Effects of Barley and Sugar-beet Pulp on Digestibility, Purine Excretion and Blood Parameters in Horses

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Summary

Sugar-beet pulp (SBP) was included at 0, 20 and 30 % in a hay: barley diet given to 3 trotters in a 3*3 Latin-square design. The apparent digestibilities of organic matter (DOM), neutral detergent fiber (NDF), nitrogen (N) and energy (DE) were not significantly affected by the change of diet. There were however indications of a decline in both DOM and DE at the highest inclusion level of SBP. With increasing intake of NDF the faecal excretion of N and the bacterial marker diaminopimelic acid (DAPA) increased while the urinary excretion of the potential microbial marker allantoin decreased. These contrasting excretion patterns could indicate a reduced absorption of microbial amino acids in the large intestine.

Wirkung von Gerste und Trockenschnitzeln auf Verdaulichkeit, Purinausscheidung und Blutparameter bei Pferden

Zuckerrübendiffusionsschnitzel (SBP) wurden einer Heu/Gerste Ration zugelegt (0, 20 und 30 %) und mit 3 Trabern im Verdaulichkeitsversuch geprüft (3 x 3 lat. Quadrat). Die scheinbare Verdaulichkeit der organischen Substanz (DOM), neutralen Detergentienfasern (NDF), des Stickstoffs (N) und der Energie (DE) wurde durch die Zulage nicht beeinflußt. Allerdings bestand tendenziell für DOM und DE ein Abfall bei der höchsten Zulagestufe. Mit steigender Aufnahme an NDF nahm die faecale N-Abgabe zu. Hierbei wurde durch einen Anstieg der Menge an Diaminopimelinsäure (DAPA) im Kot eine forcierte mikrobielle Aktivität angezeigt, andererseits fiel die renale Ausscheidung des Allantoins ab. Die gegensätzliche Veränderung dieser Ausscheidungen könnte auf eine eingeschränkte Absorption mikrobiell fixierten Proteins aus dem Dickdarm hinweisen.

Introduction

A large part of the ration carbohydrates escapes enzymatic digestion in the horse small intestine (*Argenzio*, 1975) and is subjected to microbial fermentation in the caecum and colon (*Stevens* et *al.*, 1980). The fermentation in the hindgut of the horse and in the reticulo-rumen of ruminants show many similarities (*Stevens* et *al.*, 1980) and is of importance for the energy utilization in both species. Not only the energy utilization but also the protein nutrition is affected by the activities in the hindgut. *Slade* et *al.* (1971) have shown that part of the amino acid supply in the horse is of microbial origin due to uptake from the hindgut.
In ruminants the urinary excretion of the purine derivative allantoin can be used to estimate the microbial protein supply (*McAllan*, 1982; *Lindberg* et *al.*, 1989; *Chen* et *al.*, 1991). Considering the high microbial activity in the hindgut of the horse and indications of absorption of microbial amino acids (*Slade* et *al.*, 1971) the excretion of purine derivatives could possibly also be an indicator of microbial amino acid supply in the horse.
The aim with the present experiment was to study the effect of hindgut fermentation on fiber utilization and excretion of microbial markers.

Material and Methods

Animals:
Three geldings of the Swedish Trotter Breed (7 – 10 years old) with an average initial live weight of 487 (SD 20.8) kg were used.

Feeding and diets:
The horses were fed twice daily at 8 and 15 o'clock with half the daily allowance at each feeding. The diets were composed (on dry matter [DM] basis) of hay : barley (57 : 43; diet A), hay : barley : sugar beet pulp : molasses (58 : 19 : 20 : 3; diet B) and hay : barley : sugar beet pulp: molasses (54 : 11 : 30 : 5; diet C). Minerals were given once daily and water was available at all times.

Experimental design:
The diets were fed according to a 3*3 Latin-square design. The adaptation period to a new diet was 14 d followed by total collections of faeces and urine for 3 d. Blood samples were taken from the jugular vein on the last day in each collection period.

