Application of nutrient concentration of cattle faeces to develop algorithms estimating apparent in vivo digestibility and metabolizable energy of tropical ruminant forages

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Application of nutrient concentration of cattle faeces to develop algorithms estimating apparent in vivo digestibility and metabolizable energy of tropical ruminant forages

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Abbreviations and acronyms

ADF	acid detergent fibre
CP	crude protein
DE	digestible energy
DM	dry matter
DMD	dry matter digestibility
DOMD	digestible organic matter in dry matter
GE	gross energy
g	grams
ILRI	International Livestock Research Institute
kg	kilogram
LW	live weight
ME	metabolizable energy
N	nitrogen
NDF	neutral detergent fibre
NIRS	Near Infrared Spectrometry
OM	organic matter
SEM	standard error of mean

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Introduction

In regions of sub-Saharan Africa where there are extensive rangelands, smallholder farmers depend almost exclusively on natural native pastures and forages (Leng 1990) to feed livestock regardless of the nutritive content. This is challenged by feed constraints (i.e. limitations in quality and quantity) but also because information on the nutritive content of the available pastures and forages is scarce and when available, are highly variable. This undermines decisions on feeding and feed improvement to optimize animal performance and health as well as decision-making on best husbandry practices minimizing negative environmental impact.

The nutritive value of feedstuffs is dependent on the chemical composition, its digestibility and the energy content. For natural pastures, nutritive value also depends on the prevailing climate and soil conditions. In the tropics, the native forages are generally high in fibre and low in energy, proteins and minerals, negatively affecting intake levels and digestibility (Jung and Allen 1995).

Currently, the most accurate methods of determination of digestibility are in vivo experiments. These require experimental animals, large quantities of feeds, labour and resource-intensive inputs. In sacco nylon bag technique (Orskov and Mcdonald 1979) and in vitro methods utilizing rumen liquor as microbial source, such as the Hohenheim Gas Test (Menke and Steingass 1988) and Tilley-Terry two-stage method (Tilley and Terry 1963) are substitute methods. However, these methods also require the use of surgically altered animals which many laboratories find difficult to access and maintain. Digestive microbes can also be obtained from faecal matter (van der Baan 2008). However, with faeces, there is need for calibration algorithms since the extent of digestion by faecal microbes is different from that of ruminal microbes (Omed et al. 2000). In vitro methods using enzymes (Aufrere et al. 2007) mimicking rumen microbes have been in use to avoid the problem of access to animals and variations due to rumen liquor source. However, there is a lack of standardization of commercially available batches and there are concerns as to whether two enzymes are sufficient to accurately measure ruminal digestion (Beever and Mould 2000).

Methods which do not involve wet chemistry for determination of digestibility are Near Infrared Spectrometry (NIRS) and use of prediction equations (Decruyenaere et al. 2009). NIRS utilizes a wide database on chemical composition and in vivo digestibility of various feeds for calibration to predict the digestibility of a given feed. Use of prediction equations is similar but less refined. Chemical composition of feeds is regressed against in vivo or in vitro digestibility values to derive equations that only need routine chemical analysis to calculate digestibility values. The limitations of both of these methods include inadequate databases covering the wide variations that exist in feeds, limited correlation between feed chemical characteristics and digestibility as well as propagation of errors from foundational methods used to build the equations. The advantages of these methods are that they are fast, relatively cheap (especially prediction equations that preclude the use of NIRS instrumentation and service) and can be used even in resource constrained developing countries.

The relationship between digestibility and chemical composition has had mixed outcomes. Some studies found a poor correlation between feedstuff digestibility and its chemical composition with parameters such as acid detergent lignin (ADL), crude protein (CP), crude fibre (CF), acid detergent fibre (ADF) and ash (Dardenne et al. 1993); ash, CP, CF and ether extract (EE) (Huhtanen et al. 2006); and fibre content (i.e., CF, NDF, ADF) (Madsen et al. 1997) while others found good correlation with parameters such as CP, lignin, and hemicellulose (Barton II et al. 1976); CP, CF and nitrogen free extracts (Seven and Çerçİ 2006) NDF, ADF, nitrogen (N), gross energy (GE) and EE (Stergiadis et al. 2015). The predictive

equations predicting digestibility also differ among forages, laboratories and environments (Matlebyane et al. 2009; Nousiainen 2004); for example, the USA National Research Council system has been shown to be ineffective in evaluating the nutritional quality of tropical feedstuffs (Detmann et al. 2008). These shortcomings in reliability of regression equations are minimized when specific equations are developed using the same or similar feedstuffs to the ones being tested (Madsen et al. 1997).

