

Varying the dietary supply of C and N to manipulate the manure composition of water buffalo heifers in Oman

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Abstract

Optimizing the composition of manure has the potential to reduce nutrient losses to the environment and to increase crop yields. In this study the effect of dietary ratios of carbon (C) to nitrogen (N) and neutral detergent fibre (NDF) to soluble carbohydrates (SC) on faeces composition of water buffalo heifers was assessed. Two digestibility trials were conducted with 12 animals each, fed one control and four test diets composed to achieve (1) high C/N and high NDF/SC ratios (HH), (2) low C/N and low NDF/SC ratios (LL), (3) high C/N and low NDF/SC ratios (HL) and (4) low C/N and high NDF/SC (LH) ratios. Faecal C/N ratios were generally lower than dietary C/N ratios, but the reduction was especially large for high C/N ratio diets (HH=55 %, HL=51 %). Faecal N concentration was positively correlated ($r^2 = 0.6$; $P < 0.001$) with N intake, but the increase in faecal N was more pronounced for diets that supplied low amounts of N. Faecal NDF concentration was positively related to NDF intake ($r^2 = 0.42$; $P < 0.001$), as well as the faecal C/N ratio ($r^2 = 0.3$; $P < 0.001$). Results demonstrate that C/N ratio and NDF concentration of buffalo manure were affected by diet composition. Diets with high C/N ratio and low NDF/SC ratio are preferable with regard to manure quality, but may not satisfy the nutritional requirements of producing animals, since N concentration in these diets was low and fibre concentration simultaneously high.

Keywords: *Bubalus bubalis*, dietary manipulation, manure composition, dietary C, dietary N, Oman

1 Introduction

In smallholder production systems of tropical regions, ruminant species like sheep, goats and buffaloes are often of high economic importance. The worldwide population of buffaloes (*Bubalus bubalis*) increased from 148 million head in 1990 to 194 million head in 2012, especially in India and Pakistan but also in the Near East. While the meat production from buffalo increased only slightly, milk production was largely enhanced from 44 million tones to 93 million tones in the same time period (FAOSTAT, 2012). Besides the demand for the

primary products milk and meat, the dung production of buffaloes becomes increasingly interesting as source of organic fertilizer in crop production, particularly for smallholder farmers in the Tropics.

The quality of organic fertilizers is mainly determined by their nitrogen (N) and carbon (C) content, usually expressed by the C-to-N ratio (C/N ratio). However, decomposition of organic fertilizers is additionally influenced by the forms in which carbon is available, i.e. rapidly available soluble carbohydrates or slowly degrading cell wall components like undigested NDF and lignin (Heal *et al.*, 1997). Maintaining sufficient levels of soil organic matter is crucial especially in tropical environments because of the rapid nutrient turnover in soils. Long-term effects of organic fertilization on soil OM and organic N accumulation in the soil are mainly determined by the C form (Hassink, 1994). Correspondingly, considerable amounts of C should be supplied in

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form of slowly decomposable carbohydrates and lignin to facilitate long-term nutrient release and stabilization of soil OM (Handayanto *et al.*, 1997).

Results obtained from studies with cattle or other ruminants cannot be transferred directly to buffaloes, because differences in the digestive processes of cattle and buffalo have been widely observed. It is accepted that the buffalo is superior to cattle in digesting poor-quality feeds (Robles *et al.*, 1971; Devendra, 1992; Van Soest, 1994). Nevertheless, contradictory results are reported concerning the physiological reasons. Several authors obtained higher fibre digestibility in buffaloes compared to cattle (Grant *et al.*, 1974; Hussain & Cheeke, 1996), while other studies showed that fibre digestibility was higher in cattle (Kennedy *et al.*, 1992; Puppo *et al.*, 2002). Less diverse results were obtained for the crude protein (CP) digestibility, as most authors reported similar CP digestibility in buffaloes and cattle (Sebastian *et al.*, 1970; Devendra, 1992; Puppo *et al.*, 2002). However, differences in N metabolism were observed between cattle and buffaloes, indicating that buffaloes are able to use ingested N more efficiently. Higher blood-urea contents (Kennedy *et al.*, 1987) and ammonia concentrations in the rumen (Kennedy *et al.*, 1992) were observed, resulting in lower urinary N excretion and a higher N retention (Devendra, 1992).

The conversion of plant biomass to manure by live-stock digestion processes changes the nutrient content and composition of the ingested material (Powell *et al.*, 1999; Van Vliet *et al.*, 2007). Several studies have examined the effects of dietary manipulation on manure and slurry composition and the subsequent mineralization processes in the soil and therewith the fertilizer quality, mainly defined by N concentration and C/N ratio. Most important impacts on manure or slurry quality were related to the intake of N and fibre (Kyvs-gaard *et al.*, 2000; Sørensen *et al.*, 2003; Reijs *et al.*, 2007). Studies investigating how the composition of buffalo manure is influenced by digestion processes and dietary manipulation are rare. Kennedy (1995) claimed that the buffalo's higher digestive efficiency may reduce the chances to influence the faeces composition through dietary manipulation. Challenging this perception we hypothesize that despite the buffalo's seemingly higher digestive efficiency, marked differences in C/N ratio and carbohydrate composition in the diet will persist and be reflected in faeces composition. The objectives of the present study were therefore (i) to clarify the influence of variable C/N ratio and carbohydrate composition in water buffalo diets on faeces composition, and (ii) to determine which faecal components are susceptible to changes in dietary composition by measuring the digestibility of dietary components.