Collection of samples:
The horses were kept in a metabolic stall during the collections of faeces and urine. Faeces was collected twice daily and frozen immediately. Prior to analysis the faeces was minced in a large meat mincer, mixed, subsampled and freeze dried. Urine was collected in 10 % sulphuric acid to keep pH below 3. Samples were stored at – 20 °C until analysis. Blood was collected immediately before feeding in heparinated tubes kept on ice. The samples were centrifuged immediately and the plasma was kept at – 20 °C until analysis.

Chemical analysis:
Total N was determined with a conventional macro-Kjeldahl method (*Nordisk Metodikkommitté*, 1976). Neutral detergent fibre (NDF) in hay, sugar beet pulp and faeces was analysed according to *van Soest* and *Wine* (1967) and in barley according to *Robertson* and *van Soest* (1977). Allantoin in urine samples was determined with high pressure

Table 1: Intake (g/d) of dry matter, NDF, nitrogen, and gross energy (MJ/d).

	Diet			
	A	B	C	SEM
Dry matter	6053	5880	6317	98
NDF	2515[a]	2816[b]	3112[c]	37
Nitrogen	111[a]	102[b]	109[a]	0.8
Gross energy	112	106	113	1.1

Means with different superscripts within rows differ significantly ($P<0.05$).

liquid chromatography (HPLC) using a ion-exchange (Ion-300, Tillquist, Stockholm, Sweden) column *(Jacobsson, Juremalm, and Lindberg,* unpubl.). Other purine derivatives and creatinine was analysed with HPLC using a Nova-Pak C 18 column (*Lindberg* et *al.,* 1989). Diaminopimelic acid (DAPA) in faeces was analysed according to *Czerkawski* (1974). Plasma samples were analysed for glucose and urea with standard methods at the Department of Clinical Chemistry, Swedish University of Agricultural Sciences. Plasma insulin was assayed by insulin radio-immunoassay at Equilab, Halmstad, Sweden. Gross energy in feed and faeces was determined using an iso-thermal bombcalorimeter and gross energy in urine with a adiabatic bombcalorimeter.

Statistical analysis:
Analysis of variance performed according to a Latin-square design (*Dunn* and *Clark,* 1974) using the GLM procedure (*SAS,* 1988).

Results and Discussion

There were no significant differences in DM and gross energy intakes between diets (table 1). Intake of N was significantly ($P < 0.05$) lower on diet B than on diets A and C. NDF intake differed significantly ($P < 0.05$) between diets and increased with inclusion of sugar-beet pulp (SBP). Despite the change in diet carbohydrate composition the plasma concentrations of glucose and insulin was not significantly affected. Also the concentration of plasma urea was unaffected by the diet. The average plasma concentrations of urea (mmol/l), glucose (mmol/l) and insulin (pmol/l), immediately before the morning feeding, were 4.4 (SD 0.7), 5.2 (SD 0.5) and 106 (SD 43) on diet A; 4.2 (SD 0.8), 5.5 (SD 0.3) and 76 (SD 13) on diet B; and 4.0 (SD 0.1), 5.1 (SD 0.6) and 79 (SD 22) on diet C.

Table 2: Digestibilities (%) of organic matter, NDF, nitrogen and energy, and the content of digestible energy (DE; MJ/kg dry matter) in the diets.

	Diet			
	A	B	C	SEM
Organic matter	76	77	74	2.3
NDF	63	71	69	3.6
Nitrogen	75	70	63	2.5
Energy	73	73	70	2.2
DE	13.5	13.2	12.4	0.53

Table 3: Urinary excretion (mg nitrogen [N]/d) of allantoin, uric acid, xanthine and creatinine, and faecal excretion (mg/d) of diaminopimelic acid (DAPA).

