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# Molecular structure, chemical and nutrient profiles, and metabolic characteristics of the proteins and energy in new cool-season corn varieties harvested as fresh forage for dairy cattle

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# ABSTRACT

To our knowledge, no previous research exists concerning the molecular structure and metabolic characteristics of the proteins and energy that new cool-season corn varieties provide for dairy cattle. The objectives of this study were to identify the differences in the molecular structures of proteins among several new coolseason corn varieties [Pioneer P7443R, Pioneer P7213R, Pioneer P7535R (Pioneer Hi-Bred International Inc., Johnston, IA), Hyland Baxxos RR, Hyland SR22, and Hyland SR06 (Hyland Seeds, Blenheim, ON, Canada)] using Fourier transform infrared attenuated total reflectance (FT/IR-ATR) molecular spectroscopy, and to determine the nutrient profile and supply that each variety provided for dairy cattle. The protein molecular structure studies showed that the amide I to amide II ratio ranged from 1.09 to 1.66 and that the  $\alpha$ -helix to  $\beta$ -sheet ratio ranged from 0.95 to 1.01 among the new cool-season corn varieties. Energy content was significantly different among the new varieties. We found significant differences in the protein and carbohydrate subfractions and in the ruminal degradation kinetics of the organic matter, crude protein, starch, and neutral detergent fiber of the new varieties. The new varieties had similar estimated intestinal digestibilities for rumen undegraded crude protein. However, the new varieties had significant differences in predicted total truly absorbable protein, ranging from 39 to 57 g/kg of dry matter, indicating that these newly developed varieties are satisfactory sources of truly absorbed protein for dairy cattle. Further study on the molecular structure profiles of cool-season corn in relation to its nutrient utilization and availability in dairy cattle is necessary. Key words: forage, protein molecular structure, metabolic characteristics, energy

# INTRODUCTION

Corn is a crop with a long history as a foodstuff for both animals and humans (Lauer et al., 2001; Arturo, 2003), and 40% of current global corn production is used as animal feed (Gyori, 2010). Canada harvests over 190,000 ha of corn forage, with the highest production being in Ontario (63%) and the second highest (21%) in the province of Quebec (Coors and Lauer, 2001).

The corn grown in the Canadian prairies is different from the corn varieties grown in warmer climates (Lassiter et al., 1958). The main differences are due to the shorter growing season and lower growing temperatures in the Canadian prairies compared with the areas of warm-season corn production, such as the United States (Lauer et al., 2001). The differences among varieties include changes in chemical profile and the nutrient composition of silage (Mahanna, 2010).

In corn cultivation, crop heat units (CHU) are calculated from daytime temperatures above 10°C and nighttime temperatures above 4.4°C on a cumulative daily basis from seeding to harvest. Many corn varieties for western Canada require >2,000 CHU to reach the silage harvest stage, with a kernel maturity of 45% DM.

Recently, 6 cool-season corn varieties have been developed: Pioneer P7443R, Pioneer P7213R, Pioneer P7535R (Pioneer Hi-Bred International Inc., Johnston, IA), Hyland Baxxos RR, Hyland SR22, and Hyland SR06 (Hyland Seeds, Blenheim, ON, Canada). However, no systematic research has been conducted on the molecular structure, chemical and nutrient profiles, and metabolic characteristics of the protein and energy of these newly developed cool-season corn varieties.

Fourier transform infrared attenuated total reflectance (**FT**/**IR-ATR**) molecular spectroscopy is able to detect molecular structural features in biological materials, as well as processing-induced structural changes (Doiron et al., 2009; Liu and Yu, 2010). The hypotheses of this study were that (1) the differences in the molecular structures of the proteins among the new cool-season corn varieties could be detected by

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molecular spectroscopy, (2) the magnitude of the differences among these new cool-season corn varieties was significant, and (3) the chemical and nutrient profiles of the newly developed cool-season corn varieties significantly differ from those of corn varieties grown in warm climates.

The objectives of this study were (1) to identify differences in the molecular structures of proteins among the new cool-season corn varieties using FT/IR-ATR molecular spectroscopy; (2) to investigate the differences in chemical profile, as well as the protein and carbohydrate subfractions, among the cool-season corn varieties; (3) to determine the ruminal degradation kinetics of various nutrients among the cool-season corn varieties; (4) to predict the intestinal availability of protein among the cool-season corn varieties; and (5) to reveal the metabolic characteristics of the proteins and model the amount of truly absorbable protein in the small intestine.

