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Relationship between intake of tannin-containing tropical tree forage, PEG supplementation, and salivary haze development in hair sheep and goats

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ABSTRACT

The objective of this study was to estimate the relationship between tannin binding salivary protein (TBSP) and condensed tannins (CT) intake in hair sheep and creole goats. Foliage was obtained from trees with different levels of CT content; animals were offered foliage *ad libitum*, with or without polyethylene glycol (PEG). Saliva haze development (SHD) was evaluated as evidence for TBSP. PEG consumption did not affect dry matter intake (DMI) ($P > 0.05$). Lignin ($R = -0.714$, $P < 0.001$) and Crude Protein (CP) ($R = 0.622$, $P < 0.001$) contents had a stronger association with DMI than CT ($R = 0.622$, $P < 0.011$) in sheep; no significant association was found in goats. The positive relationship between tannin intake and SHD ($P < 0.05$) was not confirmed after PEG supplementation in sheep ($P > 0.09$), but remained significant for goats ($P < 0.01$), except for those fed *Lysiloma latisiliquum* ($P = 0.07$). Foliage lignin or CP contents are better predictors of foliage intake than CT. Sheep and goats fed with tropical tree forages containing different levels of tannins exhibited differences in intake behavior; moreover, individual variations in TBSP expression helps explaining foliage DMI.

1. Introduction

Several tropical forage trees are commonly used as a source of macronutrients for ruminants in spite of their secondary metabolite content. Consumption of plant materials containing compounds (*e.g.*, tannins) may have a detrimental effect on the animals such as reduction of food intake and digestibility (Alonso-Díaz et al., 2010). Some animal species evolved defense mechanisms for such compounds; those are salivary proteins commonly named tannin binding salivary proteins (TBSP), which have a high affinity for tannins (Skopec et al., 2004; Shimada, 2006). The role of TBSP is to reduce the interaction between tannins and dietary proteins, improving the availability of the latter for the animal (Salem et al., 2007), and possibly increasing the intake of tannin-rich forages. In some animal species, the presence of tannins in the diet may stimulate TBSP synthesis as an adaptive response (McArthur et al., 1995; Clauss et al., 2005; Shimada et al., 2006). However, in small ruminants (sheep and goats) the evidence is contradictory. In Mediterranean goats, tannin consumption failed to increase TBSP

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production even after long exposure to tannin-rich plants (Hanovice-Ziony et al., 2010). On the other hand, sheep and goats fed for several years with tannin-rich forages, derived from the native vegetation, showed evidence of TBSP expression (Alonso-Diaz et al., 2012). Further studies in Mexico showed that lambs with no prior contact with tannin-rich materials had elevated TBSP (measured as increased haze development) after a short period of exposure to tannin-rich forages (Vargas-Magaña et al., 2013). On the other hands, goat kids without prior exposure to tannin-rich forages seemed to have developed TBSP as an innate feature and were unable to show a further increase in haze development after exposure to tannin-rich forages (Ventura-Cordero et al., 2015). The presence of TBSP in small ruminants from the Yucatan Peninsula has also been postulated as these studies showed a lack of increase in the intake of tannin-rich forages after polyethylene glycol (PEG) was included in the diet with the aim to block the negative effects of tannins (either astringency or post-ingestive effects) (Hernandez-Orduño et al., 2012, 2015; Revaud et al., 2014). In spite of previous evidence suggesting a relationship between tannin-rich forage consumption and TBSP in tropical sheep (Vargas-Magaña et al., 2013), there is no clear explanation on why tannins do not limit tannin-rich forage intake. Moreover, it is important to confirm the effect of PEG supplementation on forage intake and TBSP expression. Therefore, we hypothesized that tannins contained in foliage would stimulate TBSP production and this response would allow sheep and goats to ingest a larger amount of foliage. Thus, the aim of this study was to evaluate the relationship between the intake of tropical forage trees with different tannin levels and the TBSP content in hair sheep and creole goats.

2. Materials and methods

2.1. Location of the study

The experiments were carried out in the small ruminant farm at the Faculty of Veterinary Medicine and Animal Science, Universidad Autónoma de Yucatán, México. The weather of the area is classified as AW_0 (hot, subhumid, and with summer rainfall). Mean annual temperature and precipitation range from 26 to 27 °C and from 940 to 1110 mm, respectively.

2.2. Forage trees

Fresh foliage of *Piscidia piscipula, Lysiloma latisiliquum*, and *Brosimum alicastrum* leaves was used for the experiment. To minimize variability in forage chemical composition, leaves were harvested every day as a mixture from at least fifteen individual trees with more than ten years of age. The foliage was homogenized before being offered to the experimental animals. The plants for the trial were selected based on the following criteria: (a) the plants are commonly available in the region; (b) the foliage of those plants is commonly used to feed ruminants in the study area; (c) the knowledge about the condensed tannins (CT) content of the foliage. The foliage of *P. piscipula* and *L. latisiliquum* is known to have medium and high CT content, respectively (Monforte-Briceño et al., 2005; Alonso-Díaz et al., 2008). On the other hand, the foliage of *B. alicastrum* is highly palatable and with very low to null content of CT (Alonso-Diaz et al., 2009). Therefore, the latter was included as control foliage.

2.3. Experimental animals

Sheep experiment. Nine Pelibuey hair sheep with an average live weight (LW) of 27 ± 2.5 kg, and an age of eight months were used. Animals had previous experience with tannin-rich forage intake and therefore, tannin consumption (Vargas-Magaña et al., 2013).

Goat experiment. Twelve Creole goats with an average LW of 24 ± 2.5 kg, and an age of eight months were used. Animals had previous experience with tannin-rich forage intake and hence, tannin consumption (Ventura-Cordero et al., 2015).

The experimental sheep and goats were raised nematode-free by maintaining them on concrete floor pens and were fed a diet free of gastrointestinal nematode infective larvae. Before the experiments, sheep and goats were trained to consume PEG, by feeding the foliage with and without PEG for five consecutive days to ensure feed acceptance. All the animals were weighted before the beginning of each experimental period.

2.4. Feed intake

Animals were fed daily with a grain-based concentrated feed (500 g FB) in the morning (8:00 h). At 13:00 h animals were fed fresh tree foliage *ad libitum*. Each animal received foliage of one of the tree species during each period with or without PEG according to each treatment. At 17:00 h animals were offered *P. purpureum* grass *ad libitum*. Before the respective feeds were offered, orts were collected, weighed, and recorded. Feeds and orts samples were collected and stored at 4 °C until further analysis.

2.5. Tannin binding salivary protein (TBSP) in saliva samples

Saliva samples were obtained on the fifteenth day of each experimental period. Animals were sampled during the morning before feeding time. A vacuum pump was connected to Corning® PP centrifuge tubes (catalogue #430290, Corning Mexicana