Prediction of digestibility and energy concentration of catch crops – A comparison of laboratory methods

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Schätzung der Verdaulichkeit und Energiekonzentration von Ackerpferch-Zwischenfrüchten – Ein Methodenvergleich

Introduction

The energy value and digestibility of organic matter (= DOM) are important factors of feed quality. For determination of DOM *in vivo*-animal tests are necessary. As animal tests require more time, money and plant material, for the routine praxis many estimation methods are available, which differ in time and money spent, accuracy and reproducability (OPITZ V. BOBERFELD 1994). Secondary compounds, like tannins and glucosinolates (STÄHLIN 1971, ISSELSTEIN 1994), as well as fungal infection (HÖLTERSHINKEN et al. 1996, 2000) can change the activity of rumen microbes and consequently also the DOM (DEINUM and VAN SOEST 1969; VAN DER KOELEN and VAN ES 1973; TINNIMIT and THOMAS 1976; AERTS et al. 1977; KIRCHGESSNER and KELLNER 1981; OPITZ V. BOBERFELD et al. 2003).That is why especially for forage grown in low-input systems, such as suckler cow and beef cattle husbandry, or for plants, which contain secondary compounds – like glucosinolates in *Brassicaceae* – *in vitro* methods with rumen fluid seem to be most reliable for predicting digestibility of organic matter; methods, which do not consider such circumstances could implicate considerable miscalculations. Furthermore miscalculations can result from using standard regression equations for predicting digestibility of *Brassicaceae*, as this plant-group, which is rich in carbohydrates and contains glucosinolates, is taken into consideration insufficiently. Beside that common regression equations for all types of feed cause an over-

Zusammenfassung

Die Verdaulichkeit organischer Substanz und Energiekonzentration verschiedener Ackerpferch-Zwischenfrüchte, darunter drei *Brassicaceen*, wurden anhand verschiedener Labormethoden geschätzt – zwei *in vitro* Methoden mit Pansensaft, eine enzymatische Methode und eine chemische Methode. Die *in vitro* Pansensaftmethoden lieferten verlässlichere Schätzergebnisse, insbesondere wenn pilzlicher Befall oder sekundäre Inhaltsstoffe der Pflanzen zu einer Störung der mikrobiellen Aktivität im Pansen führen konnten. Die bisherigen Regressionsgleichungen sollten unter Einbeziehung von Zwischenfrüchten wie *Brassicaceen* erneuert werden, um besondere Effekte dieser Futtermittelgruppe auf die Verdaulichkeit organischer Substanz zu berücksichtigen.

Schlagworte: Verdaulichkeit organischer Substanz, Energiekonzentration, Schätzmethoden, Zwischenfrüchte, *Brassicaceen.*

Summary

The digestibility and energy concentration of different catch crops, including three *Brassicaceae*, were estimated with four laboratory methods – two *in vitro* methods with rumen fluid, one enzymatic and one chemical method. The *in vitro* methods with rumen fluid were more accurate, especially when fungal infection or secondary compounds could influence the activity of rumen microbes. The chemical method was not reliable for predicting digestibility of organic matter. The common regression equations should be renewed in consideration of catch crops, like *Brassicaceae*, and potential specific effects on digestibility of this group of feeds.

Key words: Digestibility of organic matter, energy concentration, estimation methods, catch crops, *Brassicaceae.*

estimation of low-energy feed and an underestimation of feed rich in energy; the differences in digestibility of crude nutrients cause that the median regression coefficients, calculated for the equations in consideration of all types of feed, are too high when the equation is used for low-energy feed and conversely (MENKE & STEINGASS 1987). Widespread methods for estimation of DOM beside the *in vitro*-methods with rumen fluid – Hohenheim gas test and rumen fluid-pepsin method – include the pepsin-cellulase method for predicting the enzymatic solubilisation of organic matter (= ELOS), the chemical method using the crude nutrients as well as the near infrared reflectance spectroscopy (= NIRS) as a physical method, which nevertheless entails a larger error, as it is calibrated and validated by other estimation methods.

