



Short communication

In vitro ruminal fermentation of ground and dry-rolled barley grain differing in starch content



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ABSTRACT

In vitro ruminal fermentation of ground and dry-rolled barley samples differing in starch content was evaluated using a batch culture technique. The study was arranged in a 2 (low and high starch) × 2 (ground and dry-rolled) factorial design. Gas production (GP), short chain fatty acids (SCFA), dry matter (DM) and starch disappearance were estimated at 3, 6, 12 and 24 h of incubation using rumen fluid from 3 ruminally fistulated beef cattle. Rate of GP was greater ($P < 0.05$) in both high starch and ground (2 mm) barley samples. Kinetics of DM and starch disappearance were calculated using the equation of $a + b(1 - e^{-c(t-L)})$. Starch content × processing interactions were noted for the b fraction and rate of DM disappearance. Consistently, both high starch and ground barley samples had greater ($P < 0.05$) a and b fractions, rate constant of disappearance of b fraction and effective degradability versus low starch and dry-rolled samples. Expectedly, molar proportions of individual and total SCFA were greater ($P < 0.05$) in the ground barley samples at all incubation periods. Overall, starch content (high versus low) had significant effect on the rate of GP and constant rate of DM disappearance but no effect on the constant rate of starch disappearance.

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1. Introduction

Barley grains from different sources are diverse in their chemical composition and estimated metabolizable energy contents due to geographical, environmental and genetic variations as well as their interactions (Dehghan-banadaky et al., 2007). Differences in digestibility explain most of the variation in energy content of a feed. This inherent variability in barley chemical composition leads to differences in animal performance (Yang et al., 2013). Digestibility of whole barley grain is limited by its fibrous hull and intact pericarp (Beauchemin et al., 1994) so processing is required to make the starch accessible

Abbreviations: ADF, acid detergent fiber; CP, crude protein; CV, coefficient of variation; DM, dry matter; DMD, dry matter disappearance; ED, effective degradability; GP, gas production; GV, gas volume; aNDF, neutral detergent fiber assayed with a heat stable amylase and expressed inclusive of residual ash; PI, processing index; SCFA, short-chain fatty acids.

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to microbes in the rumen. Compared with intact barley grain, processing changes both its rate and extent of degradation in the rumen. Because of rapid digestion of barley starch in the rumen, we hypothesized that starch content of barley grain can influence both rates of gas production (GP) and dry matter (DM) disappearance (DMD). The objectives of this study were to determine the effects of barley grains differing in starch content and processing (ground versus dry-rolled) on kinetics of GP, DM and starch digestibility and concentrations of short-chain fatty acids (SCFA) using a batch culture technique.

2. Materials and methods

2.1. Barley sample collection and processing

Barley samples were collected from 10 different locations in Southern Alberta. One hundred and seventy samples were collected monthly from November 2012 to March 2014. All the samples were analyzed and ranked according to their starch content and the lowest 5 samples (ranging from 539 to 590 g/kg DM and hereafter designated low starch) and highest 5 samples (ranging from 651 to 671 g/kg DM and hereafter high starch) were selected. Subsequently, these 10 samples were divided into 2 equal parts: one part was ground through a 2 mm screen (Wiley mill; Arthur H. Thomas Company, Philadelphia, PA, USA), and the other part was dry-rolled using a laboratory scale roller (Model R250.6, Kal Rob Machining, Picture Butte, AB, Canada) with rollers set at different distances in order to achieve an extent of processing expressed as processing index (PI) 0.75 ± 0.03 for each samples. Processing index was calculated as the bulk density of barley grain after rolling divided by the bulk density before rolling. Overall, 20 samples were used for the *in vitro* batch culture study.

2.2. *In vitro* incubations

Approximately 3 g of ground or dry-rolled barley was weighed in duplicate into 500 ml Ankom gas production module (RF1; a computerized system with automated pressure transducers, Ankom Technology, Macedon, NY, USA). Ruminant fluid was collected 2 h after feeding (0900 h) from three ruminally fistulated beef cattle (650 kg body weight) fed *ad libitum* a diet consisting of whole crop barley silage (700 g/kg), dry-rolled barley grain (270 g/kg), and vitamin and mineral supplement (30 g/kg) on a DM basis. All animal procedures were in accordance with the guidelines of the [Canadian Council on Animal Care \(2009\)](#). Sampling and handling of rumen fluid were as described previously ([Anele et al., 2014](#)). The pH of ruminal fluid was measured immediately (B20PI, SympHony Benchtop Meters; VWR, Edmonton, AB, Canada) and ranged from 5.78 to 6.12 throughout the study. Each Ankom gas production module received 270 ml of McDougall's buffer and 90 ml of strained ruminal fluid (3:1 ratio), after which each module was flushed with oxygen free CO₂ and sealed. Modules were incubated in an oscillating shaker at 39 °C with an oscillation speed of 125 rpm for 3, 6, 12 and 24 h. The whole process was repeated on a different date (second run) to have four analytical replicates (*i.e.* two per run). In addition, 2 blanks containing 360 ml of medium only were included for each incubation time.

Gas data obtained were fitted to exponential model ([Ørskov and McDonald, 1979](#)) as:

$$y = B(1 - \exp - c \times [t - \text{lag}]),$$

where 'y' is the cumulative volume of gas produced at time 't' (h), 'B' is the asymptotic gas volume, 'c' is the rate constant and 'lag' is the time (h) between inoculation and commencement of GP. Initial GP rate (Abs_{g}) was calculated as the product of asymptotic cumulative gas volume and rate of fermentation ([Larbi et al., 1996](#)). After incubation, bottles were placed in ice to stop fermentation. Undigested substrate was determined by high-speed centrifugation ($20,000 \times g$) of incubation residues at 4 °C for 30 min. Blanks were also centrifuged and pellet weighed and used to correct for residues from the ruminal inoculum. *In vitro* DM disappearance coefficient was calculated as:

$$\frac{[\text{Substrate incubated} - (\text{substrate pellet} - \text{blank pellet})]}{\text{substrate incubated}}$$

The same process was used to estimate starch disappearance. Kinetics of DM and starch disappearance were calculated using the equation of [McDonald \(1981\)](#):

$$y = a + b(1 - e^{-c(t-L)}) \text{ for } t > L,$$

where y, disappearance at time t; a, an intercept representing the proportion of DM/starch solubilized at initiation of incubation (soluble fraction); b, the fraction of DM/starch insoluble but degradable in the rumen; c, a rate constant of disappearance of fraction b; t, time of incubation and L, lag phase. The non-linear parameters a, b, c and L were estimated by an iterative least squares procedure ([SAS, 2002](#)). The effective degradability (ED) of DM and starch was calculated using the following equation ([McDonald, 1981](#)) with the modification of [Wulf and Südekum \(2005\)](#), which assumes no degradation occurs during the lag phase:

$$\text{ED} = a + \left(\frac{bc}{c+k} \right) \times e^{-kt},$$

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