2 Material and Methods

2.1 Experimental site

Two feeding trials, each designed as 3×3 Latin square with experimental periods of 28 days, were carried out on a private farm in Sohar, Oman, located 220 km North of Oman's capital Muscat. The first trial took place during September – November 2007 and the second trial during February – May 2008. Oman is located in the arid tropics with a mean annual precipitation of 102 mm in Sohar. During the first trial period the average daily temperature was 31 °C and the relative humidity 60 %, while average daily temperature was 28 °C and relative humidity 52 % during the second trial period. In both trial periods no rainfall occurred.

2.2 Animals

In each of the two trials, twelve Nili-Ravi buffalo heifers were selected randomly from a herd of more than 60 heifers. The animals were 1.7 – 2.0 years old and weighed 385 – 600 kg at the start of the first and second trial, respectively. Their live weight development during the trials was determined on a mechanical scale at days 1, 11, 21 and 28 of each trial period (Table 3). The animals were individually tethered under roofed paddocks with open sides and fed from large-size troughs.

2.3 Treatments and experimental design

In each of the two trials, two experimental diets were compared to one control diet. As control diet (CD), the habitual diet of the buffaloes on the private farm was retained consisting of hay, dates and a commercial concentrate. For the experimental diets the ingredients were combined in such a way that the C/N ratio and the ratio of NDF to soluble carbohydrates (sugars and starch, SC; NDF/SC) was either high (H) or low (L), resulting in four treatments, namely HH and LL (tested in trial 1) as well as HL and LH (tested in trial 2). With the exception of hay, as the forage component, the different feed-stuffs were mixed to achieve the requested composition in the different treatments and offered as concentrate mixtures. The chemical composition of the different diets is presented in Table 2. All diets contained Rhodes grass (*Chloris gayana*) hay and varying proportions of soybean (*Glycine max*) meal, crushed maize (*Zea mays*) grain, whole dates (fruits of *Phoenix dactylifera*), wheat (*Triticum* spp.) bran, cotton seed (*Gossypium hirsutum*) meal and a commercial concentrate feed (for details see Table 1).

Each trial was set up as a quadruplicate 3×3 Latin square design, with a control diet and two experimental diets. Based on their tabulated concentration (Deutsche

Table 1: Composition of the experimental diets (g/kg DM) offered to water buffalo in two feeding trials conducted in Oman.

Feedstuff	Diet				
	Trial 1 (autumn 2007)		Trial 2 (spring 2008)		Trials 1,2 CD
	HH	LL	HL	LH	
Rhodes grass hay	899	401	612	681	677
Soybean meal	78	392	27	259	
Maize		207	361		
Dates					148
Wheat bran	23				
Cotton seed meal				60	
Concentrate mix					175

HH, high C/N and high NDF/SC ratios; LL, low C/N and low NDF/SC ratios;
HL, high C/N and low NDF/SC ratios; LH, low C/N and high NDF/SC ratios; CD, control diet.

Table 2: Chemical composition of experimental diets fed to water buffalo in two feeding trials conducted in Oman.

Component		Diet				
		Trial 1		Trial 2		Trials 1, 2 CD
		HH	LL	HL	LH	
OM	(g/kg DM)	939.4	942.4	947.5	928.1	933.3
N	(g/kg OM)	15.4	36.6	17.6	29.5	14.0
C	"	419.9	403.4	414.5	408.8	403.4
NDF	"	705.4	366.8	501.2	550.7	541.9
ADF	"	368.8	192.4	259.3	307.3	289.7
ADL	"	32.4	16.2	21.2	32.1	28.6
Starch	"	12.2	122.6	197.2	60.0	66.9
Sugar	"	43.9	178.2	30.2	21.9	98.7
C/N	(g/g)	27.2	11.0	23.5	13.9	28.8
NDF/SC	"	12.6	1.2	2.2	6.7	3.3

HH, high C/N and high NDF/SC ratios; LL, low C/N and low NDF/SC ratios;
HL, high C/N and low NDF/SC ratios; LH, low C/N and high NDF/SC ratios; CD, control diet.
OM, organic matter; N, nitrogen; C, carbon; NDF, neutral detergent fiber; ADF, acid detergent fiber; ADL, acid detergent lignin.

Landwirtschaftsgesellschaft (DLG), 1997) of metabolizable energy (ME), all diets were offered at 1.45 times maintenance energy requirements of the animals. Water was offered ad libitum. Of the 28 days per trial period, 21 days served the animals' adaptation to the diet, followed by 7 days of quantification of feed intake and faecal excretion.

The total amount of feed to be given was divided into two equal portions and offered daily at 7:00h and 14:30h. At each meal, the mixed concentrates were

offered in the first place; after one hour, eventual refusals were collected and then hay was offered. Refusals were weighed immediately before the start of the second daily meal. Representative samples of feed offered and the total of refusals (separated according to feed components) were collected daily during the quantification period, weighed (electronic balance, range 0–30 kg, accuracy 0.1 kg) and dried to constant weight at ambient temperatures, weighed again and ground in a mill with a 1 mm sieve before analysis.

