Calliandra calothyrsus supplementation effects on weight gain and efficacy of control of gastrointestinal nematodes in weanling goats^{1,2}

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ABSTRACT

Calliandra (Calliandra calothyrsus) is a shrub legume with high concentration of crude protein (CP), ±22%, and condensed tannin (CT), 19 to 30%. These characteristics make it an alternative for supplementing low-quality grass diets and for reducing levels of gastrointestinal nematode (GIN) infestation. This experiment was conducted to determine the effects of feeding Calliandra on live weight (LW) gain and its efficacy against GIN in weanling goats. Eight Boer goats (11.3 kg) were randomly assigned to two treatments: supplementation with guineagrass hay (GH), or with freshly cut Calliandra (FC) leaves and fine stems. Fortnightly the animals were weighed, feces samples were collected to determine fecal egg counts (FEC), and FAMACHA® index scores were assigned. Blood samples were taken every 21 days to determine packed cell volume (PCV). Data were analyzed by using repeated measure analysis. Supplementation with FC had no significant effect on total live weight gain during 105 days (2.16 vs. 1.59 kg), FAMACHA© index (3.4 common value), nor PCV (25.8% vs. 26.6%) for FC and GH, respectively. However, FEC was significantly lower (p = 0.0014) in the group supplemented with Calliandra than in the group given GH; the respective adjusted means were 638 and 982 eggs per gram. The decrease in fecal excretion of GIN eggs could have the beneficial effect of reducing pasture contamination.

Key words: Calliandra, gastrointestinal nematodes, fecal egg counts

RESUMEN

Suplementación con *Calliandra calothyrsus* y su efecto en ganancia de peso y eficacia en el control de nematodos gastrointestinales en cabritos recién destetados

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Calliandra (Calliandra calothyrsus) es una leguminosa arbustiva con altos niveles de proteína bruta (PB), ±22%, y taninos condensados (TC), 19 a 30%. Estas características la convierten en una alternativa para la suplementación de dietas basadas en forrajes gramináceos de baja calidad y para contribuir a la reducción en la incidencia de nematodos gastrointestinales (NGI). El objetivo del experimento fue determinar el efecto de la suplementación con Calliandra sobre el desempeño productivo y la incidencia de NGI en cabros destetados. Ocho cabros de la raza Boer (11.3 kg) se asignaron aleatoriamente a uno de dos tratamientos: suplementación con heno de hierba quinea (HG) y suplementación con hojas y tallos finos de Calliandra fresca (CF). Cada dos semanas se pesaron los animales, se tomaron muestras de las heces fecales para el recuento de huevos de NGI y se registró el índice FAMACHA©. Se tomaron muestras de sangre cada 21 días para determinar el nivel de hematocrito. Los datos se analizaron usando un diseño de medidas repetidas. La suplementación con CF no tuvo efecto significativo sobre la ganancia total en peso vivo durante 105 días (2.16 vs. 1.59 kg), el índice FA-MACHA© (3.4 en común), ni el hematocrito (25.8% vs. 26.6%) para CF v HG. respectivamente. Sin embargo, el recuento fecal de huevos de NGI fue significativamente menor (p = 0.0014) en el grupo suplementado con CF que en el que recibió HG, siendo las medias ajustadas 638 y 982 huevos por gramo. La reducción en la excreción fecal de huevos de NGI puede tener un efecto benéfico en la disminución de la contaminación de las pasturas.

Palabras clave: Calliandra, nematodos gastrointestinales, recuento de huevos fecales

INTRODUCTION

Shrub and tree legumes are considered promising feed for ruminants in the tropics where animal production is limited by low availability of good quality forage, especially during the dry season. These legumes constitute an important source of protein, vitamins and minerals for livestock in tropical regions. Calliandra (*Calliandra calothyrsus*), native to the Pacific coast of Mexico (Chamberlain, 2001), is a shrub legume with high concentrations of crude protein (CP), ±22%, and condensed tannins (CT), 19 to 30%, that has excellent agronomic characteristics (Noto et al., 2004). Pamo et al. (2005) and Nherera et al. (1998) supplemented goat feeding with Calliandra and found a positive effect on weight gain. Perez-Maldonado and Norton (1996) reported that CT in Calliandra (2.3%) protected dietary protein from ruminal degradation in sheep and goats, but found neither a beneficial nor a detrimental effect on diet utilization efficiency.