### MATERIALS AND METHODS

# Corn Cultivation, Experimental Design, and Sampling

Six new cool-season corn varieties were grown at the Canada-Saskatchewan Irrigation Diversification Centre (Outlook, SK, Canada) from May to September 2011. The varieties included Pioneer 7443R, Pioneer 7213R, Pioneer 7535R, Hyland Baxxos RR, Hyland SR22, and Hyland SR06. The experimental design was a randomized completed block design. The cultivation was designed with 4 replicates (4 fields or blocks) with a total of 24 plots for the 6 varieties. Seeding was performed on May 20, 2011, and harvesting (and sampling) was performed on September 29, 2011, after 2,160 CHU had been reached.

# Molecular Spectroscopy

The molecular spectral data of the corn samples were collected and corrected for the background spectrum using FT/IR molecular spectroscopy (Jasco 4200, Jasco International Co. Ltd., Tokyo, Japan). The spectra were generated for the mid-infrared region (approximately  $4,000-800 \text{ cm}^{-1}$ ) and the fingerprint region (approximately  $1,800-800 \text{ cm}^{-1}$ ) with a spectral resolution of 4 cm<sup>-1</sup>. The FT/IR spectral data were processed using Omnic 7.3 (Spectra-Tech, Madison, WI). The regions of specific interest in this study included the protein amide I and II and the protein structure of the  $\alpha$ -helix and  $\beta$ -sheet in the infrared regions of approximately  $1,715-1,480 \text{ cm}^{-1}$  (Samadi and Yu, 2011; Liu et al., 2012).

### Univariate Spectral Analysis

The protein molecular structure spectral profile was determined from the 2 primary bands in the spectra; namely, the amide I and amide II regions (Yu, 2010; Khan and Yu, 2013). The amide I and amide II peak area absorption intensities and their ratios were calculated. Using the second-derivative functions in Omnic 7.3, the amide I peak was further resolved into several multi-component peaks in which  $\alpha$ -helices (centered at ~1,655 cm<sup>-1</sup>) and  $\beta$ -sheets (centered at ~1,630 cm<sup>-1</sup>) were identified. The intensities of the peak heights for the  $\alpha$ -helix and  $\beta$ -sheet were calculated.

#### **Rumen Degradation Procedures**

The rumen in situ degradation parameters were determined using the method described previously (Yu et al., 2002). Fresh forage samples were chopped to 1 cm in size, dried at 55°C for 72 h and ground through a Christy & Norris 10-inch feed mill (Christy Turner Ltd., Suffolk, UK) using a 2-mm screen. Approximately 7 to 9 g of each dried forage sample was weighed into a nylon bag (Nitex 03-41/31 monofilament open mesh fabric, Screentec Corp., Mississauga, ON, Canada) with dimensions of  $10 \times 20$  cm and a pore size of  $41 \,\mu\text{m}$ . The ratio of the sample size to the bag surface area averaged  $\sim 17.5 \text{ mg/cm}^{-2}$ , similar to that used in previous work (Ørskov and McDonald, 1979; Nocek and Tamminga, 1991). A polyester mesh bag  $(45 \times 45 \text{ cm with a 90-cm})$ length of nylon rope for anchoring to the cannula) was used to hold the sample bags in the rumen. The sample bags were added to the polyester mesh bag according to a gradual addition-all out schedule and incubated for 72, 48, 24, 12, 6, 2, and 0 h. The number of bags incubated for each sample was determined according to the previous work (Bal et al., 2000; Jurjanz and Monteils, 2005; Zanton and Heinrichs, 2009). The maximum number of bags in the rumen at any one time was 30 based on previous effective degradability data (Huntington and Givens, 1995). All samples were incubated for 2 runs in 3 nonlactating Friesian cows fitted with rumen cannula and fed 570 g/kg of barley silage, 100 g/ kg of alfalfa hay, 50 g/kg of dehydrated alfalfa pellets, and 280 g/kg of concentrates (containing barley, wheat, oats, canola meal, soybean meal, wheat distillers dried grains with solubles, corn gluten meal, molasses, golden flakes, canola oil, minerals, and vitamins). After incubation, the bags were removed from the rumen and, together with those samples representing 0 h, rinsed under cold water to remove excess ruminal contents. The samples were then washed with cool water and dried at 55°C for 48 h. The dry samples were stored at 4°C until further analysis.

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