During the 7 days of collection, faeces were quantitatively gathered into a plastic bowl whenever defecation occurred, whereby care was taken that faeces did not fall to the ground. If this occurred, any adhering dust was brushed away carefully before the excretion was weighed (electronic balance, range 0–30 kg, accuracy 0.1 kg). After homogenization of the excreted material, a representative sample of approximately 30 g fresh matter was taken and stored at -18°C until analysis. In this way, three to four faecal samples per animal and day were obtained, of which one (afternoon sample) was air dried and treated in the same way as described for the feeds.

2.4 Chemical analyses

Before analysis, samples were pooled as follows: For feed offered, one pooled sample was constituted per type of feed (concentrate, dates or hay) and 7-day sampling period; the same was done for the different types of residue samples for each individual animal. Likewise, the seven dried faecal samples per animal were pooled into one composite sample. Following standard procedures (Naumann & Bassler, 1976), all dried samples were analysed for concentrations of dry matter (DM) and OM. Concentrations of NDF, acid detergent fibre (ADF) and acid detergent lignin (ADL) were measured according to the methods described by Naumann & Bassler (1997). For the analysis of NDF and ADF a semi-automated Ankom 220 Fiber Analyzer (ANKOM Technology, Macedon, NY, USA) was used, whereby no decalin or sodium sulphite was used for NDF analysis. NDF, ADF and ADL values are expressed without residual ash. The determination of the concentration of sugar and starch followed the LUFF-SCHOORL-method and the polarimetric EWERS-method, respectively (Naumann & Bassler, 1976). C and N concentrations were analysed using a C/N-TCD analyser (Elementar Analysensysteme GmbH, Hanau, Germany) where 200 mg of sample material is combusted at 800°C and the concentration of gaseous C and N in the helium carrier gas is measured. Faecal samples used for the determination of C and N were morning samples of frozen faeces collected on days 2, 4 and 6. Samples were crushed, dried to constant weight at 60°C in a fan-assisted oven and ground in a mill with a 1 mm sieve before analysis.

2.5 Statistical analysis

To account for seasonal differences between the two trials the control diet was included into both trials. A pre-test showed that the data obtained from the control diets were different between the two trials, so that these were analysed individually by analysis of variance (ANOVA). Apart from the control for experimental period, the data of the control diet were not of interest

for in-depth analysis and discussion in the framework of this study.

The ANOVA was performed using the GLM procedure of SAS[®] 9.2 (SAS Institute Inc., Cary, NC, USA). Dependent variables (y) were analysed for the influence of experimental diets (D : control and two test diets) and the effect of squares which were defined as blocks. Each square (block) consisted of a 3×3 Latin square design; since twelve buffaloes were used four squares per trial were defined; animals were grouped by weight and groups of three animals were assigned to each square. The following model was used:

$$y_{ij(k)m} = \mu + S Q_m + \text{per}(S Q)_{im} + \text{buf}(S Q)_{jm} + D_{(k)} + \varepsilon_{ij(k)m} \\ (i, j, k = 1, \dots, r; m = 1, \dots, b)$$

where $y_{ij(k)m}$ = observation $ij(k)m$, μ = overall mean, $S Q_m$ = effect of square m , $\text{per}(S Q)_{im}$ = effect of period i within square m , $\text{buf}(S Q)_{jm}$ = effect of buffalo j within square m ; $D_{(k)}$ = effect of diet k ; $\varepsilon_{ij(k)m}$ = random error, r = number of diets, periods and buffaloes within each square, b = the number of squares.

Means with significant F-values were tested by the Tukey-Test. The level of significance was chosen to be $P < 0.05$. For simple linear regression analysis the REG procedure of SAS[®] 9.2 was used.

3 Results

3.1 Feed intake and faecal output

Mean values of live weight (LW, kg) of the buffaloes were not different between the experimental treatments HH and LL in trial 1 and between HL and LH in trial 2 (Table 3). The mean LW over the experimental treatments and all periods was about 83 kg higher in trial 2 than in trial 1, resulting in a higher mean metabolic weight (MW) of $112.7 \text{ kg}^{0.75}$ in the second compared to $99.5 \text{ kg}^{0.75}$ in the first trial. The organic matter intake (OMI) and the organic matter faecal output (OMFO) were different between the experimental treatments in both trials. In trial 1, OMI was 68.1 and $60.4 \text{ g/kg}^{0.75}$ in treatments HH and LL, but only 52.5 and $50.8 \text{ g/kg}^{0.75}$ in treatments HL and LH in trial 2, displaying an approximately 20 % lower OMI in trial 2. The OMFO in treatments HH and LL was 16.5 and 14.2 % of the OMI. In trial 2, OMFO was 15.9 and 11.6 % of the OMI in experimental treatments HL and LH, respectively.

3.2 Chemical composition of faeces

N concentration in faecal OM of treatments HH (2.4 %) and LL (4.0 %) in trial 1 differed significantly (Table 4), while this was not the case for the N concentration of the intermediate treatments HL (3.1 %) and

Table 3: Mean live weight (LW), mean daily organic matter intake and mean daily OM faecal output obtained from four experimental diets (HH, LL, HL, LH) and one control diet (CD) fed to water buffalo in two feeding trials conducted in Oman.