2 Material and methods

Several catch crops in arable land folds – including three *Brassicaceae* – were investigated in triennial experiments, located 160m above sea-level in the south of Gießen. The trial was established in a split-plot design with three replicates, considering the factors species, date of winter harvest, date of sowing and year (Table 1).

Table 1: Variants
Tabelle 1: Varianten

Factor	Treatment
1. Species	1.1 Winter forage rage (= leaf-rich), *Brassica napus* ssp. napus (AKELA (0), LIRATOP (00) – 12 kg ha^{-1})
	1.2 Summer forage rape (= stem-rich), *Brassica napus* ssp. *napus* (PETRANOVA (0), LIFORUM (00) – 12 kg ha^{-1})
	1.3 Stubble turnip, *Brassica rapa* ssp. *rapa* (AGRESSA, SILOGANOVA – 1,5 kg ha^{-1})
	1.4 Italian ryegras 4n, *Lolium, multiflorum* ssp. *gaudinii* (ANDY, JIVET – 50 kg ha^{-1})
	1.5 Volunteer cereals, *Hordeum vulgare* (Wintergerste-Simulation – 500 kg ha^{-1})
2. Harvest date	2.1 Early November 2.2 Middle December 2.3 Late January
3. Sowing date	3.1 Late July 3.2 Middle August
4. Year	4.1 2001/2002 4.2 2002/2003

To avoid interactions of higher order years were statistically analysed separately. The digestibility of organic matter (= DOM) was predicted with different estimation methods. The Hohenheim gas test (= HFT) estimates the DOM by the variables gas production and crude protein using the regression equation (1); the energy concentration is estimated by the variables gas production, crude protein and crude fat using equation (2) (STEINGASS und MENKE 1986; MENKE und STEINGASS 1987). Further methods for predicting the DOM were the two-stage-technique by TILLEY and TERRY (1963), where the plant material is incubated in rumen fluid and pepsin-HCl solution, and the enzymatic method with an incubation in acid pepsin- and cellulase-solution (= ELOS) (ANONYMUS 1997a). The energy value was also predicted by using regression equation (3) after analysing the crude nutrients (HOFFMANN et al. 1972; ANONYMUS 1988, 1993, 1995); the percentage digestibility of the crude nutrients was taken out of the DLG-table of feeding values for ruminants (ANONYMUS 1997b). The transformation from ME to NEL followed equation (4) according to POTTHAST et al. (1997).

Regression equations:

HFT

$$DOM\ (\%) = 24{,}59 + 0{,}7984\ \text{gas vol.} + 0{,}0496\ CP \quad (1)$$

$$ME\ (MJ) = 2{,}2 + 0{,}1357\ \text{gas vol.} + 0{,}0057\ CP + 0{,}0002859\ EE^2 \quad (2)$$

chemical method

$$ME\ (MJ) = 0{,}0312\ DEE + 0{,}0136\ DCF + 0{,}0147\ (DOM - DEE - DCF) + 0{,}00234\ CP \quad (3)$$

$$NEL\ (MJ) = ME\ [0{,}48 + 1037\ ME/(1000 - CA)] \quad (4)$$

where

gas vol. = Volume of produced gas after fermentation in rumen fluid in ml 200mg^{-1} DM in 24 h
CP = Crude protein in g kg^{-1} DM
CA = Crude ash in g kg^{-1} DM
EE = Crude fat in g kg^{-1} DM
CF = Crude fibre in g kg^{-1} DM
D- = Digestibility of crude nutrients

3 Results and discussion

Figure 1 represents the comparison of the digestibility of organic matter (= DOM), estimated with the Hohenheim gas test (= HFT), with the rumen fluid-pepsin method and with the enzymatic method (= ELOS). The results of the comparison between HFT and rumen fluid-pepsin method

