Methodology to studying digestibility in vivo

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Introduction

Digestion trials in horses are commonly accomplished to prove feed-inert effects on digestibility and to determine the content of digestible nutrients or digestible energy in the feed. For this it is essentially necessary to guarantee that the experimental ration has no adverse effect on the gut's health and digestive physiology so that the measured digestibility is affected mainly by influences which can be returned directly to the feed and its ingredients ('principle of physiological addition', PPA). How the combination of concentrates and roughage may affect the digestibility of the individual feed stuffs has been demonstrated by *Thompson* et al. (1987) and *Fehrle* (1999). As a consequence, to measure the digestibility of concentrates, mostly the difference method is needed. Merely when PPA works, experimentally determined digestibility of different feed stuffs can be summed up without significantly influencing each another ('principle of mathematical addition', PMA). Then, results from digestion trials can be generalised. To revise special or extreme effects, PPA and PMA may be neglected. However, such results are not suitable do describe the feed value. Thus, digestibility should be measured under standardised conditions. To create an uniform methodology the following aspects are of interest: Feeding conditions to prevent digestive disorders, relevant effects on digestibility and special methodical questions.

Feeding conditions

To prevent digestive disorders, the following is recommended for adult horses: The ration should contain 18-35% of crude fibre and <8% crude fat in the dry matter and should provide a structure that induces intense chewing (*Fehrle* 1999, *Zeyner* et al. 2000, *Zeyner* and *Kienzle* 2002, *Zeyner* et al. 2004). Concentrates shell not exceed 60% oft the diet (*Hoffmann* and *Fuchs* 1988). The supply of crude protein may amount to 90-150% of the individual recommendation, but should stay consistently during the study period. Despite a high individuality must be respected, the pH value in the faecal water of horses should not lie outside 6 to 7 (*Graß* 1995).

Relevant effects on digestibility

Influences on digestibility can be fundamentally discriminated by means of origin. Factors like breed, age, sex, reproductive and mental state, health and undefined individual patterns are of endogenous origin. Feed (composition, treatment, structure, quantity, energy level, feeding frequency, feeding sequence, sensoric patterns), stabling, management and exercise are exogenous. The feed itself is commonly the factor in question. Despite that, age (*Turcott* et al. 2004), reproductive state (*Davison* et al. 1991), breed (*Hoffmann* and *Fuchs* 1983, *Hoffmann* et al. 1987, *Fuchs* and *Hoffmann* 1988) and exercise (*Orton* et al. 1985, *Zeyner* et al. 2003b) seem to have an influence in horses and need to be standardised or at least clearly defined as individual study conditions. Further, an interaction between breed and ration type seems to exist (*Fuchs* and *Hoffmann* 1988). Thus, for digestion trials it can be recommend to use healthy adult but not aged (*Ralston* et al. 1989) horses, to generalise results only within the type of breed the animals used are coming from and to exercise the horses slightly during the study period. Whether horses are subjected to an exercise protocol they should meet the respective requirements always prior to the study (*Worth* et al. 1987). The feeding level affects gastrointestinal transit (*Miraglia* et al. 1992) and digestibility (*Todd* et al. 1987, *Ott* 1981, *Martin-Rosset* and *Dulphy* 1987) to a lower extent than expected, especially in comparison to ruminants.

Special methodical questions

Special methodical questions deal with the stabling of the horses, the trial protocol (character and duration of sub-periods, use of markers, sampling procedure), the handling of sampled faeces and analytical problems. One of the most essential questions is the necessary trial length, regarding feed adaptation and the yield of representative faecal samples. To get sure results the intestine, including microbiota, must be carefully and completely adapted (*Julen* et al. 1995, *Zeyner* et al. 2000, 2003a). The time needed to do so depends from the difference in the composition of the ration fed in the pre-study and study period. In most cases 7 days to change the feed and 14 days to equilibrate seem to be appropriate prior to faecal collection (*Fuchs* et al. 1987, *Zeyner* et al. 2000, 2003a). Despite pelleted and nonpelleted feed pass the digestive tract of horses completely within 84 hours after feeding (*Hintz* and *Loy* 1966), a faeces sampling period of 3 (*Goachet* et al. 2005) and 5 days (*Fuchs* et al. 1987), respectively, for total collection is recommended. When 4 N HCl insoluble ash (IA) is used as marker and faeces is sampled at several times a day, the sampling period can be reduced to 3 days (*Goachet* et al. 2005), but should stay at 5 days when faeces are only sampled in the morning (*Fuchs* et al. 1987). Markers are especially important for digestion trials with horses because by use of this method they can easily subjected to exercise. Despite others, IA seems to be a proper marker in horses (*Schurg* et al. 1977, *Sutton* et al. 1977, *Orton* et al. 1985, *Zeyner* et al. 2004a, *Goachet* et al. 2005). Contrary, lignin of any analytical origin do not work because side chains of aromatic cycles like methyl groups can split by anaerobic gut microbes (*Bergner* 1996) and thus alter the molecules. An other relevant problem deals with the sampling procedure and the pre-analytical preparation of faeces to ensure that nitrogen-losses not exceed minimal amounts (*Fuchs* et al. 1987, *Coenen*, unpublished data).

Conclusions

In vivo determined digestibility can only be generalised to describe the feed value when the experimental feeding con-

ditions were without negative effect on digestive physiology. Despite that, numerous factors may affect digestibility and special methodical questions must be taken into account. Thus, it is recommended to standardise a method to predict digestibility in horses.

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