Diet	LW (kg)	Intake (g OM/kg ^{0.75} /d)	Faecal Output (g OM/kg ^{0.75} /d)
<i>Trial 1</i>			
HH	463.8	68.1	11.3
LL	459.8	60.4	8.6
CD	466.0	67.4	10.7
SE	1.72	0.85	0.47
<i>P-values</i>			
sqa	<0.001	0.022	0.210
per(sqa)	0.005	0.370	0.012
buf(sqa)	<0.001	0.110	0.503
trt	0.064	<0.001	0.003
<i>Trial 2</i>			
HL	542.5	52.5	8.3
LH	547.3	50.8	5.9
CD	554.0	52.8	7.2
SE	2.97	0.28	0.30
<i>P-values</i>			
sqa	<0.001	0.493	0.031
per(sqa)	0.001	0.514	0.379
buf(sqa)	<0.001	0.109	0.293
trt	0.050	<0.001	<0.001

HH, high C/N and high NDF/SC ratios; LL, low C/N and low NDF/SC ratios; HL, high C/N and low NDF/SC ratios; LH, low C/N and high NDF/SC ratios; CD, control diet. SE, standard error; sqa, square; per, period; buf, buffalo; trt, treatment.

LH (3.0%) in trial 2. On the other hand, the C concentration was not different between treatments HH and LL (29.4 and 27.8%) in trial 1, but was different between the experimental treatments HL and LH (32.3 and 30.7%) in trial 2. The NDF and ADF concentrations were highest in treatment HH (76.9 and 53.9%) and lowest in treatment LL (63.5 and 39.9%), while the treatments HL and LH of trial 2 showed intermediate concentrations. The ADL concentration was highest in treatment LH (23.5%), followed by HH (19.1%). Lower levels were determined analogically for treatments LL and HL. Significant differences were observed between HH and LL in trial 1, as well as between HL and LH in trial 2, for NDF, ADF and ADL concentrations. The concentration of starch in faeces also differed between the experimental treatments in both trials, being as low as zero in treatment LH and as high

as 10.8% in treatment HL, with intermediate concentrations in treatments HH and LL in trial 1. At an average value of 0.4%, the sugar concentration in faeces was similar in treatments HH and LL in trial 1, while it differed significantly between treatments HL (1.2%) and LH (0.2%). The faecal C/N ratio discerned between the experimental treatments in trial 1, but not in trial 2. The treatments HH and LL showed a C/N ratio of 12.2 and 7.0, while treatments HL and LH had a similar C/N ratio of 11.5 and 10.6, respectively.

3.3 Intake, faecal excretion and digestibility of nutrients

The daily mean nutrient intake and daily mean nutrient faecal output were related to MW to balance for differences in LW between the two trials. The results of nutrient intake of trial 1 and trial 2 are presented in Table 5, the results of nutrient faecal output are shown in Table 6. The N intake (2.2 g/kg^{0.75}/d) as well as the faecal N output (0.34 g/kg^{0.75}/d) was highest in treatment LL. For the other treatments this relationship was not apparent. In both trials, the experimental treatments were different in N intake, but for the faecal N output, differences were only observed between treatments HH and LL. For the treatments HH and HL, which both were high in their dietary C/N ratio, a lower N digestibility was observed in comparison to treatments LL and LH (Table 7). With respect to C, the treatment with the highest intake (HH) showed the highest faecal C output, but also in this case, this relationship did not manifest in the other treatments. Nevertheless, in both trials the experimental treatments were different from each other, both for C intake and C faecal output. The intake and faecal output of NDF was highest in treatment HH (48 and 8.8 g/kg^{0.75}/d), while the treatment LL with the lowest NDF intake showed the second highest NDF output (22.5 and 5.4 g/kg^{0.75}/d). The intermediate diets HL and LH had a lower faecal NDF output, even though NDF intake was higher than in LL. The same observation was made with respect to the ADF and ADL intake and faecal output. In both trials, NDF and ADF intake as well as faecal output differed significantly between the experimental treatments, whereas differences in ADL intake were significantly different only in trial 1, while no differences between treatments were determined for faecal ADL output. The NDF digestibility was lower in the treatments LL (78%) and HL (79%) in comparison to HH (82%) and LH (85%), probably due to the low NDF/SC ratio. The starch intake and faecal output were highest for the HL treatment (10.8 and 0.95 g/kg^{0.75}/d), followed by the LL treatment (7.3 and 0.26 g/kg^{0.75}/d). The quantitative intake and excretion of starch were not related in treatments HH (0.84 and 0.04 g/kg^{0.75}/d) and LH (2.97 and 0.0 g/kg^{0.75}/d). Only very small concentra-

Table 4: Chemical composition of faeces (g/kg OM) obtained from four experimental diets (HH, LL, HL, LH) and one control diet (CD) fed to water buffalo in two feeding trials conducted in Oman.

