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The use of the *in vitro* filter bag method for predicting digestibility of forages*

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ABSTRACT

The possibility of predicting the *in vivo* organic matter digestibility of forage by estimating *in vitro* digestibility using either rumen fluid or cellulase as incubation media in the filter bag method was studied. The *in vivo* OM digestibility of 35 forage samples was determined on 6 rams and *in vitro* digestibility was determined by two methods: rumen fluid (in a Daisy^{II} Incubator)-neutral detergent (RF-ND) or cellulase solution (also in a Daisy incubator)-neutral detergent (C-ND). The coefficients of correlation between *in vivo* digestibility and that estimated by the RF-ND and C-ND methods were 0.62 and 0.69, respectively. Regression analysis shows that both tested methods have good potential for predicting *in vivo* OM digestibility.

KEY WORDS: *in vitro*, digestibility, forages, filter bag method

INTRODUCTION

The filter bag method developed by Ankom (Ankom Co, Fairport, NY) is a good alternative to classical *in vitro* digestibility methods. Most studies have used buffered rumen fluid as the incubation medium (e.g., Mabjesh et al., 2000; Wilman and Adesogan, 2000), but this method has its limitations related to standardizing rumen fluid. Recently, the use of enzymes (cellulase and xylanase) instead of rumen fluid has been studied (Ludwin and Kowalski, 2004). The aim of the present study was to determine the possibility of predicting *in vivo* forage organic matter digestibility by *in vitro* digestibility estimated using either rumen fluid or cellulase solution as the incubation medium in the filter bag method.

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MATERIAL AND METHODS

The *in vivo* organic matter digestibility of 35 forage samples from Denmark (10 maize silages, 5 whole crop barley silages, 10 green whole crop maize, 10 green whole crop barley) was determined on 6 rams, using the total collection method with a 7-day preliminary period and 7-day faecal collection. The chemical composition of feedstuffs was determined by standard methods (AOAC, 1995). The *in vitro* digestibility (IVD) was determined on the same 35 samples ground to pass through a 1 mm screen. The forages were dried in an air-forced oven for 48 h at 50°C. About 0.5 g of sample of feed was placed in a filter bag (Ankom F57;50 × 55 mm) made from polyester-polyethylene extruded filaments of a pore size about 25 μm. There were 4 bags for each feed. The bags were then placed in glass jars and incubated in a Daisy^{II} Incubator (Ankom Co., Fairport, NY). The IVD was determined by two methods: incubation of the sample for 48 h at 39.5°C in rumen fluid (RF-ND) or in a cellulase solution (C-ND) in a Daisy^{II} Incubator and then (in both methods) boiling the residue in neutral detergent for 1 h in an Ankom Fiber Analyser (for details see Ludwin and Kowalski, 2004). The results of two *in vitro* digestibility methods were subjected to one-way analysis of variance (SAS, 1996). Linear regression equations were calculated to estimate the precision of predicting *in vivo* digestibility when using both filter bag methods.

RESULTS

The variation in chemical composition of feedstuffs (Table 1) was sufficient to use this set of samples in the comparison. The average *in vivo* OM digestibility was 72.0%, and varied from 63.2 to 78.5%. The IVD of all forages estimated by the C-ND method was significantly higher than when estimated by RF-ND. When the set of samples was divided according to types of forages, the differences between *in vitro* methods were significant for maize and silages but they were not significant for barley and green forages (Table 2). Coefficients of correlation between *in vivo* OM digestibility and IVD estimated by the RF-ND and C-ND methods were 0.62 ($P<0.01$) and 0.69 ($P<0.01$), respectively. The linear regression equations for both methods are: $y=0.672x + 26.33$ (RF-ND; $n=35$; $r^2=0.39$; SE=3.10) and $y=0.6939x + 22.766$ (C-ND; $n=35$; $r^2=0.48$; SE=2.86), respectively.

Table 1. Chemical composition of feed, % DM

Item	OM	CP	CF	NDF	ADF	Starch
Mean	95.63	9.06	22.49	43.37	24.94	27.21
Standard deviation	0.83	1.42	3.47	3.74	2.76	6.08
Minimum	93.35	6.54	17.58	35.69	20.56	11.51
Maximum	96.85	12.45	35.14	50.49	31.88	35.51

Table 2. Means of *in vivo* OM and *in vitro* digestibility (%) using two filter bag methods: rumen fluid-neutral detergent (RF-ND) and cellulase-neutral detergent (C-ND)

Item	N	*In vivo* OM digestibility	RF-ND		C-ND		Main effect[1]
			mean	SD	mean	SD	
All forages	35	72.0	68.0	1.9	71.0	0.8	**
Maize	20	74.2	69.5	1.9	73.6	0.8	***
Whole crop barley	15	69.2	66.1	1.8	67.5	0.7	NS
Silages	15	73.4	67.7	2.0	72.1	0.7	**
Green whole crop	20	71.0	68.3	1.7	70.2	0.8	NS

