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Short communication

Fermentative capacity of equine faecal inocula obtained from clinically normal horses and those predisposed to laminitis

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ABSTRACT

The aim of the study was to assess the fermentative capacity of faecal inocula obtained from 14 mature grass-kept horses; 7 clinically normal horses (NOR) and 7 that were predisposed to laminitis (LAM). Freshly voided faeces were collected from each animal and stored at −20 °C prior to transportation on ice to the laboratory where they were used as inocula for *in vitro* digestibility determinations, using the gas production technique of Theodorou et al. (1994), to assess the ability of each inoculum to ferment grass hay, starch or inulin. Gas production curve fitted parameters; asymptotic gas production (*A*), half time of asymptotic gas production (*B*), inflection point (t_I) and maximal fractional rate of substrate degradation (MFR) were similar for the two incoula sources; NOR and LAM. With the exception of pH, which was lower ($P<0.001$) in bottles inoculated with LAM compared to NOR (6.50 *vs.* 6.89, respectively), analysis of vessel contents following fermentation also showed no difference between inocula groups. Consequently, it would appear that there is no difference in the fermentative capacity of inocula obtained from clinically normal

Abbreviations: *A*, asymptotic gas production; *B*, half time of asymptotic gas production; DML, dry matter loss; GI, gastrointestinal; GP, gas production; LAM, horses predisposed to laminitis; MFR, maximal fractional rate of substrate degradation; NOR, horses with no previous history of laminitis; t_I, inflection point; TVFA, total volatile fatty acid; VFA, volatile fatty acid.

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horses and those predisposed to laminitis. Furthermore, the GP technique appears to be valuable tool for evaluating the fermentative capacity of equine faecal inocula.

1. Introduction

Laminitis has widespread implications for equine welfare (Hinkley and Henderson, 1996). There are many aetiological factors associated with equine laminitis; however, pasture-induced laminitis appears to be the most common in the UK (Slater et al., 1995). One uncertainty about laminitis is why only certain individuals appear to be predisposed, even when grazing identical pastures? One of the most credible hypothesis is that differences in individual susceptibility might occur at the level of the large intestine (Bailey et al., 2004), with possible differences in the bacterial populations. Despite the importance of intestinal microbial populations in host animal health, there is a dearth of information on the microbial ecology of the equine hindgut. While microbial diversity has an important role to play in hindgut function and disease, the effect this has on the ability to degrade certain feedstuffs is of equal importance. Studies have reported large variations in caecal fermentation parameters between ponies fed similar diets (McLean et al., 2000; Moore-Colyer et al., 2000; de Fombelle et al., 2001) and it has been hypothesised that these differences may be attributable to microbial diversity, which may be particularly evident in animals with a history of laminitis. Nevertheless, *in vivo* investigation of this requires surgically modified animals; thus the *in vitro* gas production (GP) technique of Theodorou et al. (1994) has been used to assess the effects of feedstuffs on the large intestinal environment (McLean et al., 1997; Murray et al., 2005a,b) and to assess the fermentative capacity of equine faecal inocula as an indicator of hindgut microbial activity (Murray et al., 2006). Consequently, the aim of the experiment reported here was to examine the ability of faecal inocula obtained from clinically normal horses and those predisposed to laminitis to ferment grass hay, starch or inulin using the *in vitro* gas production technique of Theodorou et al. (1994).

2. Materials and methods

2.1. Experimental design

Fourteen mature grass-kept (maintained at grass 24 h a day) horses from the International League for the Protection of Horses in Norfolk provided the faecal inocula for the study. Faecal samples were collected from 7 clinically normal horses (NOR) and from 7 that were predisposed to laminitis (LAM) during the month of July 2005. Horses predisposed to laminitis were included in the study if they were diagnosed with acute laminitis 3 or more times during the preceding 3 years. Freshly voided faeces were collected from each animal and stored at −20 °C prior to transportation on ice to the laboratory where they were used as inocula for *in vitro* digestibility determinations, using the gas production technique of Theodorou et al. (1994), to assess the ability of each inoculum to ferment grass hay, starch or inulin. The distance between the animals and the laboratory necessitated the need for storage of the inocula at −20 °C and for transportation on ice.

2.2. In vitro gas production

The methods employed for the gas production technique were as described by Theodorou et al. (1994), with the exception of the preparation of the microbial inocula. Each sample of frozen faeces was defrosted and subsequently incubated to a temperature of 38 °C. Each faecal inoculum was combined with culture medium in a ratio of 1:2, and macerated in a blender (Kenwood BL300, Kenwood Ltd., Harant, Hants, UK) for 20 s. The resultant inoculum was strained through a triple layer of muslin and dispensed under CO_2 immediately after extraction.

Each faecal inoculum (14 in total) was used to inoculate three identical series of bottles containing 1 g dry matter (DM ± 0.5%) of either hay (ground to pass through a 1 mm dry mesh screen), soluble