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Effects of *Havardia albicans* supplementation on feed consumption and dry matter digestibility of sheep and the biology of *Haemonchus* contortus*

H.H. Galicia-Aguilar, L.A. Rodríguez-González, C.M. Capetillo-Leal, R. Cámara-Sarmiento, A.J. Aguilar-Caballero, C.A. Sandoval-Castro*, J.F.J. Torres-Acosta*

Facultad de Medicina Veterinaria y Zootecnia, Universidad Autónoma de Yucatán, Km. 15.5 Carretera, Mérida-Xmatkuil, A.P. 4-116 Itzimna, 97100 Mérida, Yucatán. Mexico

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ABSTRACT

This study evaluated effects of Havardia albicans foliage intake on sheep dry matter digestibility (DMD) and included post-mortem evaluation of Haemonchus contortus infection (i.e., worm burdens, worm female length, fecundity). Fifteen hair sheep $(16 \pm 4.2 \text{ kg})$ live weight (LW)) raised free of gastrointestinal nematodes were used. Sheep were fed a basal diet: grain based concentrate and Pennisetum purpureum grass (900 and 100 g/kg DM, respectively) and were infected on day 0 with 3000 H. contortus infective larvae. On day 28 post-infection (PI) sheep were distributed to three groups being: Control (100 g/kg DM of basal diet), HA group (basal diet 600 + H. albicans 400) and the HA + PEG group (basal diet 600 + H. albicans 400) + Polyethylene glycol (PEG). The H. albicans foliage was supplied for 13 d including 7 d of adaptation (i.e., day 28-40 PI). The content of condensed tannins (CT), total polyphenols (TP) and total tannins (TT) was determined in the foliage. Concentrate, grass and H. albicans intake (g DM/kg LW^{0.75}), DMD and H. contortus egg excretions were determined. Sheep were slaughtered at day 41 PI to assess adult worm burdens, female worm lengths and fecundity (i.e., eggs in utero). Foliage had (g/kg DM): 71.5 CT, 61.3 TP and 66.9 TT. Experimental groups had similar feed intakes, and intake of H. albicans was not affected by PEG. The DMD of HA+PEG and HA groups was lower than Control (P<0.05). Post-mortem worm burdens were similar in all groups, but the H. contortus females in the HA group were shorter and had reduced fecundity (P<0.05). Adding PEG (HA+PEG) eliminated the effect of H. albicans intake on worm length. A short period of H. albicans intake affected the DMD of sheep and reduced the H. contortus female worm length and fecundity. The use of this type of tannin-rich foliage could bring nutritional and anti-parasitic benefits to the ruminants which consume them.

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Abbreviations: AH, anthelmintic; CP, crude protein; CT, condensed tannins; DMD, apparent dry matter digestibility; EPG, eggs/g of feces; HA, Havardia albicans; L₃, infective larvae (or larvae stage 3); LW, liveweight; ME, metabolizable energy; MP, metabolizable protein; PEG, polyethylene glycol; PI, post infection; PSM, plant secondary metabolites; TFEC, total fecal egg count; TP, total polyphenols; TT, total tannins.

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^{*} Corresponding authors. Tel.: +52 999 942 3200; fax: +52 999 942 3205.

E-mail addresses: ccastro@uady.mx (C.A. Sandoval-Castro), tacosta@uady.mx (J.F.J. Torres-Acosta).

1. Introduction

Some fodder plants have nutraceutical properties which are defined as nutritional and pharmacological benefits which can improve the productive performance of animals (Min and Hart, 2003; Heckendorn et al., 2007). There are several reviews dealing with effects of condensed tannins (CT) in ruminants as increasing feed metabolizable protein (MP) uptake, as well as anthelmintics (Hoste et al., 2006, 2012; Waghorn, 2008; Alonso-Díaz et al., 2010). Although the evaluation of tropical tannin-rich resources, such as *Havardia albicans* or *Lysiloma latisiliquum*, has shown a clear *in vitro* anthelmintic (AH) effect against *Haemonchus contortus*, the *in vivo* AH effects are less well studied. Experiments with *L. latisiliquum* foliage showed positive AH effects against the development stages and reproductive biology of adult parasites (Brunet et al., 2008; Martínez-Ortíz-de-Montellano et al., 2010), and the latter showed that parasitized animals increased intake of *L. latisiliquum* foliage. Evaluation of *H. albicans* foliage, a legume tree with higher levels of CT than *L. latisiliquum*, also showed that parasitized sheep ate more tannin-rich fodder, causing a reduction on their dry matter digestibility (DMD) and a clear reduction of the quantity of *H. contortus* eggs per g of feces (EPG) (Méndez-Ortíz et al., 2012). The latter study lacked *post-mortem* information to corroborate effects of *H. albicans* CT on development and reproductive biology of *H. contortus*.

