that cellulose and xylans are not equally digested by ruminants. More extensive digestion of the cellulose component was demonstrated in a series of experiments with different forages both in vitro and in vivo (Ben-Ghedalia and Miron, 1984; Ben-Ghedalia and Rubinstein, 1984, 1985; Morris and Bacon, 1977). Consequently, the proportion of hemicellulose in the residual CW is increased. The cell walls (CW) of cereal and leguminous forages, representing mono- and dicotyledonous sources, differ in terms of chemical, structural and biodegradational properties. For example, the ratio of lignin to matrix polysacchrides in the CW is approximately 1:5 in monocots as compared with 1:1 in dicots (Ben-Ghedalia and Rubinstein, 1984, 1985). The digestibility of the matrix polysaccharides is determined by the degree of branching of the xylan chains and by the extent to which they are interlinked with lignin (Brice and Morrison, 1982; Tanner and Morrison, 1983). Although it is understood that the digestive tract of the ruminant exerts a leveling effect with respect to the composition of the ingested CW, there is a question whether the original "fingerprint" of the forage plant CW could still be recognized in the fecal material. This is of particular interest when considering the response of screened manure fiber (SMF) of various sources to chemical treatments, since it was found that hemicellulose-rich lignocelluloses respond better to hydrolyzing or saponifying treatments whereas lignin-rich materials are more effectively treated by oxidative agents (Miron and Ben-Ghedalia, 1982; Shefet and Ben-Ghedalia, 1982).

Another question of interest is the effect of the level of forage intake (within a fixed total dry matter (DM) intake) on neutral detergent fiber (NDF) composition of SMF, the biodegradation of SMF by rumen microbes, and its response to chemical treatments. This is of particular interest, as it is well-known that increasing forage intake speeds the rate of passage of fiber from the rumen and increases the amount of undigested, but potentially digestible, fiber in the feces (Evans, 1981).

Table I. Composition (g/100 g of DM) of Hays Fed to the Heifers

component	VH	OH
NDF	59.4	71.7
cellulose	37.4	37.6
hemicellulose	9.45	28.2
lignin (permanganate)	12.5	5.73
NDF non-glucose polysacch	15.6	21.9
ND-sol carbohydr	27.1	13.1
in vitro OMD	53.0	55.5

The use of vibrating screeners for the separation of cattle manure into fiber and liquid presents a well-balanced solution to the ecological problem of manure disposal in the dairy herd, since the fiber can be refed and the liquid and fine particles recycled to the soil (Wilkinson, 1980). The expanding use of this technology motivated this research.

The objective of this study was to examine the response of SMF of different sources, originating from cereal or legume hay fed at different levels, to ozone and NaOH treatments in terms of chemical composition and biodegradation by rumen microbes.

MATERIALS AND METHODS

Animals, Feeds, and Separation of SMF. Sixteen Holstein heifers with an average weight of 360 kg each were allocated to four dietary treatments with four heifers per treatment. The rations (7.89 kg of DM/day) consisted on a DM basis of (I) 70% vetch hay (VH) + 30% concentrate, (II) 30% VH + 70% concentrate, (III) 70% oat hay (OH) + 30% concentrate, and (IV) 30% OH + 70% concentrate. The chemical composition and in vitro digestibility of the forages are given in Table I. The concentrate mixture was a 15% crude protein commercial concentrate, consisting per 100 kg of 37 kg of corn, 25 kg of wheat bran, 14 kg of soybean meal, 10 kg of barley, 10 kg of sorghum, and 4 kg of a mineral mixture. The heifers were fed the complete feed mixture (hay + concentrate) twice daily at 0800 and 1400 h. Following 14 days of adaptation, fecal grab samples were taken twice per day, 2 h postfeeding for 4 days. Fecal samples were pooled on a DM basis into one large sample for each treatment and kept at −20 °C until separation of the fiber fraction.