Diet	N	C	NDF	ADF	ADL	Starch	Sugar	C/N ratio
<i>Trial 1</i>								
HH	24.1	294.0	768.9	538.6	191.1	3.4	3.5	12.2
LL	39.8	278.3	635.7	398.7	156.0	29.8	4.6	7.0
CD	28.0	307.1	717.1	513.3	198.4	6.3	2.5	11.0
SE	0.53	8.48	22.59	15.94	7.43	2.36	0.74	0.30
<i>P-values</i>								
sqa	0.890	0.627	0.315	0.570	0.903	0.534	0.271	0.523
per(sqa)	0.078	0.246	0.060	0.117	0.042	0.402	0.046	0.096
buf(sqa)	0.103	0.794	0.834	0.856	0.349	0.644	0.533	0.635
trt	<0.001	0.089	0.003	<0.001	0.003	<0.001	0.162	<0.001
<i>Trial 2</i>								
HL	31.4	323.0	651.6	375.0	160.4	107.9	12.0	11.5
LH	29.7	307.2	721.6	526.5	235.2	0.0	2.0	10.6
CD	27.0	303.5	740.8	556.2	219.1	2.0	3.1	11.4
SE	2.47	3.05	13.90	15.28	8.74	11.35	0.75	0.42
<i>P-values</i>								
sqa	0.265	0.0003	0.157	0.430	0.136	0.720	0.352	0.192
per(sqa)	0.255	<0.001	0.552	0.370	0.398	0.376	0.142	<0.001
buf(sqa)	0.379	0.006	0.099	0.252	0.584	0.297	0.723	0.202
trt	0.467	0.001	0.001	<0.001	<0.001	<0.001	<0.001	0.273
HH, high C/N and high NDF/SC ratios; LL, low C/N and low NDF/SC ratios; HL, high C/N and low NDF/SC ratios; LH, low C/N and high NDF/SC ratios; CD, control diet. SE, standard error; sqa, square; per, period; buf, buffalo; trt, treatment. OM, organic matter; N, nitrogen; C, carbon; NDF, neutral detergent fiber; ADF, acid detergent fiber; ADL, acid detergent lignin.								

tions of sugar were found in faecal output; differences in sugar intake between the experimental treatments were significant in both trials, while statistically significant differences in the amount of sugar excreted via faeces were only obtained for treatments HL and LH in trial 2. The C/N ratio of the ingesta differed between the treatments in both trials, but for the faecal output only in trial 1. Treatment HH had a C/N ratio in intake of 27.2 and of 12.2 in faecal output, equivalent to a decrease in the C/N ratio of 55% from feed to faeces. A similar decrease was obtained in treatment HL from 23.3 to 11.5 (51%). In treatments LH and LL the decrease was less distinct, namely from 14.1 to 10.6 (25%) and from 11.5 to 7.0 (38%), respectively. The NDF/SC ratio in the ingested feed was different between experimental treatments in both trials, but differences could not be determined for the faecal output because for 6 and 11 animals receiving diets HH and LL (and 15 receiving CD), fae-

cal NDF/SC ratios were >100 due to very low (<0.5%) concentrations of SC in faecal DM.

3.4 Relationship between nutrient intake and faecal output

Simple regression analysis was applied to determine relationships between chemical components in intake and in faecal output (Table 8). Only regression equations with r^2 -values of 0.30 and above are presented. The concentrations of N and starch in faeces were positively and moderately related to the N and starch concentrations in intake with r^2 -values of 0.60 and 0.58, respectively. The NDF and ADF concentrations in faeces were related to NDF and ADL concentration in intake, and the ADF concentration in faeces was additionally negatively influenced by the starch concentration in intake ($r^2 = 0.55$). The ADL concentration in faeces was related only to the ADL concentration in intake.

The C/N ratio of faecal output was weakly correlated to the N and NDF concentration in intake, and to an even lesser extent to the C/N ratio of intake.

4 Discussion

The four experimental diets prepared for the two trials were formulated to differ from each other in C/N ratio (high, low) and NDF/SC ratio (high, low). The experimental design allowed to compare statistically the diets HH (high C/N and NDF/SC ratios) and LL (low C/N and NDF/SC ratios) in trial 1, as well as the diets HL (high C/N and low NDF/SC ratios) and LH (low C/N and high NDF/SC ratios) in trial 2. The control diet was not considered explicitly in the discussion as they were only used to control for the effect of trial period.

4.1 C/N ratio in diet and faeces

Mean values of the C/N ratios in faeces were lower than those of the respective diets in all treatments, but to different extents. Strongly reduced C/N ratios in faeces, as obtained in treatments HH and HL have also been observed in cattle faeces (Delve *et al.*, 2001) and in cattle slurries (Reijs *et al.*, 2007). The N concentration of the diet had the strongest influence on the faecal C/N ratio (Table 8, $r^2 = 0.34$), because it also determined the N concentration of faeces (Table 8, $r^2 = 0.60$), which is in accordance with results of other studies (Sørensen *et al.*, 2003; Powell *et al.*, 2006). The increase in faecal N concentration was higher in diets with low N concentrations, which is also reflected in the lower N digestibility of diets with high C/N ratio (HH and HL). It has been observed that total N excretion is diverted from faeces to urinary urea N, when N concentrations of diets are increasing (Kebreab *et al.*, 2001; Castillo *et al.*, 2001; Broderick, 2003). Animals fed diets with high C/N ratio and therefore low N concentrations excrete N mainly via faeces. The accumulation of N in faeces of animals fed diets with high C/N ratio may also result to some extent from NDF-bound N. In accordance with the presented findings Delve *et al.* (2001) reported that ruminal digestion decreased high C concentration of low quality plant material to a larger extent than the relatively lower C concentration of high quality plant material, while fibre-bound N increased in faeces, resulting in a narrower faecal C/N ratio especially with fibrous diets. The possibility to increase the faecal N concentration in order to increase the amount of N available after manure application to the soil is limited, since increasing the N concentration of diets results only in moderate increases in faecal N, while excess N is excreted via urine. NDF-bound N is less labile and therefore less prone to losses after manure application to the soil. However, from the obtained results it can not be determined if it is possible

to increase the proportion of NDF-bound N by dietary manipulation.