Several researchers have indicated that plants containing CT may have anthelmintic properties (Lange et al., 2006; Paolini et al., 2003a; Molan et al., 2002), and feeding these plants is considered an alternative to prevent development of anthelmintic resistance in populations of gastrointestinal nematodes (GIN). Pagán et al. (2005) in a study at six small ruminant farms in different regions of Puerto Rico found resistance to Benzimidazole and Levamisole in both semiarid and humid regions. The most pathogenic nematode parasite of small ruminants in tropical and

subtropical regions is *Haemonchus contortus*, "barber pole worm". This parasite is more problematic in regions where rain is abundant during the entire year. Therefore, clinical outbreaks and mortalities due to Haemonchosis in sheep and goats of all ages can occur at any time (Waller, 2002).

Information on tannin-containing tropical forages and their efficacy in combating GIN and improving weanling goat performance is limited. Tannins are subdivided into two groups; hydrolysable tannins (polymers of gallic or ellagic acid esterified to a core molecule) and proanthocyanidins (flavonoid polymers), more commonly referred to as CT. Hydrolysable tannins are more susceptible to enzymatic and nonenzymatic hydrolysis and usually are more soluble in water. The principal difference between these two groups is the greater capability of the CT to form complexes with proteins, whereas this property is limited in hydrolysable tannins, because they are rapidly degraded (Hagerman et al., 1992). The CT in ruminant diets can have both positive and negative effects, depending on its concentration. According to Barry and McNabb (1999), the ideal concentration of CT in forage legumes is 2 to 4%. Tannins in higher concentration reduce intake, digestibility of protein and carbohydrates, and animal performance (Reed et al., 1990). Moderate levels of CT have been reported to increase the efficiency of nitrogen utilization through greater flow of intact protein to the duodenum (McNabb et al., 1993).

The objective of this study was to determine the effects of supplementation with Calliandra on live weight gain and on efficacy against GIN in weanling goats grazing native pastures.

MATERIALS AND METHODS

The research was conducted at the Alzamora Farm, University of Puerto Rico, Mayagüez Campus. The duration of the experiment was 105 d; the first 15 d constituted an adaptation period. Eight Boer goats, with average live weight (LW) 11.3 kg, were randomly assigned to two treatments: supplementation with guineagrass hay (GH; *Panicum maximum* Jacq.), or with leaves and fine stems of fresh Calliandra (FC). The Calliandra was cut daily and offered to the animals in individual pens. Supplementation was at 20% of the expected dry matter (DM) intake (3% of LW daily). After the morning feeding, the goats grazed native grass pastures (*Dichanthium annulatum, Heteropogun contortus, Cynodon dactylon*) and had water available ad libitum.

Dry matter content of the forages was determined by drying the samples at 60° C in an oven for 24 hours. Nitrogen concentration was determined according to AOAC (1990) procedure and CP was calculated as N*6.25. Neutral detergent fiber (NDF) concentration was de-

termined according to the procedures described by Van Soest et al. (1991). Samples of Calliandra obtained from the Alzamora Farm forage plots at 49-d regrowth were sent to the Agricultural Experiment Station, Stephenville, Texas A&M University, and analyzed for concentrations of extractable CT, protein-bound CT, and fiber-bound CT. This procedure was performed by using Calliandra as standard (Rivera, 2006). The same Calliandra plants provided forage at a similar stage of regrowth for use in this study.

Fortnightly the goats were weighed, feces samples were collected for determining fecal egg count (FEC) by using modified Mc. Master System (Kaplan, 2002), and FAMACHA© index scores were assigned. Blood samples were taken by venipuncture every 21 days from individual animals to determine packed cell volume (PCV). The goats were dewormed at the beginning of the experiment. Fortnightly, animals found to have FAMACHA© index scores of 4 and 5 were dewormed with products having levamisole hidrochloride and febendazole as active ingredients.

The live weight, FEC and PCV data were subjected to repeated measure analysis. The FAMACHA© index data were analyzed by Pearson's chi-square test and chi-square MV-G2 test. The SAS/STAT 9.1 program was used for these procedures (SAS, 2004).

RESULTS AND DISCUSSION

The chemical composition of the grazed and supplemented forage is presented in Table 1. Condensed tannin concentration (CT) of Calliandra harvested at 49-d regrowth and determined by using a Calliandra standard and expressed on a dry basis was 19% for extractable CT, 1.42% for protein-bound CT, and 0.50% for fiber-bound CT (Rivera, 2006). After the adaptation period (15 d) the goats consumed all of the supplemental forages offered in both treatments. The types of supplementation had no significant effect on live weight (p > 0.05); mean values (calculated as initial weight + final weight/2) were 12.6 and 12.4 kg for FC and GH, respectively. The trend over time in live weight is shown in Figure 1, where numerical differences between treatments of 682, 228 and 453 g are observed on days 73, 88 and 105, respectively.