SMF was separated on a laboratory scale using a 2-mm net screen and repeated washings with tap water until complete clearing of nonfibrous material. The four SMF materials were

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Table II. Effect of Chemical Treatments on the Content (g/100 g of DM) of NDF Fractions in SMF Originating from Cattle Fed either VH or OH (Mean Values of Three Replicates per Treatment ± SE)

source of fecal fiber in diet	chemical treatment: control	NaOH	O_3
	NDF		
30% VH	87.4 ± 1.79	79.0 ± 1.66	57.3 ± 0.29
70% VH	86.0 ± 0.24	77.9 ± 0.18	54.3 ± 0.31
30% OH	88.7 ± 0.98	81.2 ± 0.49	55.1 ± 0.78
70% OH	84.9 ± 0.68	76.9 ± 0.56	53.5 ± 1.18
	Lignin		
30% VH	22.3 ± 0.27	24.6 ± 0.90	9.93 ± 0.32
70% VH	19.6 ± 0.17	22.4 ± 0.58	7.17 ± 0.45
30% OH	16.6 ± 0.83	17.5 ± 0.40	5.53 ± 0.09
70% OH	17.2 ± 0.34	18.2 ± 0.26	7.50 ± 0.25
	Hemicellulose		
30% VH	28.4 ± 0.61	17.4 ± 0.42	12.6 ± 0.84
70% VH	32.3 ± 0.93	17.9 ± 0.82	14.6 ± 1.25
30% OH	33.2 ± 0.75	19.7 ± 0.89	13.4 ± 1.11
70% OH	32.5 ± 1.07	21.9 ± 0.78	12.3 ± 1.67
	Cellulose		
30% VH	35.4 ± 0.13	36.0 ± 0.25	32.6 ± 0.56
70% VH	32.7 ± 1.13	35.4 ± 0.30	33.2 ± 0.49
30% OH	36.7 ± 0.03	39.8 ± 0.64	35.4 ± 0.24
70% OH	33.3 ± 1.00	34.5 ± 1.02	32.0 ± 0.36

dried in an oven at 50 °C and kept in the laboratory until subjected to the chemical treatments.

Chemical Treatments. Each of the four SMF materials was divided into three subsamples to be used for control (untreated SMF), NaOH treatment, and ozone treatment. Chemical treatments were performed in triplicate. Each of the control SMF materials was subdivided into three replicates, as done with the SMF subjected to the chemical treatments. The NaOH treatment was done by adding 100 mL of 6% NaOH to 100 g of SMF-DM. The NaOH-treated SMF samples were kept at room temperature for 1 week, after which the materials were freeze-dried and used for analyses. For the ozone treatment, batches of 50 g of SMF-DM were moistened to 40% and ozonized in glass columns (80 × 5 cm) until complete decolorization. For this purpose a Fischer ozone generator, Model 501, was used, supplying 2 g of ozone/h. Under these conditions, ozone consumption was 1 g/5 g of DM of treated SMF.

Analytical Procedures. The detergent fractionation system of Goering and Van Soest (1970) was used to determine and prepare the neutral detergent fiber, acid detergent fiber (ADF), and permanganate lignin. Hemicellulose was calculated by difference (NDF − ADF).

In vitro digestibility (IVD) of the organic matter and monosaccharide residues was measured in triplicate by using the rumen liquor–acid pepsin method of Tilley and Terry (1963). Monosaccharides in whole SMF material and in NDF preparations were determined in ball-milled material after hydrolysis in 12 M H_2SO_4 for 1 h at room temperature and continued hydrolysis in 0.5 M H_2SO_4 at 100 °C for 5 h. Inositol was added as an internal standard, and the released monosaccharides were determined as their alditol acetate derivatives after reduction and acetylation according to Blakeney et al. (1983). The conditions for separation by gas–liquid chromatography were those of Bacon and Gordon (1980). Uronic acids in hydrolysates were determined colorimetrically according to Blumenkrantz and Asboe-Hansen (1973).

Calculations. Monosaccharides were determined in whole SMF and in NDF preparations, and the fraction of soluble monosaccharides was derived by difference. For any of the parameters examined, the effect of treatments was the predominant factor affecting the variation within SMF samples. Therefore, standard error (SE) was calculated to show the variation within the replicates of the chemically treated SMF samples.