[1] RF-ND vs C-ND; ** - P<0.01, *** - P<0.001, NS - not significant

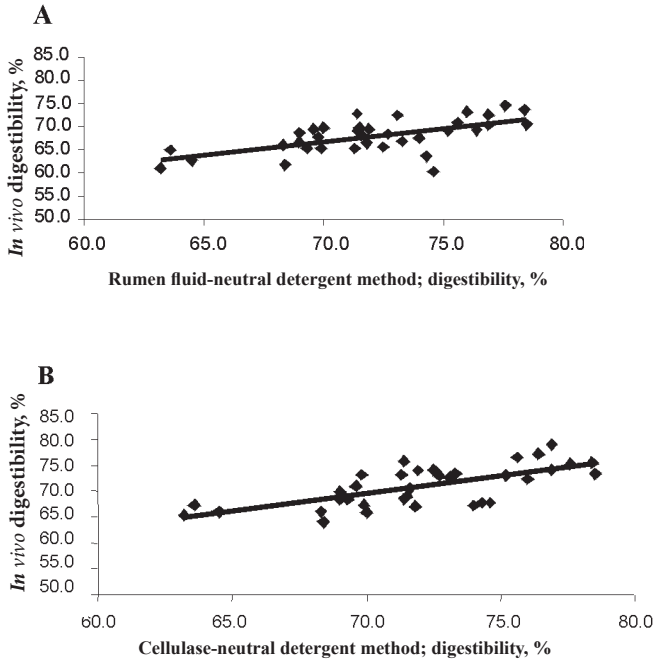

Figure 1. The relationship between *in vivo* OM digestibility and filter bag *in vitro* digestibility when measured using rumen fluid-neutral detergent (A) or cellulase-neutral detergent (B)

DISCUSSION

This study showed that the filter bag method could be effectively used to predict *in vivo* digestibility of forages. It allows simultaneous incubation of many samples and different samples can be incubated at the same time in the same medium (Holden, 1999). The incubation medium, however, had a significant effect on IVD, with the C-ND method giving slightly higher digestibility coefficients. Regression

analysis shows that both tested methods have a good potential for predicting *in vivo* OM digestibility. However, both the higher correlation coefficient and lower SE suggest that using cellulase instead of rumen fluid can be even more advantageous. Using enzymes also seems to be easier for standardization.

CONCLUSIONS

It can be concluded that the filter bag method is an easy and precise tool for studying *in vivo* digestibility. Enzymes have potential as an alternative medium to rumen fluid in this technique.

REFERENCES

Holden L.A., 1999. Comparison of methods of *in vitro* dry matter digestibility for ten feeds. J. Dairy Sci. 82, 1791-1794

Ludwin J., Kowalski Z.M., 2004. The effect of different sources of xylanase on *in vitro* dry matter digestibility measured by the filter bag method. J. Anim. Feed Sci. 13, Suppl. 2, 71-74

Mabjesh S.J., Cohen M., Arieli A., 2000. *In vitro* methods for measuring the dry matter digestibility of ruminant feedstuffs: comparison of methods and inoculum source. J. Dairy Sci. 83, 2289-2294

SAS, 1996. Release 6.12 for Windows. SAS Institute Inc., SAS Campus Drive, Cary, NC

Wilman D., Adesogan A., 2000. A comparison of filter bag methods with conventional tube methods of determining the *in vitro* digestibility of forages. Anim. Feed Sci. Tech. 84, 33-47

STRESZCZENIE

Wykorzystanie metody woreczków filtracyjnych w szacowaniu strawności pasz objętościowych

Określano możliwość szacowania strawności *in vivo* pasz objętościowych na podstawie strawności *in vitro* oznaczonej metodą woreczków filtracyjnych. Strawność *in vivo* masy organicznej 35 pasz (zielonki i kiszonki z kukurydzy oraz z całych roślin jęczmienia) oznaczono metodą klasyczną na 6 trykach, strawność *in vitro* oznaczano 2 metodami: płyn żwacza (inkubacja w inkubatorze Daisy^{II})-detergent neutralny (RF-ND) lub roztwór celulazy (także w inkubatorze Daisy^{II})-detergent neutralny (C-ND). Współczynniki korelacji prostej pomiędzy strawnością *in vivo* i oznaczoną metodami RF-ND i C-ND wynosiły odpowiednio 0,62 i 0,69. Analiza regresji wskazuje, że obydwie metody *in vitro* mogą być stosowane do szacowania strawności *in vivo* masy organicznej pasz objętościowych.