There was no effect (p > 0.05) of type of supplementation on PCV, the mean values being 25.8% and 26.6% for FC and GH, respectively. The means of PCV observed were well above 19% (critical value) for all samplings (Figure 2). Goats with PCV under this critical value are considered anemic (Vatta et al., 2001). There were four individual values under 19% during the experiment; the overall dispersion was from 14% to 38%.

According to the Chi-square test, FAMACHA© index score was not dependent (p > 0.05) on type of supplementation. In both FC and GH

Nutrient	Grazed Forage	Guineagrass hay	Fresh Calliandra
DM (%)1	29.4	88.9	47.3
CP (%) ²	5.4	4.9	14.3
NDF (%) ²	77.4	71.8	64.8

Table 1.—Chemical composition of grazed and supplemental forages.

groups FAMACHA© index mean was 3.4. All individual animal index values assigned were 2, 3 or 4; 3 was the mode. FAMACHA© index trends are shown in Figure 3.

The interaction between supplementation treatment and period of sampling was significant for FEC. Fecal egg count was significantly lower (p = 0.0014) in the group supplemented with Calliandra than in the group given GH at 88 d, with respective adjusted means of 638 and 982 eggs per gram. The trends in egg excretion rate and number of animals dewormed are shown in Figure 4. Fewer animals were dewormed under the FC than under the GH treatment during the first 73 d of the experiment (Figure 4). The egg excretion levels in goats of the GH group increased excessively at 88 d. For this reason, all animals were

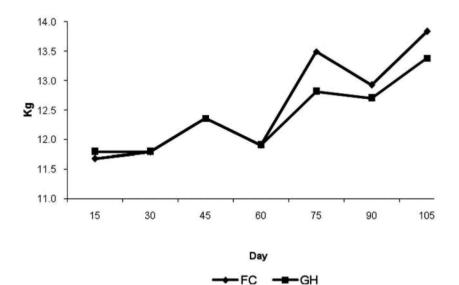


FIGURE 1. Mean live weight of grazing goats supplemented with fresh Calliandra (FC) and guineagrass hav (GH).

¹DM = dry matter, CP = crude protein, NDF = neutral detergent fiber.

²Dry matter basis.

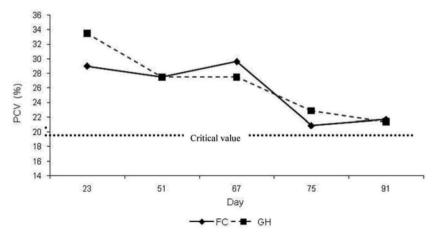


FIGURE 2. Mean packed cell volume (PCV) of grazing goats supplemented with fresh Calliandra (FC) and guineagrass hay (GH).

dewormed; similar egg excretions between the two treatments were observed at the next sampling (105 d).

Supplementation with freshly cut Calliandra leaves and fine stems at 20% of the expected DM intake was not enough to improve weight gain significantly under the conditions of the experiment (Table 2). This lack of effect on weight gain does not agree with previous results in goats. Ebong (1996) examined the effect of graded levels (0, 25, 50,

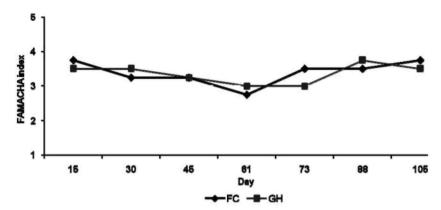


FIGURE 3. Mean FAMACHA© index scores of grazing goats supplemented with fresh Calliandra (FC) and guineagrass hay (GH).

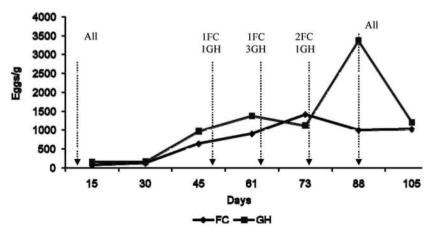


FIGURE 4. Mean fecal egg count (FEC) of grazing goats supplemented with fresh Calliandra (FC) and guineagrass hay (GH). Number of dewormed animals from each treatment ($\underline{\vdots}$).