RESULTS

The effect of treatments on the content of NDF and NDF fractions in SMF is shown in Table II. There was only a slight variation in NDF content within the untreated SMF sources. Three distinct groups of NDF values, distributed according to chemical treatments with no overlap, are clearly noticeable, implying that the major factor affecting the variation of NDF content in the SMF sources of this study was the effect of chemical treatments. The NaOH treatment had a slight solubilizing effect (10% or less), whereas ozone was far more effective in this regard (~38%). Lignin content of untreated SMF was somewhat higher in materials originating from vetch than in those originating from oat hay; however, the between-feeds difference in lignin content (see Table I) has been leveled off. The effect, if any, of NaOH on the content of lignin was in the direction of a slight increase. Notwithstanding, ozone degraded up to two-thirds of the lignin of SMF, almost equaling its content to that of the forages ingested. (For comparison, see Table I.)

As with the NDF fraction, three distinct groups of hemicellulose values, distributed according to chemical treatments, are clearly noticeable. Considering the hemicellulose content, it seems that the limited efect of the NaOH treatment on NDF content of SMF was mediated via its solubilizing action on the hemicellulose fraction. Ozone was more effective than NaOH in solubilizing hemicellulose. Cellulose content of SMF sources was not affected by chemical treatments. Moreover, the variation in cellulose content within and between chemical treatments was small.

The effect of treatments on the composition of monosaccharide residues in whole SMF materials is shown in Table III, and their distribution between NDF and cell solubles is presented in Table IV. Neither ozone nor NaOH had any appreciable effect on monosaccharide composition. Moreover, between-forages differences in carbohydrate composition (Table I) were almost completely eliminated in SMF as a result of the digestive processes in the gut. Whereas chemical treatments did not affect overall carbohydrate composition of SMF, they did affect the distribution of monosaccharides in NDF and cell solubles fractions. Generally, glucose was NDF-bound and hardly affected by treatments. The effect of chemical treatments was expressed mainly on the xylan and the related matrix (hemicellulose) monosaccharides. Ozone exerted the most effective solubilizing action on those

Table III. Effect of Chemical Treatments on the Composition (g/100 g of DM) of Monosaccharide Residues[a] of Screened Manure Fiber Originating from Cattle Fed either VH or OH

monosacch residue	30% VH in the diet			70% VH in the diet			30% OH in the diet			70% OH in the diet		
	control	NaOH	O_3	control	NaOH	O_3	control	NaOH	O_3	control	NaOH	O_3
glucose	30.9	31.4	30.2	33.4	29.7	32.7	32.1	32.7	34.9	33.9	31.0	31.8
xylose	16.8	16.0	15.8	15.4	14.0	16.0	18.4	17.8	17.5	20.0	16.8	17.3
arabinose	5.58	5.47	4.91	2.61	2.70	3.30	5.23	5.94	5.29	3.13	3.70	3.52
galactose	1.92	1.58	1.86	1.15	1.45	2.05	1.60	1.60	1.06	1.35	1.32	1.40
mannose	1.02	0.98	0.99	0.85	1.03	1.32	0.80	0.93	0.50	0.79	0.70	1.03
rhamnose	0.26	0.24	0.27	0.40	0.27	0.33	0.19	0.13	0.21	0.16	0.11	0.31
uronic acids	5.56	5.25	5.27	6.31	5.93	5.24	5.02	4.98	4.34	4.44	4.47	5.53

[a] Released by hydrolysis; see Materials and Methods.

Table IV. Effect of Chemical Treatments on the Distribution (%) of Monosaccharide Residues[a] in NDF and in Cell Solubles (CS) of Screened Manure Fiber

		30% VH in the diet			70% VH in the diet			30% OH in the diet			70% OH in the diet		
		control	NaOH	O_3	control	NaOH	O_3	control	NaOH	O_3	control	NaOH	O_3
glucose	NDF	100	100	97.6	96.7	94.2	100	100	93.2	96.2	100	100	100
	CS			2.36	3.30	5.80			6.80	3.80			
xylose	NDF	96.2	71.9	49.5	98.1	78.0	58.2	100	89.6	51.7	98.0	100	52.8
	CS	3.80	28.1	50.5	1.90	22.0	41.8		10.4	48.3	2.00		47.2
arabinose	NDF	94.0	74.8	21.0	94.6	60.0	27.0	100	100	26.6	100	83.4	25.3
	CS	6.00	25.2	79.0	5.40	40.0	73.0			73.4		16.6	74.7
galactose	NDF	57.2	48.5	19.6	64.8	31.2	18.0	53.0	51.5	50.2	39.4	33.0	13.0
	CS	42.8	51.5	80.4	35.2	68.8	82.0	48.0	48.5	49.5	60.6	67.0	87.0
uronic acids	NDF	74.7	83.4	43.5	78.4	74.0	40.6	82.2	99.6	43.3	100	71.2	45.2
	CS	25.3	16.6	56.5	21.6	26.0	59.4	17.8	0.40	56.7		29.8	54.8

[a] Released by hydrolysis; see Materials and Methods.