4.2 NDF/SC ratio in diet and faeces

The NDF/SC ratio, being the second determinant in diet formulation, was not calculated for faeces due to the very high digestibilities (>90%) of SC in the experimental diets so that often no SC were found in faeces. This would result in very high or mathematically unfeasible NDF/SC ratios in faeces of all experimental diets. An exception was the HL diet, where high faecal starch and sugar concentrations were observed resulting in lower sugar and starch digestibilities compared to the other diets, although still above 90%. This might be due to the high maize content (36%) in the HL diet. Starch resistant to rumen degradation has often been observed in maize-based diets, and certain amounts also escape the digestion in the small intestines and are excreted with faeces (Hindle *et al.*, 2005; Svihus *et al.*, 2005). The starch concentration in faeces was positively correlated with the starch concentration in intake (Table 8, $r^2 = 0.58$). This shows that the concentration of SC in faeces can be influenced by choosing feed sources rich in starch resistant to fermentation.

Of interest from the point of view of manure quality is how the diverse fibre fractions in faeces, described here by NDF, ADF, ADL, are influenced by the diet composition. The fibre fractions are digested to different extents in the ruminant digestive system and will be decomposable at different rates when faeces are applied as fertilizer to the soil. In the present study, all fibre fractions showed an accumulation in faeces, depending on their proportion in the diet. Since ADL is almost indigestible for ruminants (Van Soest, 1994), the concentration of ADL in faeces was correlated with its intake (Table 8, $r^2 = 0.33$). The picture is not that clear for NDF and ADF, as different cell wall constituents of different ruminal degradation patterns are summarized under these terms. NDF in diets with low NDF/SC ratios (LL and HL) were less digestible. This has also been observed in several *in vivo* and *in vitro* studies with cattle and sheep (Kennedy & Bunting, 1992; Olsen *et al.*, 1999; Sveinbjörnsson *et al.*, 2006). On the one hand this was attributed to a decreasing pH in the rumen with increasing SC content of the diet, which negatively affects the activity of cellulolytic bacteria, and therefore increases the NDF excretion in faeces. On the other hand, rumen microorganisms prefer to use the easy available SC first, so that the lag time for fibre degradation increases and less fibre is degraded during the period the digesta remains in the reticulo-rumen (Kennedy & Bunting, 1992). Especially the digestibility of the hemicellulose is sensitive to starch-associated changes in the ruminal environment. In the present study the digestibility of hemicel-

Table 5: Mean intake of chemical components (g/kg LW0.75/d) of water buffaloes fed four experimental diets (HH, LL, HL, LH) and one control diet (CD) in two feeding trials conducted in Oman.

Diet	N	C	NDF	ADF	ADL	Starch	Sugar	C/N ratio	NDF/SC ratio
<i>Trial 1</i>									
HH	1.05	28.60	48.00	25.09	2.09	0.84	2.17	27.15	15.95
LL	2.19	24.40	22.54	11.83	1.12	7.29	3.33	11.15	2.20
CD	0.95	27.19	36.55	19.53	1.86	4.55	6.50	28.77	3.30
SE	0.03	0.35	0.53	0.28	0.16	0.09	0.05	0.05	0.06
<i>P-values</i>									
sqa	0.076	0.024	0.069	0.065	0.964	0.087	0.008	0.498	0.867
per(sqa)	0.637	0.369	0.285	0.282	1.000	0.439	0.260	0.708	0.753
buf(sqa)	0.536	0.109	0.076	0.073	0.999	0.549	0.114	0.685	0.741
trt	<0.001	<0.001	<0.001	<0.001	0.004	<0.001	<0.001	<0.001	<0.001
<i>Trial 2</i>									
HL	0.93	21.73	25.72	13.29	1.24	10.82	1.59	23.26	2.08
LH	1.47	20.77	28.20	15.71	1.52	2.97	1.12	14.11	6.92
CD	0.74	21.28	28.60	15.28	1.46	3.57	5.17	28.76	3.27
SE	0.01	0.11	0.12	0.06	0.08	0.10	0.02	0.08	0.06
<i>P-values</i>									
sqa	0.781	0.486	0.461	0.419	0.997	0.665	0.371	0.969	0.911
per(sqa)	0.353	0.518	0.648	0.648	0.848	0.500	0.550	0.394	0.369
buf(sqa)	0.170	0.110	0.288	0.240	1.000	0.394	0.425	0.370	0.309
trt	<0.001	<0.001	<0.001	<0.001	0.065	<0.001	<0.001	<0.001	<0.001

HH, high C/N and high NDF/SC ratios; LL, low C/N and low NDF/SC ratios; HL, high C/N and low NDF/SC ratios; LH, low C/N and high NDF/SC ratios; CD, control diet. SE, standard error; sqa, square; per, period; buf, buffalo; trt, treatment.
OM, organic matter; N, nitrogen; C, carbon; NDF, neutral detergent fibre; ADF, acid detergent fibre; ADL, acid detergent lignin.