The objectives of this study were therefore to develop prediction equations to predict dry matter digestibility (DMD), digestible organic matter in dry matter (DOMD) and metabolizable energy (ME) of tropical feedstuffs using proximate composition (i.e. ash, NDF, ADF, CP and GE) of forages and faecal matter of cattle from in vivo studies.

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Materials and methods

Study site

Studies were performed on the International Livestock Research Institute (ILRI), Nairobi, Kenya campus. The first study (Korir et al. 2016) (further on referred to as the N-balance study) was conducted between May and August 2014. The second study (Goopy et al. in press) was a sub-maintenance feeding study which was conducted between June 2016 and January 2017. A third study on effect of feeding tropical grasses intercropped with or without legumes on cattle performance conducted between April and November 2018 provided the dataset for validation of equations developed using datasets from N-balance and sub-maintenance studies.

Animals and study design

a. N-balance study

In brief, n = 6 Friesian yearling steers (live weight (LW) (mean + standard error of mean (SEM)) = 178.5 ± 24.4 kilogram (kg)); and n = 6 Boran yearling steers (LW (mean ± SEM) = 128.4 ± 12.5 kg) were provided with ad libitum access to clean water in individual pens. Prior to the study the animals were treated for internal parasites with an anthelminthic and for external parasites with an acaricide wash. Initial LW of the animals was determined before the start and subsequent LW recorded once per week during the study period. Details are available in Korir et al. (2016).

A 3 X 3 Latin square design in duplicate was chosen with three treatments (i.e. control (CON) of wheat, Triticum aestivum, daily supplementation (Daily) and bi-daily supplementation (Bi-d) with Calliandra calothyrsus), three periods (each lasting 28 days with 14 days of adaptation and 14 days of measurement) and two breeds (Friesian and Boran). The animals were stratified by LW and randomly assigned to a group of n = 4 animals. Each group was randomly allocated to a treatment diet in the first period. After each period, the groups were randomly allocated to another treatment diet, until all the groups received each diet throughout the study period.

b. Sub-maintenance feeding study

In total n = 12 Boran steers (LW (mean ± SEM) = 183.3 ± 4.3 kg, age = 18 months) were housed, treated and weighed in a similar manner to those in N-balance study described above.

A 3 X 3 Latin square design in duplicate was chosen with four treatments (i.e. fed Rhodes grass, Chloris gayana at 40% of maintenance energy requirement (MER), 60% MER, 80% MER and 100% MER) and five periods (each lasting 49 days with 21 days of adaptation, 14 days of measurement and 14 days of recovery). The animals were stratified by LW and randomly assigned to a group of n = 3 animals. Allocation of groups to treatment diets were done in a similar manner to that in the N-balance study above.

c.   Tropical grasses feeding study (validation dataset)

In this study, n = 18 Boran steers (LW (mean ± SEM) = 230.5 ± 5.0 kg, age = 18 months) were housed, treated and weighed in a similar manner to those in studies described above.

A 3 X 3 Latin square design in duplicate was chosen with six treatments (i.e. fed Napier (Pennisetum purpureum), Brachiaria (Brachiaria brizantha), C. gayana) grasses planted alone, and these grasses planted in intercrop with Lablab, Lablab purpureus) and two periods (each lasting 28 days with 14 days of adaptation and 14 days of measurement). The animals were stratified by LW and randomly assigned to a group of n = 3 animals. Allocation of groups to treatment diets were done in a similar manner to that in the studies above.

Diets, feeding and sample collection

a.   N-balance study

In brief, the animals were fed once daily at 0930 hours with ad libitum chaffed T. aestivum straw hay only, offered at 120% of the previous seven-day intake in a feeding trough in the CON treatment. In the Daily treatment, ad libitum T. aestivum straw hay was supplemented with 20 g/kg LW of air-dried C. calothyrsus leaves and in the Bi-d treatment, ad libitum T. aestivum straw hay was supplemented every other day with 40 g/kg LW of air-dried C. calothyrsus leaves. The amount of C. calothyrsus fed was calculated on the previous weekly LW offered in a separate bucket.

