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Short communication

Influence of pectin on intestinal digestion of chromogens in steers

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ABSTRACT

The study was conducted to determine the effect of duodenal infusion of pectin on the intestinal digestion of chromogens in order to project future research to reduce the yellow pigmentation in adipose tissue of grass-fed cattle. Four steers (462.5 ± 48 kg) with cannulas in the rumen and proximal duodenum were used in a 4 × 4 Latin square design to evaluate the effect of duodenally infused pectin on intestinal digestion of β-carotene and chlorophylls-*a* and -*b*. The basal diet consisted of 97% alfalfa hay and 3% of a mineral premix. Dry matter intake was restricted to 9.6 kg/d. All steers were infused with 2.97 g d^{-1} β-carotene via the ruminal cannula. Chromium oxide was administered simultaneously with β-carotene as an inert digesta marker. Experimental treatments consisted of daily infusions of 0 (saline alone), 18.5, 46.25, and 92.5 g d^{-1} of pectin. Infusion of treatments into the pyloric region of the abomasum was accomplished using a drip line inserted via the duodenal cannula and regulated to dispense respective treatments over a period of approximately 10 min every 3 h. There were no treatment effects on intestinal digestion of OM. Pectin infusion did not affect ($P = 0.90$) intestinal digestion of chlorophyll-*a*, but decreased intestinal digestion of β-carotene ($P = 0.03$) and chlorophyll-*b* ($P = 0.02$). β-carotene digestion tended to decrease with increasing level of pectin infusion (linear component, $P = 0.09$). It was concluded that pectin may affect intestinal digestion of β-carotene and chlorophyll-*b*.

1. Introduction

Harvesting cattle either directly or shortly following a period of grazing can result in carcass discounts due to yellow pigmentation of adipose tissue (Reynoso et al., 2004). Of the plant chromogens involved, β-carotene (the most abundant of the pasture carotenoids) is the major contributor to this yellow discoloration (Mora et al., 1999). Discoloration is largely a function of pigment concentration (Kneifel et al., 1992). Pectin has an antagonistic effect on absorption of carotenoids in monogastrics (Erdman et al., 1986; Judd and Truswell, 1985), while the pectin contained in the ration of ruminants, is completely degraded by ruminal microorganisms and does not reach into the small intestine. The objective of this study was

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to evaluate the potential of pectin by duodenal infusion, as an antagonist of intestinal β-carotene and chlorophylls-*a* and *b* absorption in ruminants.

2. Materials and methods

Animal care and handling was in accordance with the guidelines of the Mexican Council on Animal Care (SAGARPA, 1999).

2.1. Animals and management

Four steers Holstein (462.5 ± 48 kg) with ruminal and a proximal duodenal cannulas (Zinn and Plascencia, 1993) were used were used in a 4 × 4 Latin square design to evaluate the influence of pectin on chromogen digestion. Steers were maintained in individual pens (2.42 × 3.74 m) with free access to water. Fifteen d prior to initiation of the study, steers were treated for internal and external parasites (Ivomec Plus® Merck, Rahawy, NJ).

The basal diet consisted of 97% alfalfa hay and 3% of a mineral premix. Dry matter intake of the basal diet was restricted to 9.6 kg/d (2.34% of body weight) divided into equal portions fed at 0700 and 1900 postprandial h. In addition to basal diet, all steers were infused with 2.97 g d^{-1} β-carotene (30% purity, Alfadeita®, Mexico) via the ruminal cannula. Chromium oxide (38.4 g^{-1} d^{-1}) was administered simultaneously with β-carotene as an inert digesta marker. Dosages of β-carotene and chromium oxide were infused twice daily, 40 min before of each meal (0620 and 1820 postprandial h).

Experimental treatments consisted of daily infusions of 0 (saline alone), 18.5, 46.25, and 92.5 g d^{1} of pectin (Citrus Pectin E 440, Cargill®, Germany). Pectin was diluted in 1 L of saline solution maintained at 37 °C. Infusion of treatments into the pyloric region of the abomasum was accomplished using a drip line inserted via the duodenal cannula and regulated to dispense respective treatments over a period of approximately 10 min. Treatments were infused in equal portions every 3 hours (0020, 0320, 0620, 0920, 1220, 1520, 1820, 2120 postprandial h daily).

2.2. Sampling

Experimental periods consisted of a 10-d diet adjustment period followed by a 4-d collection period. During the collection period duodenal and fecal samples were taken from all steers, twice daily as follows: d 1, 0800 and 1400; d 2, 0930 and 1530; d 3, 1100 and 1700; and d 4, 1230 and 1830 postprandial h. Individual samples consisted of 250 mL of duodenal fluid and 200 g (wet basis) of fecal material. Alfalfa hay was sampled weekly, and pooled within each period. Duodenal samples were pooled within steer and sampling day. Fecal samples were pooled within steer and period. Duodenal and fecal samples were stored at −20 °C until analysis.

2.3. Analytical procedures

Pectin, measured as galacturonic acid was determined by spectrophotometry using m-feniphenol (Ahmed and Labavitch, 1977). Alfalfa, duodenal and feces samples were analyzed for all or part of the following: DM (65 °C for 48 h, AOAC, 2003), OM (by ashing at 600 °C for at least 8 h, AOAC, 2003) and chromium (atomic absorption spectrophotometry). Chlorophylls and β-carotene were determined on freshly thawed alfalfa, duodenal and fecal samples, and then all data were adjusted on a dry basis. Sample extractions of β-carotene, chlorophyll-*a* and -*b*, were performed using a modification of the technique of Bruinsma (1963): (1) weighed samples of alfalfa hay (0.5 g), duodenal chyme (5.0 g) and feces (0.5 g) were placed in 50 mL amber flasks; (2) 20 mL of acetone and 0.01% of a mixture of BHA: BHT (1:1) was added into of each flask; (3) flasks were shaken at 200 rpm orbital shaking incubator (Lab-Line, Melrose Park, Illinois, USA) for 3 h at room temperature, avoiding contact with natural light; (4) samples were filtered on Whatman # 2 paper, washing with 20 mL of petroleum ether; (5) filtrate was stirred continuously for approximately 5 min; (6) after phase separation, the lower phase was discarded, and this procedure was repeated 2–3 times; (7) acetone was removed with 2 or 3 successive washes with distilled water; (8) 10 mL of 40% NaOH was added to acetone extract and shaken; (9) the bottom layer bottom was removed and washed 1–3 times with distilled water to remove the NaOH (confirmed with 3 drops of phenolphthalein); (10) Samples were then washed 1–3 times with 20 mL of 10% $NaSO_4$; 11) the bottom layer was extracted and placed in 50 mL volumetric flasks and gauged with acetone/petroleum ether/water (85:10:5, vol/vol/vol); 12) samples were filtered through 0.45 micron Acro-disks (Gelman Acrodisc Krackeler Scientific, Albany, USA). Filtrate was analyzed for β-carotene, chlorophyll-*a* and -*b* according to HPLC procedures of Armenta and Guerrero (2009) and Franco et al. (2010).

2.4. Statistical analysis

The experiment was analyzed as a 4 × 4 Latin square. The statistical model for the trial was as follows: $Yijkl = \mu + Pti + Pk + \varepsilon ijkl$, where μ is the mean effect, *Pt* is pectin level, *P* is period effect, and ε is the residual error. In addition, polynomial contrast comparisons were conducted on control *vs.* pectin, and linear and quadratic effects of pectin level.