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Effect of quebracho-chestnut tannin extracts at 2 dietary crude protein levels on performance, rumen fermentation, and nitrogen partitioning in dairy cows

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ABSTRACT

Our objective was to determine the effects of a tannin mixture extract on lactating cow performance, rumen fermentation, and N partitioning, and whether responses were affected by dietary crude protein (CP). The experiment was conducted as a split-plot with 24 Holstein cows (mean ± standard deviation; 669 ± 55 kg of body weight; 87 ± 36 d in milk; 8 ruminally cannulated) randomly assigned to a diet of [dry matter (DM) basis] 15.3 or 16.6% CP (whole plot) and 0, 0.45, 0.90, or 1.80% of a tannin mixture in three 4 × 4 Latin squares within each level of CP (sub-plot). Tannin extract mixture was from quebracho and chestnut trees (2:1 ratio). Dietary CP level did not influence responses to tannin supplementation. A linear decrease in DM intake (25.5 to 23.4 kg/d) was found, as well as a linear increase in milk/DM intake (1.62 to 1.75) and a trend for a linear decrease in fat-and-protein-corrected milk (38.4 to 37.1 kg/d) with increasing levels of tannin supplementation. In addition, there was a negative linear effect for milk urea N (14.0 to 12.9 mg/dL), milk protein yield (1.20 to 1.15 kg), and concentration (2.87 to 2.83%). Furthermore, the change in milk protein concentration tended to be quadratic, and predicted maximum was 2.89% for a tannin mixture fed at 0.47% of dietary DM. Tannin supplementation reduced ruminal NH_3-N (11.3 to 8.8 mg/dL), total branched-chain volatile fatty acid concentration (2.97 to 2.47 mol/100 mol), DM, organic matter, CP, and neutral detergent fiber digestibility. Dietary tannin had no effect on intake N (587 ± 63 g/d), milk N (175 ± 32 g/d), or N utilization efficiency (29.7 ± 4.4%). However, feeding tannin extracts linearly increased fecal N excretion (214 to 256 g/d), but reduced urinary N (213 to 177 g/d) and urinary urea N (141 to 116 g/d) excretion. Decreasing dietary CP did not influence milk production, but increased N utilization efficiency (milk N/N intake; 0.27 to 0.33), and decreased milk urea N (15.4 to 11.8 mg/dL), ruminal NH_3-N (11.0 to 9.3 mg/dL), apparent digestibility of DM (66.1 to 62.6%), organic matter (68.2 to 64.3%), and CP (62.9 to 55.9%), as well as urinary N excretion (168 vs. 232 g/d). Results of this study indicated beneficial effects of 0.45% tannin extract in the diet on milk protein content. Increasing tannin extract levels in the diet lowered urinary N excretion, but had detrimental effects on DM intake, milk protein content, milk protein yield, and nutrient digestibility.
Key words: quebracho and chestnut tannins, nitrogen, protein, dairy

INTRODUCTION

Tannins are naturally occurring plant polyphenols with multiple phenolic hydroxyl groups that may form complexes with proteins, metal ions, AA, and polysaccharides (Makkar, 2003). They are broadly classified into hydrolysable (polyphenolic units esterified to a carbohydrate core) and condensed tannins (polymers of covalently bonded flavonols). Hydrolysable tannins and condensed tannins may have both adverse and beneficial effects on animal performance depending on their concentration, chemical structure, and the physiological state of the animal and composition of the diet (Makkar, 2003).

Tannins have the capacity to bind protein through hydrogen bonds forming a tannin–protein complex that is stable in the rumen (pH 5.0 to 7.0) and resistant to rumen microbial degradation but dissociated in the low pH environment of the abomasum (Makkar, 2003). Thus, tannins can reduce the amount of protein that is digested in the rumen and increase the flow of protein to the small intestine. Conversely, tannins may reduce nutrient digestibility by inhibiting digestive enzyme and rumen microbial activity (Makkar, 2003). Dose-response studies investigating effects of tannin extracts on animal performance, nutrient utilization efficiency, and N partitioning in lactating dairy cows are scarce. Furthermore, to the best of our knowledge,

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no studies have evaluated the effects of a combination of condensed and hydrolysable tannins and their possible interactions with dietary CP level in lactating dairy cows. For example, tannins may not affect protein digestion when fed at low levels and diets have excess protein. However, when dietary protein is close or below requirements, tannins may be detrimental for animal performance (Waghorn, 2008).