Prior to fresh feeding, refusals were removed and weighed. Faeces excreted over 24 hours in the individual pens was collected during the final seven days of each period. Total faecal weight was determined. Samples of the refusals were pooled by treatment, homogenized and about 200 g was taken and stored in zipped polythene bags at -20 $^{\circ}$C. Samples of T. aestivum straw hay and C. calothyrsus were also taken and stored in similar manner to the refusals. Faecal samples were homogenized by hand and about 500 g subsampled into labelled foil trays then dried in forced-air oven at 50°C to constant weight. They were then cooled in a desiccator, reweighed, and stored in zipped polythene bags at room temperature pending proximate analysis. Further details are available in Korir et al. (2016).

b.   Sub-maintenance feeding study

The animals were fed once daily at 0930 hours with chaffed C. gayana hay at 40%, 60%, 80% and 100% their MER. For animals on the 100% MER treatment, 20% of the energy intake was fed as 48:52 cotton seed meal: molasses mix offered in a separate bucket. Sample collection (i.e. faeces and refusals), weighing, pooling, homogenisation and storage data were collected in a similar manner to the N-balance study.

c.   Tropical grasses feeding study

The animals were fed once daily at 0930 hours with chaffed fresh grasses (P. purpureum planted alone; B. brizantha planted alone; C. gayana planted alone; P. purpureum intercropped with L. purpureus; B. brizantha intercropped with L. purpureus; and C. gayana intercropped with L. purpureus). Sample collection (i.e. faeces and refusals), weighing, pooling, homogenisation and storage data were collected in a similar manner to the studies above.

Laboratory proximate nutrient analysis

The dried feed, refusal and faecal samples were milled (Model MF 10B, IKA Werke, Willmington, N.C., USA) to pass through a one millimetre sieve and analysed for DM at 105°C overnight in a forced-air oven (Method 967.1 according to AOAC (1990)) followed by crude ash determination in a muffle furnace (Model N 11, Nabertherm, Bremen, Germany) at 550°C for four hours (Method 924.05 according to AOAC (1990)). The NDF and ADF were determined by detergent analysis (Van Soest et al. 1991) using an ANKOM200 Fibre Analyser (ANKOM Technology, Macedon, USA). The NDF was determined with the use of sodium sulphate with heat stable α-amylase. The GE concentrations were determined by bomb

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calorimetry using Parr 6200 Isoperibolic calorimeter (Parr Instrument Company, Illinois, USA). Total N in the samples was analysed by micro-Kjeldahl method (Digester and distiller: Model DK and UDK 129 respectively, VELP Scientifica, Usmate, Italy; Titrator: Model 716 DMS Titrino, Metrohm AG, Herisau, Switzerland) (Method 988.05 according to AOAC (1990)). The CP of the samples was determined using the N concentration multiplied by 6.25. All the analyses were done in duplicate and the threshold standard deviation between the replicates set at 5%.

Nutrient digestibility and ME intake

Apparent in vivo and proximate DM digestibility (DMD), digestible organic matter in DM (DOMD) and CP digestibility (CPD) (all in g/100 g) were calculated as:

DMD, g/100 g = DMoffered – DMrefusal – DMfaecal (1)

DOMD, g/100 g = DOMDoffered – DOMDrefusal – DOMDfaecal (2)

CPD, g/100 g = CPoffered – CPrefusal – CPfaecal (3)

The ME intake (MEI) was calculated as digestible energy (DE) (i.e, GE intake – faecal GE) less urinary and methane (CH_4) energy. Due to lack of empirical data on urinary energy, urinary and CH_4 energy was assumed to be 19% of the DE.

MEI, MJ/kg DM = DE – (Urinary + CH_4) Energy

= 0.81 * DE (4)

where (urinary + CH_4) energy = 0.19 * DE

Statistical analysis

Descriptive statistics of variables were determined using R 3.4.4 (R Development Core Team 2016). Tukey honest significant difference was used to compare means and significance declared at $p < 0.05$.

General linear modelling was done using faecal nutrient concentrations as predictor variables and in vivo apparent DMD and DOMD, and ME intake as the response variables. Outliers and points with high leverage were successively identified using component and residual plots, plots of Cook's distance against individual observations, and standardized residuals against leverage defined as:

Cutoff = 4/(n-k-1)

where n = number of observations;

and   k = number of explanatory variables.