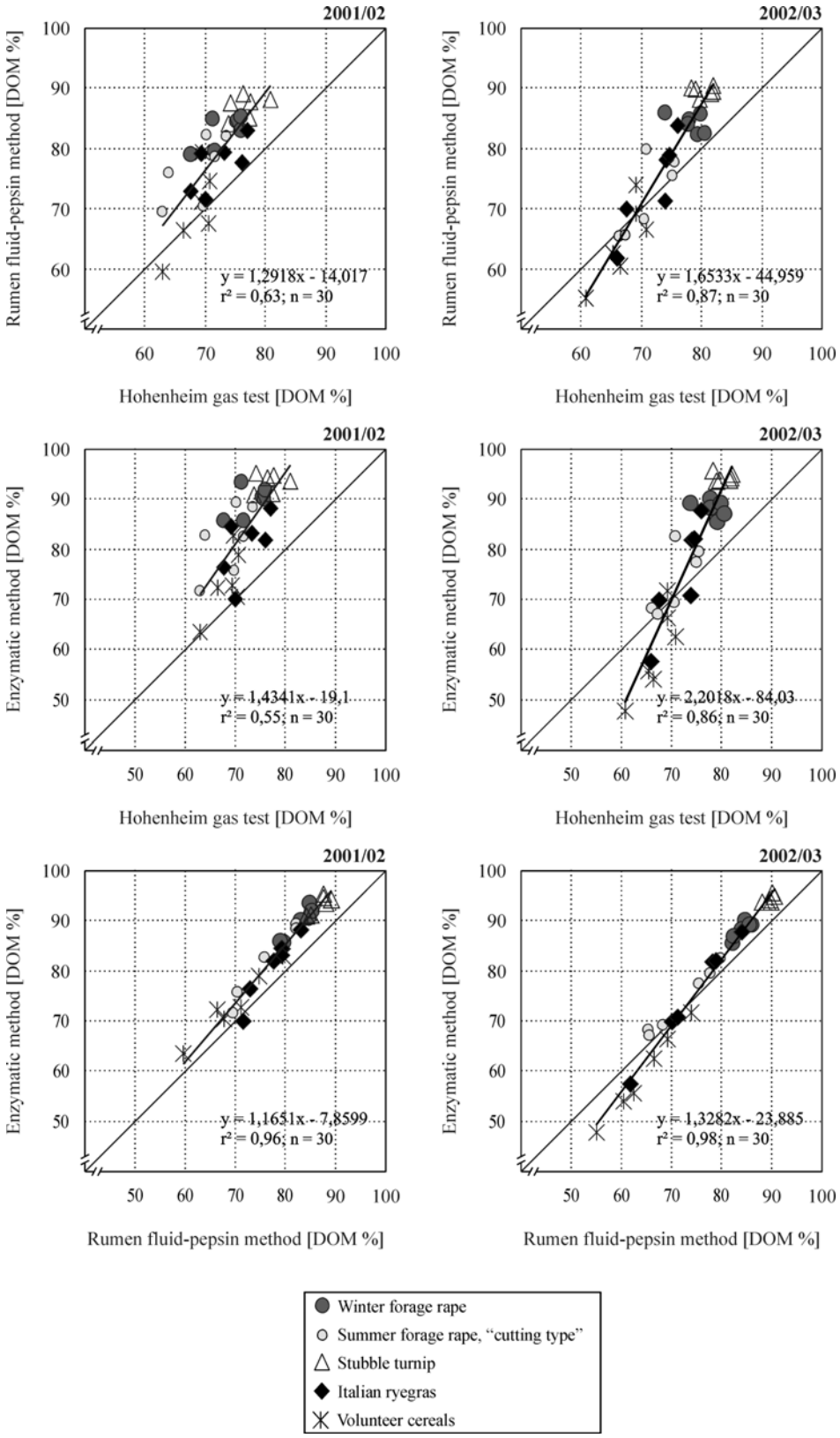

Figure 1: Digestibility of organic matter estimated by HFT, rumen fluid-pepsin method and enzymatic method
Abbildung 1: Verdaulichkeit organischer Substanz geschätzt mit dem HFT, der Pansensaft-Pepsin-Methode und der enzymatischen Methode

are similar to the results, when HFT is compared with ELOS.