Table 6: Mean faecal excretion of chemical components (g/kg LW0.75/d) of water buffaloes fed four experimental diets (HH, LL, HL, LH) and one control diet (CD) in two feeding trials conducted in Oman.

Diet	N	C	NDF	ADF	ADL	Starch	Sugar	C/N ratio
<i>Trial 1</i>								
HH	0.27	3.30	8.77	6.04	2.08	0.04	0.05	12.22
LL	0.34	2.37	5.44	3.42	1.49	0.26	0.04	7.01
CD	0.30	3.29	7.66	5.53	1.20	0.07	0.03	11.04
SE	0.01	0.12	0.39	0.28	0.18	0.02	0.01	0.30
<i>P-values</i>								
sqa	0.142	0.045	0.856	0.487	0.621	0.377	0.312	0.523
per(sqa)	0.001	0.001	0.003	0.010	0.500	0.210	0.030	0.100
buf(sqa)	0.216	0.086	0.271	0.558	0.982	0.760	0.316	0.635
trt	0.002	<0.001	<0.001	<0.001	0.073	<0.001	0.367	<0.001
<i>Trial 2</i>								
HL	0.26	2.71	5.39	3.05	1.32	0.95	0.10	11.46
LH	0.18	1.82	4.27	3.10	1.41	0.00	0.01	10.58
CD	0.20	2.17	5.32	4.01	1.55	0.02	0.02	11.44
SE	0.02	0.12	0.21	0.16	0.09	0.11	0.01	0.42
<i>P-values</i>								
sqa	0.442	0.725	0.706	0.671	0.854	0.566	0.467	0.193
per(sqa)	0.320	0.561	0.374	0.634	0.664	0.459	0.452	0.001
buf(sqa)	0.504	0.074	0.033	0.122	0.184	0.369	0.556	0.203
trt	0.062	<0.001	0.003	0.001	0.181	<0.001	<0.001	0.273

HH, high C/N and high NDF/SC ratios; LL, low C/N and low NDF/SC ratios; HL, high C/N and low NDF/SC ratios; LH, low C/N and high NDF/SC ratios; CD, control diet. SE, standard error; sqa, square; per, period; buf, buffalo; trt, treatment.
OM, organic matter; N, nitrogen; C, carbon; NDF, neutral detergent fiber; ADF, acid detergent fiber; ADL, acid detergent lignin.

Table 7: Digestibility (%) of chemical components of water buffaloes fed four experimental diets (HH, LL, HL, LH) and one control diet (CD) in two feeding trials conducted in Oman.

Diet	OM	N	C	NDF	ADF	Starch	Sugar
<i>Trial 1</i>							
HH	83.4	74.1	88.4	82.4	75.0	94.9	97.9
LL	85.8	84.4	90.3	77.7	69.6	96.5	98.8
CD	84.2	68.3	87.9	80.4	73.9	98.5	99.6
SE	0.50	1.00	0.45	1.05	1.56	0.92	0.40
<i>P-values</i>							
sqa	0.072	0.079	0.037	0.440	0.667	0.925	0.422
per(sqa)	<0.001	0.001	0.003	0.021	0.025	0.088	0.086
buf(sqa)	0.059	0.120	0.057	0.541	0.643	0.275	0.307
trt	0.012	<0.001	0.006	0.021	0.065	0.048	0.028
<i>Trial 2</i>							
HL	84.1	72.2	87.5	79.1	77.0	91.3	93.7
LH	88.3	88.1	91.2	84.9	80.2	100.0	98.9
CD	86.4	73.7	89.8	81.4	73.8	99.5	99.6
SE	0.61	2.66	0.51	0.75	1.03	1.00	0.50
<i>P-values</i>							
sqa	0.980	0.471	0.687	0.752	0.668	0.662	0.409
per(sqa)	0.683	0.380	0.510	0.365	0.632	0.618	0.440
buf(sqa)	0.067	0.619	0.061	0.036	0.101	0.339	0.409
trt	<0.001	0.001	<0.001	<0.001	0.002	<0.001	<0.001

HH, high C/N and high NDF/SC ratios; LL, low C/N and low NDF/SC ratios; HL, high C/N and low NDF/SC ratios; LH, low C/N and high NDF/SC ratios; CD, control diet.
SE, standard error; sqa, square; per, period; buf, buffalo; trt, treatment.
OM, organic matter; N, nitrogen; C, carbon; NDF, neutral detergent fiber; ADF, acid detergent fiber; ADL, acid detergent lignin.

Table 8: Significant linear regression equations with r^2 values >0.3 between the concentration of chemical components in intake and in faecal output (g/kg OM) of water buffalo fed four different experimental diets ($n=12/diet$).