These points were checked again to eliminate any possible errors in entry or justification for their existence. Where there were no errors and justification was not possible, the high leverage points were removed from the data and the process repeated until all such points were removed. Four such points were removed in the model. These corresponded to data from two animals who developed health problems in the course of the N-balance study. The linear model was run again and adjusted R-squared was used to determine the goodness of fit; all variables which were not statistically significant ($p > 0.05$) were excluded from the model. This was repeated until all the variables in the model were statistically significant. Stepwise multiple linear regression was then used to come up with the best prediction equation using the lowest Akaike information criterion values. Residual analysis was then performed to determine randomness of distribution, normality and homoscedasticity. When all these conditions were met, the model was declared valid. Further, diagnostic plots were

used to confirm the residual analysis. Where a curve was noted in the residual versus fitted plot, possibility of including a quadratic term in the model was explored and included if it improved the model. Where outliers were noted in the Normal Q-Q plot, their justification or exclusion was explored and executed if it improved the model. Homoscedasticity of data and leverage of points was once again tested after changes in the model.

Internal evaluation was performed to validate the developed prediction equations using root mean square prediction error (RMSE) and mean absolute error (MAE).

$$RMSE = \sqrt{[1/n \sum(P-A)^2]} \qquad (5)$$

$$PE = 1/n\sum(P-A) \qquad (6)$$

where P = predicted value, A = observed value.

An external evaluation of the developed equations was done in a similar manner to internal evaluation but using the validation dataset from the tropical grasses feeding study.

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Results and discussion

Nutrient concentrations and nutrient digestibility

The range of proximate nutrient concentrations in feed samples were 91–94 g DM, 85–88 g organic matter (OM), 2–31 g CP, 18–79 g NDF, 11–47 g ADF, all per 100 g DM, and 17–20 MJ per kg DM of GE, while faecal nutrient concentrations ranged between 84–93 g DM, 72–79 g OM, 6–8 g CP, 58–66 g NDF, 35–39 g ADF, all per 100 g DM, and 17–18 MJ per kg DM of GE (Table 1). Like the N-balance study, the faecal OM and fibre concentrations were lower than their concentrations in feeds.

This can be attributed to the fact that part of these proximate fractions was digested and thus only the undigested portion of the nutrients remain. The faecal CP concentrations were higher in the N-balance study than that of the diet offered which may be indicative of high endogenous losses which is characteristic of a high-fibre diet containing antinutritional factors (Tamminga et al. 1995) such as found in C. calothyrsus (Premaratne and Perera 1999). This was not the case in the relatively good quality diet in the sub-maintenance feeding study where the faecal CP content was numerically lower than that of the offered diet for all treatments. Generally, the GE concentration of the feeds and the faeces were similar, a finding which agrees with that of Osoro and Cebrian (1989) in sheep fed fresh pastures. This may be due to the concentration effect of ash and indigestible cell wall material in the faeces which although have high to GE are of low nutritive value.

Generally, the nutrient digestibility (Table 2) in the sub-maintenance feeding study was higher than in the N-balance study with CP digestibility negative in the latter similar to findings of overall faecal N loss of 18–45 g per day by Koster et al. (1996) in cows fed a low CP diet. This lower digestibility can be attributed to the low quality of the feeds used in the N-balance study especially regarding CP which was below the minimum threshold (< 7 g/100 g DM), particularly in the non-supplementation treatments, to ensure proper functioning of rumen microbes (Korir et al. 2016). The range of the apparent digestibility of DM and OM found (Table 2) is in agreement with that of similar feedstuffs (Goopy et al. 2018).

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Table 1. The proximate concentrations (mean ± SEM) of feeds, feed refusals and faecal samples from: the Nitrogen balance study (Korir et al., 2016) (May to August 2014), the sub-maintenance feeding study (Goopy et al. 2016, in press), and the tropical grasses feeding study (April–November 2018) carried out at ILRI, Nairobi, Kenya.