Table V. Effect of Chemical Treatments on the in Vitro Organic Matter Digestibility (%) of SMF Originating from Cattle Fed either VH or OH (Mean Values ± SE)

source of fecal fiber in diet	control	NaOH	O_3
30% VH	11.5 ± 0.29	36.3 ± 0.61	70.0 ± 1.16
70% VH	17.7 ± 0.97	46.5 ± 0.42	74.9 ± 0.45
30% OH	11.8 ± 1.01	51.5 ± 0.06	70.7 ± 0.62
70% OH	20.5 ± 0.87	53.2 ± 0.26	75.1 ± 1.07

polysaccharides. It is interesting to note that, in the untreated SMF samples (controls, Table IV), glucose, xylose, and arabinose—the typical NDF components—were virtually NDF-bound, whereas galactose and uronic acids, which may be found in microorganisms and in pectin, were present also in the cell solubles fraction. In vitro digestibility values of the organic matter (IVOMD) are presented in Table V and those of the monosaccharide components of whole SMF in Figure 1. In vitro organic matter digestibility (OMD) values of untreated SMF were very low, but the effect of treatments was remarkable and the difference between treatments was clearly distinguishable. The ozone treatment increased the IVOMD up to the range 70–75%. The effect of 6% NaOH was more modest, upgrading the SMF to the level of a low-quality forage (36–53%). It appears that, among the untreated SMF materials, those originating from the high-forage treatments (70% hay) were more digestible than those from the low-forage treatments (30% hay). It seems also that the oat hay originating SMF samples responded better to the NaOH treatment than those originating from vetch hay.

The increase in digestibility of the monosaccharide components of whole SMF as affected by treatments was higher than that found with the OM. The IVD of the individual monosaccharides was raised to the level of 80–90% in the ozone-treated materials. The effect of sodium hydroxide on the digestibility of glucose and xylose, the major monosaccharides, was weaker, but the effect of ozone and of NaOH on the IVD of arabinose was similar.

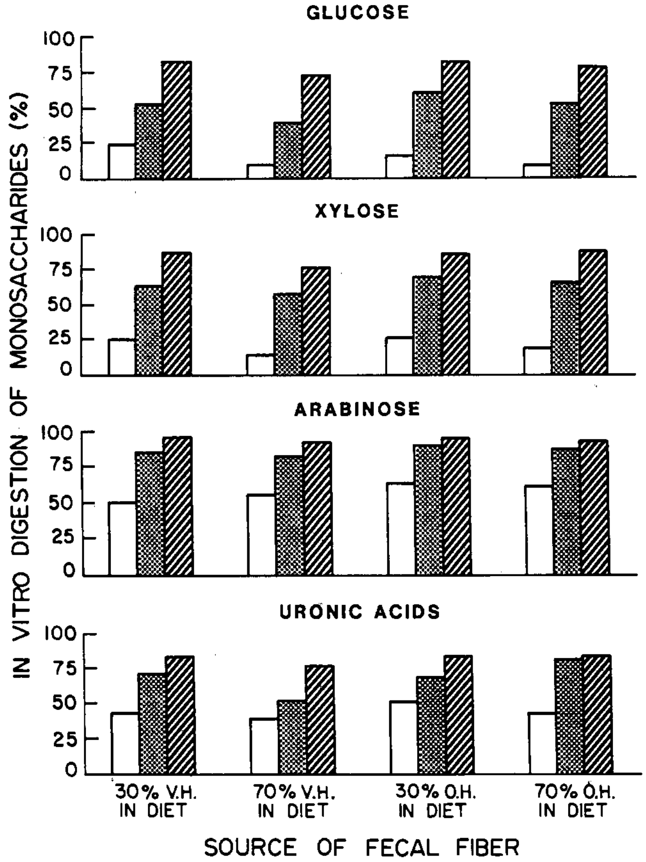

Figure 1. In vitro digestibility of monosaccharides released by hydrolysis in four sources of chemically treated SMF: □, control; ▥, 6% NaOH-treated; ▨, ozone-treated.