75%) of substitution of Calliandra leaf meal (CLM) instead of supplemental soybean meal (SBM) on animal performance, and he found the highest daily gains in animals supplemented with 50:50 SBM:CLM. The weight gain obtained with the 75:25 SBM:CLM supplement (83.9 g/d) was markedly higher than that of the FC treatment in the present study (20.5 g/d). Nherera et al. (1998) also reported higher weight gain (42 g/d) with a diet of maize stover and air-dried Calliandra than that obtained in this study with FC supplementation. These differences could be due to low quality of the grazed forage in this study, especially to its low concentration of CP (5.43%). Pamo et al. (2002) fed goats with a basal diet of tropical forages supplemented with 380 g/d of fresh

Table 2.—Live weight gain and deworming frequency in grazing weanling goats supplemented with GH (guineagrass hay) and FC (fresh Calliandra).

	Mean		
-	FC	GH	SE
Initial weight (kg)	11.7	11.8	0.8
Final weight (kg)	13.8	13.4	0.8
Total weight gain (kg)	2.2	1.6	_
Daily weight gain (g)		15.1	1.6
Times that each animal was dewormed		3.3	_
Average interval between consecutive dewormings (days)	47.3	37.2	-

leaves of Calliandra, corresponding to 6 g N, and observed weight gains of 13.8 g/d; this value is closer to that of the present study.

There was no effect of supplementation with Calliandra on PCV and FAMACHA© index, but fecal egg excretion was decreased. These results are similar to observations of Paolini et al. (2003a) in a study with Saanen goats experimentally infected with *Haemonchus contortus*. Quebracho extracts (5% of the diet DM) were administered as a source of tannins to the treatment group. A decrease in egg excretion was observed, but there were no significant differences in the blood parameters or number of worms counted at necropsy. Fecundity per capita of the worms present in the tannin-fed group of goats was significantly reduced. These results suggest that the effect of condensed tannins on nematode fecundity is the principal mode of action of CT. Lange et al. (2006) reported a decrease in fecal egg excretion when sheep were fed *Lespedeza cuneata* hay. Thereafter when hay feeding was suspended, there was an increase in fecal egg excretion. There was no effect from supplementation on the number of larvae in the abomasum.

Paolini et al. (2003b) conducted two experiments which led them to suggest a relationship between the effect of the CT and the parasitic stage of gastrointestinal nematodes (GIN). In the first experiment a group of goats were infected with L3 (infective stage) of *Trychostrongy-lus colubriformis* and *Telodorsagia circumcincta*. After 7 d, quebracho extract was administered. In this case the tannins had an effect on egg excretion and fecundity of the female nematodes, but no effect on the number of nematodes. In the second experiment, in which the animals were infected as before but the quebracho extract was administered immediately, there was an effect from tannins on the number of nematodes but not on fecundity of the female parasites. Min et al. (2004) observed a decrease in fecal egg excretion and a reduction in hatching and development of larvae in goats grazing forage containing CT (*Lespedeza cuneata*, 46 g extractable CT/kg DM).

These results from different studies support the hypothesis of Lange et al. (2006) that CT affect fertility of the female nematodes in the abomasum but not their viability. This hypothesis is consistent with the reduction in fecal egg excretion and lack of effect on PCV and FAMACHA observed in the present study. Additionally, the results obtained by Min et al. (2004) and Paolini et al. (2003b) showed an effect on establishment of larvae in the abomasum. However, the mechanism by which the tannins act is not well known. Butter et al. (2001) studied the effect of quebracho tannin on nematodes in the small intestine of mice. They found effects of CT on *Nippostrongylus brasiliensis*, which, although closely associated with the mucosa, actually dwells within the small intestinal lumen. There was no effect on *Trichinella spiralis*,

which penetrates the small intestinal mucosa. However, in an in vitro study with adult nematodes of *N. brasiliensis* and *T. spiralis* incubated in quebracho tannins, both species were affected. These results suggest that tannins have a toxic effect that requires direct contact between parasite and tannin. On the other hand, Molan et al. (2000), in an experiment on larval migration of *Trichostrongylus colubriformis*, observed a paralysis effect because the larvae exposed to CT did not pass through the pores of a screen, but the viability remained high at 87 to 94%. This finding suggests an effect on nematode muscular activity. Additional studies are necessary in order to understand the mechanism of CT action on GIN and the effects of the parasite stage on this relationship.

The decrease in fecal excretion of GIN eggs achieved with Calliandra supplementation could have the benefit of reduced pasture contamination by infective larvae. Supplementation with this legume is an alternative for control of gastrointestinal nematodes of goats under tropic conditions, and a mitigant to the problem of anthelmintic resistance.

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