Linear Regression Equations	r^2	SE	P	n
$N_FOM = 18.46 + 0.51 \times I_N$	0.60	0.051	<0.001	71
$STA_FOM = -27.55 + 0.59 \times I_STA$	0.58	0.061	<0.001	72
$NDF_FOM = 418.82 + 0.50 \times I_NDF$	0.42	0.073	<0.001	72
$NDF_FOM = 454.25 + 8.73 \times I_ADL$	0.46	1.122	<0.001	72
$ADF_FOM = 219.97 + 0.50 \times I_NDF$	0.33	0.084	<0.001	72
$ADF_FOM = 228.38 + 9.67 \times I_ADL$	0.46	1.254	<0.001	72
$ADF_FOM = 575.74 - 1.02 \times I_STA$	0.55	0.110	<0.001	72
$ADL_FOM = 86.17 + 4.04 \times I_ADL$	0.33	0.686	<0.001	72
$CN_FOM = 14.27 - 0.17 \times I_N$	0.34	0.029	<0.001	72
$CN_FOM = 3.00 + 0.01 \times I_NDF$	0.31	0.003	<0.001	72
$CN_FOM = 6.40 + 0.19 \times I_CN$	0.29	0.036	<0.001	72

X_FOM, concentration of X in faecal organic matter (OM); I_X, concentration of X in organic matter intake; N, nitrogen; STA, starch; NDF, neutral detergent fiber; ADF, acid detergent fiber; ADL, acid detergent lignin; CN, carbon-to-nitrogen ratio.

lulose, calculated as the difference between NDF and ADF, was lower ($P=0.012$) in LL (81 %) compared to HH (88 %) and in tendency also in HL (84 %) compared to LH (89 %) ($P=0.075$; data not shown). The faecal ADF concentration was also influenced by the NDF/SC ratio of the diets, with higher ADF digestibility in diets with a high NDF/SC ratio (HH and LH). This indicates that high amounts of SC also influenced the digestibility of cellulose. This is confirmed by the moderately negative correlation between faecal ADF concentration and starch intake (Table 8, $r^2 = 0.55$), indicating the influence of starch intake on ADF digestibility, which was also observed by Mertens & Loften (1980).

Although NDF and ADF digestibilities were lower in diets with low NDF/SC ratios, the total faecal fibre output is much higher when buffaloes were fed with high NDF/SC ratio diets. Aiming at slowly degrading fibre fractions in faeces, the results therefore suggest a formulation of diets with high NDF/SC ratio. This was achieved especially in combination with a high dietary C/N ratio.

4.3 Dietary manipulation to improve manure quality

Organic fertilizers with C/N ratios below 20 to 25 have been observed to show fast mineralization of N after application to the soil (Senesi, 1989; Myers *et al.*, 1994). All manures in the current study had C/N ratios below this threshold, and also in other studies C/N ratios of faeces were well below this point (Sørensen *et al.*, 2003; Powell *et al.*, 2006). Delve *et al.* (2001) determined an extreme C/N ratio of 86 in barley straw and a C/N ratio of 27 in the faeces of cattle fed only on this straw. Overall it has to be accepted that livestock manures are organic fertilizers with fast N mineralization rates, which can be hardly influenced by dietary manipulation. It has also to be taken into account that manipulating diets to obtain a favourable C/N ratio in faeces are physiologically not balanced, as they mainly contain fibre and only very low amounts of N, so that energy concentration and OM digestibility may be limiting for producing ruminants.

The N concentration of faeces depends on the N concentration of the diet, so that the faecal N concentration can be influenced by dietary manipulation. In temperate regions, intensive systems reduce N content in diets or improve dietary protein quality to reduce N emissions from livestock production systems. Excess supply of N is avoided and the N utilization efficiency of the animals is attempted to be maximized (e.g. Børsting *et al.*, 2003). The problem of excess N is probably less severe in tropical organic crop production. However, the timing of fertilizer application is crucial, so that mineralized N is supplied to the growing crop rather than lost by leaching or volatilization. Therefore especially fibre-bound

N, which is less degradable in the soil, is favourable. The results suggest that manure from animals feeding on diets with high C/N ratios contains more fibre-bound N compared to diets with low C/N ratio. Also here it has to be taken into account that diets with high C/N ratios may limit animal productivity.

The decomposition of C in the soil depends on the C form. In diets C is mainly present in form of SC, NDF or lignin, so that the composition of the C fraction can be influenced by dietary manipulation. Particularly an increased dietary lignin concentration results in increased lignin concentrations in faeces, since lignin is the least decomposable cell wall fraction and almost indigestible for ruminants. To achieve long-term effects with regard to the stability of soil OM, moderate lignin concentrations in manures are desirable. Also high concentrations of other cell wall fractions in diets, lead to high fibre concentrations in faeces. However, lignin and other slowly fermentable fibre fractions like cellulose are strongly negatively correlated to the OM digestibility of ruminant diets (Van Soest, 1994) and therefore possibilities to increase manure lignin concentrations are limited by the expected decrease in animal performance.

5 Conclusions

The results of the present study confirm that manure composition of water buffalo heifers is influenced by diet composition formulated on the basis of C/N ratio and NDF/SC ratio. Despite the reportedly high fibre digestion capacity of the buffalo, digestive processes did not alleviate the manifestation of diet characteristics in faeces. This is of importance when aiming at a specific manure quality for fertilization purposes in crop cultivation. Although there was a strong correlation between ingestion and faecal excretion of N, the faecal C/N ratio reflected poorly the dietary C/N ratio. High dietary NDF and N were the factors influencing faeces composition most, but modulating effects were also achieved by the inclusion of starch into the diet resulting in lower fibre digestibility. With regard to manure quality a diet with high C/N ratio and low NDF/SC ratio is preferable, but diets with high C/N ratio as a result of high fibre, low N and low SC concentrations may not satisfy the nutritional requirements of producing animals.

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