Study	Samples	n	Treatment	DM	OM	CP	NDF	ADF	GE
				g/100 g FM	g/100 g DM				MJ/kg DM
Nitrogen balance	T. aestivum straw hay	3	N/A	87.7 ± 0.52	93.6 ± 0.64	2.0 ± 0.11	80.7 ± 0.57	46.5*	17.6*
	C. calothyrsus	3	N/A	89.7 ± 0.31	93.6 ± 0.42	25.8 ± 0.40	40.5 ± 0.28	nd	20.2*
	Faecal	11	CON	23.6 ± 1.03	86.2 ± 0.41	6.1 ± 0.13	66.1 ± 0.76	39.1 ± 0.43	17.6 ± 0.11
	Faecal	11	Daily	23.4 ± 0.89	86.8 ± 0.48	7.5 ± 0.74	57.6 ± 5.64	35.1 ± 3.40	17.9 ± 0.07
	Faecal	10	Bi-D	25.0 ± 1.37	86.4 ± 0.48	7.5 ± 0.77	58.1 ± 5.61	35.1 ± 3.38	17.7 ± 0.36
Sub-maintenance feeding	C. gayana hay	1	N/A	86.1	92.3	6.9	72.2	43.0	17.3
	Cotton seed	1	N/A	91.8	92.2	31.4	52.0	37.0	20.3
	Molasses	1	N/A	73.7	92.7	5.1	nd	nd	16.1
	Faecal	15	40%	9.3 ± 0.5	8.3 ± 0.2	6.1 ± 0.18	64.7 ± 2.80	39.3 ± 2.41	17.4 ± 0.08
	Faecal	15	60%	13.4 ± 0.54	11.7 ± 0.23	9.0 ± 0.14	62.0 ± 0.63	36.0 ± 0.63	17.3 ± 0.13
	Faecal	15	80%	15.5 ± 0.59	13.1 ± 0.18	10.0 ± 0.16	61.4 ± 0.58	35.7 ± 0.68	17.5 ± 0.15
	Faecal	15	100%	19.5 ± 0.44	16.5 ± 0.25	15.5 ± 0.24	62.1 ± 1.13	37.2 ± 0.96	17.6 ± 0.08
Tropical grasses feeding	P. purpureum	10	NP	15.9 ± 0.86	85.0 ± 0.57	8.9 ± 0.59	64.1 ± 0.58	41.1 ± 0.79	nd
	B. brizantha	10	BR	25.0 ± 1.89	88.3 ± 0.26	8.4 ± 0.32	65.6 ± 0.58	40.1 ± 0.83	nd
	C. gayana	10	RH	27.7 ± 1.79	89.6 ± 0.23	7.6 ± 0.33	68.8 ± 0.75	42.3 ± 0.97	nd
	P. purpureum - L. purpureus intercrop	10	NP-LA	16.7 ± 1.16	84.9 ± 0.63	8.8 ± 0.46	65.5 ± 0.81	41.8 ± 0.79	nd
	B. brizantha - L. purpureus intercrop	10	BR-LA	26.2 ± 1.60	88.7 ± 0.29	8.2 ± 0.32	66.2 ± 1.08	40.7 ± 1.13	nd
	C. gayana - L. purpureus intercrop	10	RH-LA	27.8 ± 1.57	89.0 ± 0.51	7.7 ± 0.48	67.9 ± 0.83	41.5 ± 0.83	nd
	Faecal		NP	16.3 ± 7.97	79.4 ± 13.35	7.8 ± 0.22	56.4 ± 12.37	37.1 ± 13.30	nd
	Faecal		BR	16.4 ± 9.21	82.6 ± 5.80	7.3 ± 0.14	56.2 ± 9.02	35.8 ± 9.95	nd
	Faecal		RH	17.7 ± 3.95	83.7 ± 6.19	7.2 ± 0.22	57.5 ± 12.78	36.7 ± 10.96	nd
	Faecal		NP-LA	15.9 ± 6.21	78.7 ± 14.70	7.6 ± 0.32	54.5 ± 14.33	36.8 ± 9.95	nd
	Faecal		BR-LA	17.7 ± 6.06	82.6 ± 6.52	7.5 ± 0.14	54.4 ± 15.03	34.4 ± 13.75	nd
	Faecal		RH-LA	17.7 ± 6.31	82.6 ± 2.71	7.6 ± 0.37	56.3 ± 9.42	35.5 ± 9.82	nd

ADF = acid detergent fibre; Bi-D = bi-diurnal supplementation with C. calothyrsus; BR = B. brizantha planted alone; BR-LA = B. brizantha intercropped with L. purpureus; C. calothyrsus = Calliandra calothyrsus; C. gayana = Chloris gayana; CON = control treatment feeding T. aestivum straw hay alone; CP = crude protein; Daily = once a day supplementation with C. calothyrsus; DM = dry matter; GE = gross energy; L. purpureus = Lablab purpureus; N/A = not applicable; nd = not determined; NDF = neutral detergent fibre; NP = P. purpureum planted alone; NP-LA = P. purpureum intercropped with L. purpureus; OM = organic matter; RH = C. gayana planted alone; RH-LA = C. gayana intercropped with L. purpureus; SEM = standard error of mean.