Furthermore Figure 1 shows, that there is a larger agreement between rumen fluid-pepsin method and ELOS; the bisecting line represents the total agreement of two methods. The rumen fluid-pepsin method as well as ELOS estimate the DOM – especially of the highly digestible *Brassicaceae* (= stubble turnip and winter forage rape) – higher than the HFT. However the graduation of the different feeds in view of their digestibility mostly agrees. The methods – the rumen fluid-pepsin method and ELOS on the one hand and the HFT on the other hand – show a better agreement, when the DOM is estimated of varieties, which contain higher concentrations of structural substances, like volunteer cereals or some variants of annual ryegrass and summer forage rape "cutting-type" (Table 2). The last mentioned variety contains high concentrations of cellulose, hemicellulose and lignin particular at the early date of sowing (= late July), which is connected with a narrow leaf/stem-ratio. Regarding the annual ryegrass, concerned are the variants grown over a long vegetative period – early date of sowing (= late July) and late date of winter harvest (= late January).

In the second year of investigation some variants of these varieties are more digestible according to the HFT, what could be connected with fungal infection and high concentrations of lignin (Table 2) in this rainy winter. Relating to the lower estimation values according to the HFT, there are several possible explanations, like the drop out of the treatment with pepsin-HCl, whereby proteins are digested and further the DOM increases, because of the solution of hemicelluloses in the acid treatment. BUGHRARA and SLEPER (1986) also estimate lowest DOM by ELOS without previous pepsin-HCL treatment. Furthermore a part of the carbohydrates do not enter in the gas production, measured by the HFT. With the rumen fluid-pepsin method as well as with ELOS these carbohydrates are soluted and measured as *in vitro*-digestible. The volume of gas production is also connected to the ratio of the short chain fatty acids acetate, propionate and butyrate, which are the endproducts of the decomposition of carbohydrates in the rumen. Varieties, which contain larger concentrations of sugar and starch – like stubble turnip and winter forage rape – entail an increase of propionate and butyrate, while cellulose changes the ratio in favour of acetate (KIRCHGESSNER 2004). High concentrations of propionate are connected with a lower gas produc-

Table 2: Crude nutrients [%], structural substances, ergosterol [mg kg^{-1} DM], glucosinolates [µmol g^{-1} DM] and gas production [ml 200 mg^{-1} DM in 24 h], n = 18

Tabelle 2: Rohnährstoffe [%], Gerüstsubstanzen, Ergosterol [mg kg^{-1} DM], Glucosinolate [µmol g^{-1} DM] und Gasbildung [ml 200 mg^{-1} DM in 24 h], n = 18