	Diet			
	A	B	C	SEM
Allantoin N	4 657	3964	2983	197
Uric acid N	957	740	672	88
Xanthine N	684	735	557	61
Creatinine N	11 443[a]	9097[b]	6925[c]	188
DAPA	549	674	753	36

Means with different superscripts within rows differ significantly ($P<0.05$).

The substitution of barley by SBP decreased the intake of starch and increased the intake of fermentable fibers. This should be expected to increase the hindgut fermentation (*Stevens* et *al.,* 1980).
Despite the marked increase in NDF intake its digestibility was not significantly affected. This indicates a high potential of the horse to utilize fermentable fibers in the hindgut. It should be noted, however, that there was a non-significant decrease in OM and energy digestibilities at the highest inclusion level of SBP (table 2). This also resulted in a non-significant decrease in the DE content of diet C.

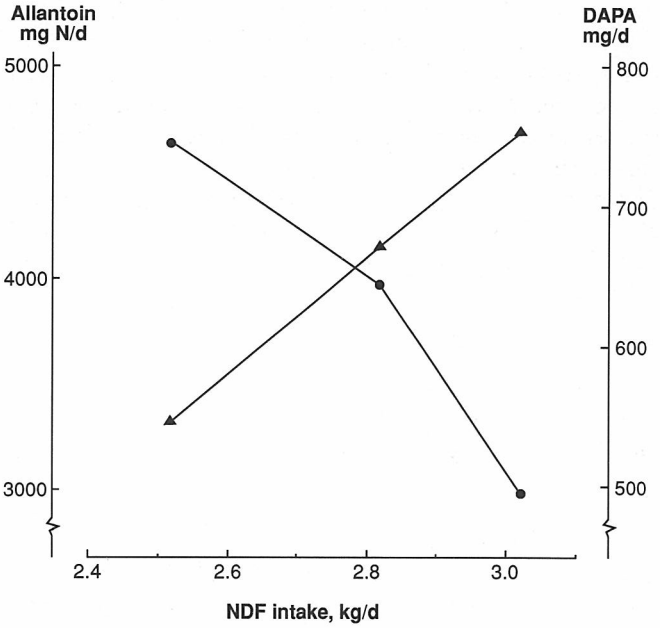

Fig. 1: The influence of increasing intake of neutral detergent fibre (NDF) on the urinary excretion of allantoin (●, mg N/d) and the faecal excretion of diaminopimelic acid (DAPA; ▲, mg/d) in horses.

The increase in NDF intake resulted in a marked but non-significant reduction in the digestibility of N (table 2). The increase in excretion of faecal N indicates an increase in hindgut fermentation in agreement with observations made in pigs by *Gargallo* and *Zimmerman* (1981). The simultaneous increase ($P < 0.09$) in faecal excretion of the bacterial marker DAPA (table 3) and faecal N supports this suggestion.

The urinary energy loss (% of DE) did not differ significantly between diets and was on average 5.3 (SEM 0.4). The urinary N loss was not significantly affected by the diet and was on average 65.8 (SEM 5.7) g per day.

The urinary excretion of allantoin, on the other hand, decreased with inclusion of SBP ($P < 0.11$), while the urinary excretion of uric acid and xanthine was unaffected by the change of diet. Also the creatinine excretion was significantly ($P < 0.05$) reduced with increasing inclusion of SBP.

The urinary excretion of allantoin ranged from 29 to 45 mg N/kg metabolic body weight ($W^{0.75}$). This is 5 – 8 times higher than the endogenous allantoin excretion in sheep and 2 – 3 times higher than the endogenous allantoin excretion in cattle (Chen at al., 1991). If the endogenous allantoin excretion of the horse is comparable to that of ruminants the observed urinary excretion indicates an exogenous supply of nucleic acids. The exogenous nucleic acids could be of microbial origin.
With increasing intake of NDF and an increasing faecal excretion of the bacterial marker DAPA there was a non-significant decrease in the urinary excretion of allantoin (fig. 1). These contrasting excretion patterns could possibly indicate a reduced absorption of microbial amino acids in large intestine.

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