*samples pooled to give one composite sample

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Table 2. Nutrient digestibility (arithmetic mean ± SEM) of i) T. aestivum straw hay (with and without C. calothyrsus supplementation) obtained with yearling steers fed at maintenance; ii) C. gayana hay (with and without cotton seed and molasses supplementation) obtained with 18-month-old steers fed at 40%, 60%, 80% and 100% maintenance energy requirement feeding level; and iii) B. brizantha, P. purpureum, and C. gayana all planted alone or each intercropped with L.purpureus obtained with yearling steers fed at maintenance level; studies carried out between May 2014 and November 2018 at ILRI, Nairobi, Kenya.

Parameter	Study		
	N-balance[i)]	Sub-maintenance[ii)] feeding	Tropical grasses[iii)] feeding
DMD (g/100 g)	57.1 ± 1.22	56.3 ± 0.86	59.3 ± 0.72
OM digestibility (g/100g)	60.2 ± 1.13	59.6 ± 1.35	62.1 ± 0.72
CP digestibility (g/100g)	15.2 ± 3.77	59.6 ± 1.54	64.2 ± 0.91

Digestibility and ME intake algorithms using faecal nutrient concentrations

Generally, the results here show that faecal proximate composition is a very weak predictor of ME and digestibility of feedstuffs.

a.   Apparent digestibility algorithms

The models predicting DMD and DOMD showed poor fit (i.e. adjusted R-squared of between 0.05 and 0.10, equations Tables 3, 4 and 5) and weak correlations between predicted and actual values (i.e. 0.3), but with low prediction errors (i.e. 3–5% for digestibility). The best predictors of DMD were faecal DM, ADF and NDF while the best predictors of DOMD were NDF, ADF and CP when data from both studies were used. The quality of equations 1a, 1b, 3a and 3b could have been compromised by inclusion of both ADF and NDF variables due to the possible collinearity of the two variables. However, removal of one or both the terms rendered the model worse according to the subsequent adjusted R-squared. Similarly, the best predictors of apparent digestibility when only data from the sub-maintenance feeding study were used were faecal NDF and ADF while only feacal DM was a predictor of digestibility when one data from N-balance study was considered. The negative relationship between apparent digestibility with faecal DM and faecal ADF can be attributed to the fact that the quantity of these nutrients in faeces is inversely proportional to the extent of their digestibility.

This is because they have no source within the gastro-intestinal tract and their presence in faeces means they were in the feed but were not fully digested. Indeed, an increase of 10 g/100 g DM of fibre fractions in grass have been found to decrease DOMD of fresh-cut grass by 5.1 g/100 g DM (Stergiadis et al. 2015a), frozen grass by 7.7–9.0 g/100 g DM (Givens et al. 1990a) and by 8.0–10.9 g/100 g DM in grass silages (Yan and Agnew 2004).

Table 3. Equations predicting in vivo DMD, DOMD and ME intake of feedstuffs in cattle using proximate composition of faeces data from both N-balance and sub-maintenance feeding studies carried out at International Livestock Research Institute, Nairobi between May and August 2014, and June 2016 and January 2017 respectively (n = 92)

Parameter	Equation	Adj. R[2]	p-value	r	RMSE	PE	Equation
DMD (g/100 g DM)	52.8 − 0.3fDM + 0.4fNDF − 0.4fADF	0.059	0.043	0.320	4.0	±5	1a
DOMD (g/100 g DM)	28.5 + 0.6fNDF − 0.5fADF + 1.0fCP	0.098	0.008	0.370	4.0	±5	1b
ME intake (MJ/day)	75.3 − 1.8fDM − 1.9fADF + 1.8fCP	0.361	<0.001	0.660	7.0	±22	1c

Adj. R[2] = adjusted R-squared; DMD = dry matter digestibility; DOMD = digestible organic matter in dry matter; fADF = faecal acid detergent fibre; fCP = faecal crude protein; fDM = faecal dry matter; ME = metabolizable energy; n = number of observations; fNDF = faecal neutral detergent fibre; PE = mean error = mean (predicted value − observed value); r = correlation coefficient between actual and predicted values; RMSE = root mean squared error = square root (mean (predicted value − observed value)[2])