Dschaak et al. (2011) reported a decrease in DMI, but no change in digestibility, milk production, and ruminal fermentation when cows were fed quebracho tree (*Schinopsis* spp.) extracts at 3% of diet DM. However, Ahnert et al. (2015) observed a reduction in apparent total-tract nutrient digestibility when quebracho extracts were fed at 4 and 6% of diet DM but not when fed between 1 and 2% of dietary DM to growing dairy heifers. Sliwinski et al. (2004) and Liu et al. (2013) reported no effect of chestnut tree (*Castanea sativa*) extracts on DMI and milk production when fed at either 0.49 or 1% of diet DM. Previous studies have also shown that tannin extracts obtained from quebracho (Dawson et al., 1999; Komolong et al., 2001) or chestnut (Sliwinski et al., 2004) can decrease urinary N excretion. Ammonia emission from manure is highly correlated with urinary urea N excretion (Aguerre et al., 2010), which is most vulnerable to volatilization after conversion to NH_3 on the barn floor (Muck and Richards, 1983). Thus, a tannin mix from quebracho (condensed tannin) and chestnut tree extracts (hydrolysable tannins) fed at a level that does not compromise animal performance might be used as a dietary strategy to reduce excretion of environmental labile urinary N in manure.

Thus the first hypothesis evaluated in this trial was that dietary supplementation of a mixture of quebracho and chestnut tannin extracts has no effect on animal performance, digestibility, and rumen function but it reduces excretion of environmental labile urinary N. The second hypothesis stated that the effect of tannin supplementation is influenced by dietary CP level. Thus, our main objective was to determine the dose response to 4 levels of a tannin mix extract fed at 2 levels of dietary CP on lactating cow performance, nutrient digestibility, rumen parameters, and urinary N excretion.

MATERIALS AND METHODS

Animals and Treatments

The experiment was conducted as a split-plot with 2 levels of dietary CP evaluated in a complete randomized design (whole plot) and 4 levels of tannin extract evaluated within each level of dietary CP in three 4 × 4 Latin squares (sub-plot). Twenty-four multiparous lactating Holstein cows (mean ± SD; 669 ± 55 kg of BW; 87 ± 36 DIM) were first randomly assigned to 1 of 2 dietary treatments of either 15.3 or 16.6% CP on a DM basis. A 1.3-percentage-unit difference between dietary CP levels was achieved by substituting soybean meal and expeller soybean meal for ground corn in a custom-made concentrate mix (Table 1). The 12 cows in each dietary CP level were then randomly assigned to 1 of 4 dietary treatments including 0, 0.45, 0.90, or 1.80% (DM basis) of a tannin extract from quebracho and chestnut trees. One of the 3 Latin squares within each CP level included ruminally cannulated cows.

The tannin extract (Bypro; Silvateam, Indunor S.A., Argentina) contained (by weight) one-third chestnut tannin extract and two-thirds quebracho tannin extract. Quebracho and chestnut extracts were separately analyzed by matrix-assisted laser desorption/ionization time of flight (**MALDI-TOF**) mass spectrometry according to Pizzi et al. (2009). Analysis by MALDI-TOF of the quebracho extract indicated the following composition (DM basis): 84.3% condensed flavan-3-ols (predominantly profisetinidin), 10.7% oligomers of flavan-3-ols (catechin and epicatechin dimers), and 5% carbohydrate derivate (dimers of pentose, monocarboxylic acid of hexose, and 6-carbon sugars). The MALDI-TOF chestnut extract analysis indicated the following composition (DM basis): 7.9% digalloyl glucose, 5.0% trigalloyl glucose, 16.5% pentagalloyl glucose, and 70.6% oligomers of digalloyl glucose, trigalloyl glucose, and pentagalloyl glucose. A more detailed description of the chestnut extract structure has been reported by Pizzi et al. (2009). Rice hulls were replaced proportionally to the tannin included in the diet (Table 2). Diets were offered as TMR once daily at 0700 h allowing for 5 to 10% orts. Ingredient mix was adjusted based on weekly forage DM analysis. Treatment sequences within Latin square were balanced for carryover effects in subsequent periods. Each period lasted 21 d and consisted of 14 d for diet adaptation and 7 d for data and sample collection.

Cows were housed in tie-stall barns bedded on rubber mats with wood shavings as bedding and had free access to water throughout the experiment. Care and handling of the animals was conducted as outlined in the guidelines of the University of Wisconsin College of Agricultural and Life Sciences Animal Care and Use Committee.

Feed and Milk Sampling and Analyses

Samples of silages, premixes, and cottonseed were collected on the last day of each period and stored at −20°C. Total mixed ration and orts samples were col-