	Winter forage rape			Summer forage rape			Stubble turnip			Italian ryegras			Volunteer cereals		
	min	$\bar{x}$	max	min	$\bar{x}$	max	min	$\bar{x}$	max	min	$\bar{x}$	max	min	$\bar{x}$	max
CA	10,3	**16,0**	27,2	9,2	**16,0**	29,3	10,4	**13,8**	17,9	11,9	**14,4**	19,2	11,2	**15,7**	23,4
CP	17,2	**21,5**	24,4	13,3	**18,7**	23,9	16,8	**22,0**	25,5	17,5	**22,7**	26,4	21,8	**24,4**	28,0
EE	1,6	**2,3**	2,8	1,3	**2,0**	3,4	1,1	**1,5**	1,9	2,4	**3,4**	4,9	2,0	**2,7**	3,4
CF	12,5	**15,1**	18,5	13,6	**20,2**	28,2	10,6	**13,1**	16,4	19,0	**21,6**	25,4	19,2	**21,5**	24,3
2001/02 NDF	22,3	**32,6**	61,3	24,5	**41,0**	83,5	19,9	**24,6**	31,8	40,0	**52,7**	71,5	45,4	**59,4**	84,2
ADF	16,5	**29,8**	52,5	20,7	**32,6**	70,2	15,8	**21,4**	32,2	21,7	**28,8**	44,1	21,2	**32,1**	49,1
ADL	1,0	**2,0**	4,4	1,3	**2,8**	5,1	1,0	**1,9**	3,0	1,0	**2,2**	6,5	1,5	**2,5**	3,8
Ergosterol	1,0	**32,7**	88,2	1,0	**22,9**	89,7	6,9	**43,2**	128,5	1,0	**56,6**	143,1	1,0	**113,7**	288,0
Glucosinolates	2,8	**8,8**	18,1	2,7	**6,9**	11,0	6,3	**18,5**	33,5	–	–	–	–	–	–
Gas production	37,1	**47,2**	57,7	32,9	**43,6**	53,3	44,1	**51,6**	60,0	35,2	**45,5**	61,9	30,6	**39,5**	45,2
CA	6,4	**10,1**	18,1	4,0	**8,6**	17,9	9,1	**12,8**	15,5	7,5	**10,4**	17,0	4,6	**8,1**	11,9
CP	13,8	**16,4**	20,2	8,1	**11,6**	15,7	10,0	**15,6**	19,4	12,1	**16,6**	21,4	10,0	**14,6**	22,5
EE	1,5	**2,5**	2,9	0,9	**1,8**	2,6	0,8	**1,1**	1,5	2,1	**2,7**	3,5	0,9	**1,7**	2,3
CF	13,2	**16,2**	20,2	16,7	**24,8**	30,8	11,2	**13,0**	14,7	18,0	**23,4**	28,6	21,5	**26,6**	33,5
2002/03 NDF	24,9	**28,8**	33,3	29,2	**39,2**	47,2	21,1	**25,4**	31,8	40,5	**55,1**	68,5	52,8	**61,5**	83,2
ADF	19,7	**23,4**	27,6	24,1	**31,3**	37,2	17,8	**22,0**	27,6	23,5	**29,5**	36,0	27,5	**33,1**	47,3
ADL	1,8	**2,3**	3,0	2,5	**4,4**	6,5	1,1	**1,9**	2,7	1,3	**2,9**	4,9	2,8	**3,9**	4,9
Ergosterol	7,6	**47,9**	101,6	2,1	**21,9**	53,6	3,7	**28,1**	55,3	0,0	**130,3**	376,0	27,7	**102,5**	251,6
Glucosinolates	14,1	**20,9**	30,4	8,6	12,2	18,8	10,3	**17,3**	23,0	–	–	–	–	–	–
Gas production	45,8	**56,9**	62,7	43,7	**50,6**	58,8	53,3	**60,3**	67,6	39,9	**49,3**	55,9	33,6	**43,8**	53,5

tion; contrarily the gas production increases with high concentrations of acetate (BEEVER and MOULD 2000; WILLIAMS 2000). According to HOFFMANN et al. (1972) feeds rich in carbohydrates and poor in crude fibre can cause disturbances of normal rumen activity and fermentation involving a loss of energy. Such a loss of energy, what particularly can be important to stubble turnip and winter forage rape, is not recorded by ELOS. Add to this that digestibility of more than 90% seems to be improbable for winter grown forage investigated in this research. Another difference between the methods is the regression equation used in the HFT. This equation is calibrated and recommended for all types of feed and could implicate an overestimation of low energy feeds and an underestimation of feeds with a higher energetic value, because the regression coefficients are a compromise for these different types of feed; this explains the small variation between the values measured with the HFT (MENKE und STEINGASS 1987). The higher estimated values with ELOS in opposite to the rumen fluid-pepsin method can be connected with specific effects of secondary compounds or fungal infection, here enzyme react less sensitiv than rumen microbes (OPITZ V. BOBERFELD et al. 2003). The comparison of the energy values, estimated with the HFT and with the chemical method, yields no agreement between these methods (Figure 2). The disadvantage of the chemical method is the insufficient classification of the carbohydrates by the fraction of crude fibre and further the poor subdivision of the varieties of forage rape in the DLG-table (ANONYMUS 1997b); the DOM of the investigated types of forage rape differ clearly, because of variations in leaf/stem-ratio (BERENDONK 1982; NEFF 2005; OPITZ V. BOBERFELD und NEFF 2006).