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Table 4. Equations predicting in vivo DMD, DOMD, and ME intake of feedstuffs in cattle using proximate composition of faeces data from N-balance feeding study carried out at International Livestock Research Institute, Nairobi between May and August 2014 (n = 32)

Parameter	Equation	Adj. R^2	p-value	r	RMSE	PE	Equation
DMD (g/100 g DM)	67.2 – 0.4fDM	0.059	0.096	0.300	4.0	±3	2a
DOMD (g/100 g DM)	68.7 – 0.3fDM	0.049	0.118	0.280	4.0	±3	2b
ME intake (MJ/day)	71.2 – 2.1fDM	0.487	<0.001	0.710	7.0	±5	2c

Adj. R^2 = adjusted R-squared; DMD = dry matter digestibility; DOMD = digestible organic matter in dry matter; fDM = faecal dry matter; ME = metabolizable energy; n = number of observations; PE = mean error = mean (predicted value – observed value); r = correlation coefficient between actual and predicted values; RMSE = root mean squared error = square root (mean (predicted value – observed value)^2)

Table 5. Equations predicting in vivo DMD, DOMD and ME intake of feedstuffs in cattle using proximate composition of faeces data from sub-maintenance feeding carried out at International Livestock Research Institute, Nairobi between June 2016 and January 2017 (n = 60)

Parameter	Equation	Adj. R^2	p-value	r	RMSE	PE	Equation
DMD (g/100 g DM)	41.5 + 0.7fNDF – 0.7fADF	0.056	0.079	0.300	3.0	±3	3a
DOMD (g/100 g DM)	36.1 + 0.7fNDF – 0.6fADF	0.063	0.065	0.310	4.0	±3	3b
ME intake (MJ/day)	66.8 – 1.3fDM – 0.6fNDF + 3.7fCP	0.412	<0.001	0.670	6.0	±5	3c

Adj. R^2 = adjusted R-squared; DMD = dry matter digestibility; DOMD = digestible organic matter in dry matter; fADF = faecal acid detergent fibre; fCP = faecal crude protein; fDM = faecal dry matter; fNDF = faecal neutral detergent fibre; ME = metabolizable energy; n = number of observations; PE = mean error = mean (predicted value – observed value); r = correlation coefficient between actual and predicted values; RMSE = root mean squared error = square root (mean (predicted value – observed value)^2)

On the other hand, positive relationship of faecal CP with apparent digestibility could be indicative of undigested microbial mass from high ruminal microbe turnover indicative of high feed digestion, assuming that it is as a result of high endogenous CP losses.

Positive relationship with faecal NDF may be attributed to the fact that high faecal NDF may be due to the net concentration effect indigestible fibre fractions which may also be an indicator of the high extent of digestion of other fractions.

b. Metabolizable energy intake algorithms

One prediction equation (i.e. 1c in Table 3) was developed for ME intake using data from both studies and two equations (i.e. 2c and 3c in Tables 4 and 5 respectively) were developed using nitrogen balance and sub-maintenance feeding studies data alone. Generally, the models showed a poor to moderate fit between ME intake and faecal nutrient concentrations (i.e. adjusted R-squared between 0.4–0.5) similar to a weak to moderate positive correlation between predicted and actual ME intake values (i.e. 0.7). However, the prediction errors were relatively high based on the combined data from both studies (i.e. 22 MJ/day) but very low when the data from individual studies were used alone (i.e. 5 MJ/day). The models developed using data from individual studies alone performed better (in terms of fit, correlation and prediction errors) than that developed using the combined dataset from both studies.