This study evaluated effects of *H. albicans* foliage intake on sheep DMD, *H. contortus* egg excretion and worm burdens, as well as the female worm length and fecundity.

2. Materials and methods

2.1. Animals and experimental facilities

Twenty-one hair sheep lambs (Pelibuey \times Dorper) \sim 3 mo old were selected. Lambs were kept free of parasitic nematodes from birth by maintaining them under conditions which avoided contact with gastrointestinal nematode infective larvae in feed as well as maintaining them in concrete floor pens (Torres-Acosta, 1999). The parasite naïve status of the lambs helped to avoid interference of the lambs' immune systems on the parasitological results of this study. Parasite naïve lambs were confirmed free of infection and were allocated to individual metabolic cages. Lambs were fed at 45 g DM/kg liveweight (LW) of concentrate feed and *P. purpureum* grass (900 g and 100 g/kg DM, respectively).

2.2. Infective larvae and artificial infection of animals

Lambs were infected with an oral dose of 3000 H. contortus infective larvae (L_3) per lamb as per guidelines for the evaluation of efficacy of AH in ruminants (Wood et al., 1995). The H. contortus L_3 were obtained from CENID-PAVET-INIFAP in Cuernavaca (Mexico). Doses of L_3 larvae were refrigerated at 5 °C until 2 h before infection when they were kept at ambient temperature (i.e., 25 °C). On day 28 post-infection 15 of the 21 original lambs were selected to continue because they were showing evidence of a sub-clinical infection (i.e., presence of nematode eggs in the feces).

2.3. Experimental treatments

Three criteria were considered for group allocation, being to achieve: (a) similar LW/group ($16 \pm 4.2 \text{ kg LW}$), (b) similar H. contortus fecal egg counts ($2573 \pm 733.6 \text{ HPG}$) and (c) similar number of males and females/group (4:1 respectively). Lambs were assigned to one of three experimental groups being: Control group, basal diet (concentrate feed mixture and grass; 900:100 g/kg DM); HA group, basal diet and H. albicans fodder (600:400 respectively) and HA+PEG group, basal diet and H. albicans foliage (600:400 respectively)+25 g of polyethylene glycol (PEG E-3350 Pluracol®, Polioles, Mexico City, Mexico) diluted in 25 ml of water. The PEG was orally administered daily.

2.3.1. Concentrate feed mixture

The feed mixture was formulated according to AFRC (1993) requirements for growing sheep. The ingredients used were: ground sorghum grain (350 g), wheat bran (300 g), soybean meal (100 g), ground *Cynodon nlemfluensis* hay (100 g), ground turkey litter (100 g), sugar-cane molasses (30 g) and mineral salt (20 g/kg DM).

2.3.2. Foliages

Chopped *Pennisetum purpureum* grass was fed *ad libitum* every morning. *H. albicans* foliage was harvested daily in the morning from at least 5 trees and the leaves were manually mixed. A total of 14 *H. albicans* trees were used. The rationale for the amount of *H. albicans* fodder fed to the HA+PEG and HA groups was to allow *ad libitum* foliage intake without jeopardizing lamb performance. *H. albicans* intake reported by Méndez-Ortíz et al. (2012) was used as the criteria for the initial amount offered (*i.e.*, 350 g DM/lamb/d).

2.4. Animal feeding

Individual lambs were weighed weekly and this LW was used to adjust the amount of feed fed to each to a level of 45 g DM/kg LW. The groups consuming *H. albicans* had an adaptation period of 7 d, suggested by data of Méndez-Ortíz et al.

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