4 Conclusion

For the prediction of digestibility of organic matter *in vitro*-methods with rumen fluid are evidently more suitable than enzymatic or chemical methods. Especially in the case of fungal infection or for plants containing secondary compounds the estimation is more accurate, because the activity of rumen microbes can be influenced. The chemical method is not reliable for predicting digestibility of organic matter. Regarding the Hohenheim gas test it is recommendable to create new equations, in consideration of *Bras-*

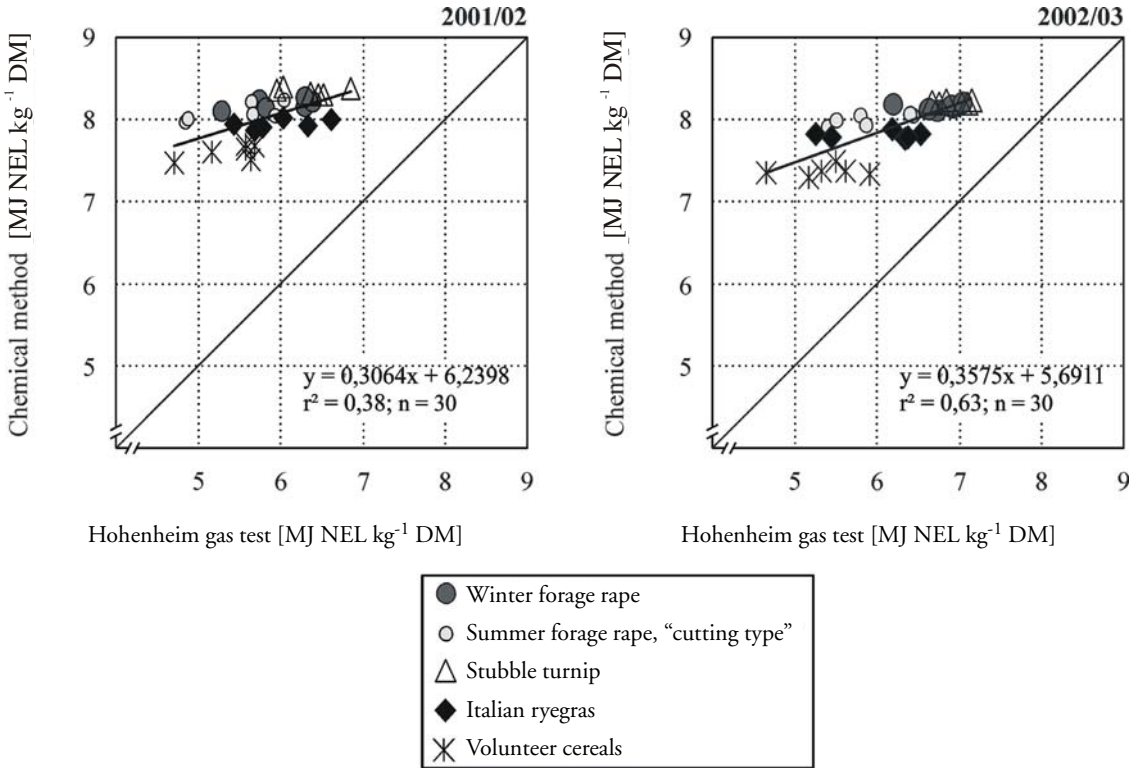

Figure 2: Energy value [MJ NEL kg^{-1} DM] estimated by HFT and chemical method
Abbildung 2: Energiekonzentration [MJ NEL kg^{-1} TM] geschätzt mit dem HFT und der chemischen Methode

sicacea, as the specific effects of this group of feeds rich in carbohydrates and containing secondary compounds do not flow into the regression equations up to now.

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