The best predictors of ME intake were faecal: DM, CP, ADF and NDF. Stergiadis et al. (2015a) using chemical composition of fresh-cut grass to predict ME also found low correlation (i.e. 0.4–0.5) with N and ADF as the best predictors but also ash, water soluble carbohydrates and ether extract, the latter two parameters not tested here. The ME was negatively related to faecal DM, NDF and ADF possibly because high faecal concentrations of these fractions shows low digestibility of a feedstuff hence low ME intake from such feedstuff. Using combined data from both studies and data from sub-maintenance study alone, faecal CP was positively related to ME intake. Stergiadis et al. (2015a) and Givens et al. (1990b) also found that feedstuff N is positively related to ME. High faecal N/CP could be indicative of N partition towards the faeces as opposed to the urinary route hence lower urinary energy losses meaning high ME intake (Givens et al. 1989). It could also mean high microbial mass from ruminal digestion (indicative of efficient ruminal digestion hence high ME intake) but poor post-ruminal digestion or high endogenous losses, both of which would lead to low ME intake in which case a negative relationship would be expected. High faecal NDF and ADF indicates poor digestibility or high feed passage rate both of which would result in low ME intake. Similarly, if the feedstuff NDF and ADF was high to begin with, there would be depressed feed intake due to rumen fill which would still lead to low ME intake (i.e. negative relationship). Development of ME intake algorithms using sub-maintenance feeding data alone avoided any noise that could have been present due to insufficient CP supply to ruminal microbes in the N-balance study (Table 5). This was also partly informed by the findings

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that forage-based diets of low CP have low organic matter digestibility which increase on CP supplementation (Karsli and Russell 2002). There is need to investigate this relationship further to find out a plausible reason and/or the right relationship using a different database.

c.   Validation of the digestibility equations using data from study on feeding tropical grasses

Using an independent dataset to validate the equations shows that equations derived from N-balance study were better predictors of both DMD and DOMD (Table 6, equations 2a and 2b) than equations derived from sub-maintenance study alone or those derived from the two datasets combined. This is probably because the diet fed to the animals in this study whose data was used to derive digestibility equations was closest to the optimal diet used in the study of feeding tropical grasses. This is because, of the four animals fed below optimum protein (control: CON), two developed health problems and therefore their data, which corresponded to high leverage points, was excluded when the equations were developed leaving data from optimally fed animals to derive the digestibility equation. Overall, prediction of DMD was much better than that of DOMD. It is not immediately apparent why this is so. It is hoped that the reason may become clearer as more data is added to the initial dataset. Due to lack of information on ME intake in the study on feeding tropical grasses, only DMD and DOMD equations were validated.

Table 6. Validation of equations predicting in vivo DMD and DOMD (both in g/100 g DM) of feedstuffs in cattle using proximate composition of faeces data from a study on feeding studies tropical grasses carried out at International Livestock Research Institute, Nairobi between April–November 2018 (n = 36)

Source of data	Parameter	Equation	RMSE	PE	Eq
Combined N-balance and Sub-maintenance studies	DMD	52.8 - 0.3fDM + 0.4fNDF - 0.4fADF	5.9	±5	1a
	DOMD	28.5 + 0.6fNDF - 0.5fADF + 1.0fCP	11.8	±11	1b
N-balance study alone	DMD	67.2 - 0.4fDM	4.4	±4	2a
	DOMD	68.7 - 0.3fDM	4.5	±4	2b
Sub-maintenance study alone	DMD	41.5 + 0.7fNDF - 0.7fADF	6.4	±5	3a
	DOMD	36.1 + 0.7fNDF - 0.6fADF	10.1	±9	3b

DMD = dry matter digestibility; DOMD = digestible organic matter in dry matter; Eq = equation; fADF = faecal acid detergent fibre; fCP = faecal crude protein; fDM = faecal dry matter; ME = metabolizable energy; n = number of observations; fNDF = faecal neutral detergent fibre; PE = mean error = mean (predicted value – observed value); RMSE = root mean squared error = square root (mean (predicted value – observed value)^2)

Conclusions

Equations relating faecal chemical composition to digestibility and ME are weak but may be improved by using a wider, more varied diet as well as more samples. The above findings demonstrate the possibility of using cattle's faecal proximate, fibre and energy composition to predict the apparent digestibility and ME content of feedstuffs. Of possible interest is that only four faecal parameters were of importance (i.e. DM, CP, NDF and ADF). Analyses of these parameters, in addition to GE used in ME estimation, is fairly simple, routine, cheap and does not need sophisticated laboratory infrastructure. Ideally, equations should be developed from studies carried out at maintenance level feeding assuming no nutritional constraints. However, nutritional constraints in smallholder farming is more often the rule than the exception. Therefore, such equations should be developed from studies which reflect the feeding situations similar to the prevailing conditions. This will require a large database of in vivo (and in vitro) studies reflecting the different situations of feeding under constraints. This study, therefore, is a first step towards development of such a database and the accompanying prediction equations with the ultimate aim of finally proposing prediction equations that are appropriate for smallholder feeding situations.

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