DISCUSSION

The question whether SMF could be characterized chemically according to its origin, i.e., the type of forage ingested, was raised in this study. Tables II and III provide

Table VI. In Vitro Digestibility (%)[a] of Monosaccharides in OH and VH[b]

	in whole material		in cell walls	
monosacch residue[c]	OH	VH	OH	VH
glucose	65.7 ± 1.01	58.7 ± 0.88	61.0 ± 1.15	55.3 ± 0.95
xylose	52.2 ± 1.90	42.4 ± 2.19	51.5 ± 1.95	34.3 ± 2.52
arabinose	61.1 ± 0.21	74.8 ± 1.30	58.6 ± 0.28	61.8 ± 1.97
galactose	75.3 ± 0.14	84.0 ± 0.18	49.7 ± 0.25	58.8 ± 0.45
mannose	89.8 ± 0.19	76.7 ± 0.33	57.7 ± 0.09	63.5 ± 0.52
rhamnose	61.3 ± 3.26	70.0 ± 0.83		46.7 ± 1.46
uronic acids	75.0 ± 0.25	83.2 ± 1.10	62.7 ± 0.50	42.6 ± 3.70
total monosacch	62.8 ± 1.06	64.4 ± 0.78	57.3 ± 1.22	51.2 ± 1.07
IVOMD	57.4 ± 0.16	57.6 ± 0.03		

[a] Mean values ± SE based on three in vitro replicates. [b] From Ben-Ghedalia and Rubinstein (1984). [c] Released by hydrolysis; see Materials and Methods.

an answer to this question: The digestive tract exerts a remarkable leveling effect on the composition of the ingested forages. Differences in composition between oat and vetch hay (Table I) are almost completely eliminated and cannot be recognized in the corresponding SMF samples. However, it appears that the original nature of the chemical bonds within the CW macromolecule is still preserved to some extent in the SMF materials. This is evident from the differential response of oat-SMF and vetch-SMF to NaOH treatment with respect to IVOMD (Table V).

It is clear from this study that the predominant effect on SMF composition and digestibility was that of the chemical treatments. Irrespective of SMF source, ozone was the most effective treatment in terms of both NDF solubilization and of increasing the IVOMD (Table V; Figure 1). These phenomena correspond well with previous data on SMF (Ben-Ghedalia and Rubinstein, 1986) and with other lignocelluloses showing that high-lignin (dicot) plant residues are sensitive to oxidizing (ozone) treatments (Shefet and Ben-Ghedalia, 1982). SMF, irrespective of source, can be considered as high-lignin materials. To illustrate the magnitude of the treatments effect, some data on the IVD of monosaccharide residues of oat and vetch hays are presented in Table VI. Ozone increased the in vitro digestibility of total glucose and total xylose of SMF to a level (80–90%) much higher than found in the hays (Table VI; Figure 1). The effect of NaOH in this respect was within the range of, or somewhat higher than, the respective values of the forages. The significance of this finding is that the chemical treatments applied to SMF were able to convert the later into a foragelike material.

In summary, untreated SMF is of little value as a feed alternative irrespective of origin. The variation in its composition is not large as compared with conventional forages (Ben-Ghedalia and Rubinstein, 1986). Treated SMF from cattle seems to be suitable for recyling through ruminants since it contains a substantial fraction of potentially digestible carbohydrates. Sodium hydroxide treatment, which proved to be particularly effective with SMF of monocot origin, is suggested as a starting point, since the technology is simple and available. NaOH-treated SMF is suggested as a feed alternative for growing heifers and other low-producing ruminants. Ozone-treated SMF can be considered as a potential component in the rations of high-producing ruminants. This is, however, closely associated with developing the ozonation technology. Further experiments, in vivo, are necessary to substantiate the results of this study prior to application in the field.

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Registry No. NaOH, 1310-73-2; O_3, 10028-15-6; glucose, 50-99-7; xylose, 58-86-6; arabinose, 147-81-9; galactose, 59-23-4; mannose, 3458-28-4; rhamnose, 3615-41-6; lignin, 9005-53-2; hemicellulose, 9034-32-6.

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