



# Effects of condensed tannins on live weight, faecal nitrogen and blood metabolites of free-ranging female goats in a semi-arid African savanna

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## ABSTRACT

Current understanding of the effects of condensed tannins (CTs) on productivity of mixed-feeding ruminants is largely based on simple laboratory and feeding experiments. These experiments do not allow mixed feeders such as goats to adequately employ their behavioural and physiological responses to plant secondary metabolites. In a field experiment, we investigated the effects of CTs on growth performance of goats. We hypothesized that CTs reduce blood circulatory nutrient and increase nitrogen in faeces. We divided 45 yearling females into three groups of 15 animals that were orally dosed daily with either CTs, polyethylene glycol 6000 (PEG, a polymer that neutralizes dietary tannins), or water (control). We measured the average daily gains, live weights, faecal nitrogen and four blood metabolites from each goat during dry and wet seasons. Live weights increased over time in both dry ( $P < 0.001$ ) and wet seasons ( $P < 0.001$ ). The average daily gain was consistently greatest for animals dosed with PEG and least for those dosed with CTs. Goats dosed with CTs had the greatest faecal nitrogen and the least blood protein concentrations, while the opposite was true for PEG goats in both seasons. Blood urea and non-esterified fatty acids indicated a negative influence of CTs on energy and protein metabolism. We concluded that CTs limit growth and PEG mitigates the negative effects of CTs on growth performance of free-ranging mixed feeding ruminants.

## 1. Introduction

Although trees and shrubs are an important food resource for wild and domestic herbivores in African savanna ecosystems (Bergstrom, 1992), they are nearly always endowed with condensed tannins (CTs). Being found in approximately 80% of woody plants, CTs are the most abundant chemical defences produced by plants (Bryant et al., 1991). Current knowledge of the effects of CTs on productivity of large herbivores is largely based on simple laboratory and short-term feeding experiments, which do not adequately capture the complex behavioural and physiological responses employed by animals in response to CTs. However, it is through these experiments that it is now known that tannins can either be detrimental or beneficial to the herbivores and environment, depending on the tannin type and concentration in the forages (Min et al., 2003; Piluzza and Bullitta, 2010).

For example, at low to moderate quantities (20–45 g CT/kg DM) CTs bind with and provide protection to the dietary protein from degradation by rumen microbes thereby increasing the efficiency of protein

digestion and absorption later in the small intestines (Waghorn, 2008; Piluzza et al., 2013). Forages containing low levels of CTs may lower the internal parasite burden with positive consequences for animal growth performance (Lisonbee et al., 2009; Piluzza et al., 2013). On the other hand, at high concentrations (> 55 g CT/kg DM), tannins are known to reduce feed intake, and reduce live-weight gain with detrimental consequences for productivity (Waghorn and McNabb, 2003). Increased faecal nitrogen excretions (Kumar and Vaithyanathan, 1990; Owens et al., 2012), reduced growth hormone titre (Barry, 1984) and reduced blood nitrogen (Silanikove et al., 1997) have also been observed from animals exposed to tannin-rich forages. In addition to forming irreversible complexes with dietary proteins, excessive CTs reduce lipid digestion (Barry and Manley, 1986) and bind carbohydrates to form indigestible complexes with the cell wall material (Reed et al., 1990).

Research has also shown polyethylene glycol (PEG) to bind CTs irreversibly over a wide range of conditions and thus reducing the formation of protein-tannin complexes (Silanikove et al., 1996; Silanikove

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et al., 1997) that can be either beneficial or detrimental to the animal depending on the [CTs]. Given that pen experiments oversimplify herbivore responses to CTs, we conducted field experiments with the main objective to determine the effects of CTs on growth performance of free-ranging goats in a semi-arid savanna. We combined live weight measurements with those of nutrition-related blood metabolites to increase the accuracy of assessing nutritional state and welfare of free-ranging goats (Chester-Jones et al., 1990; Ndlovu et al., 2007). We thus compared average daily gain (ADG), live weight, faecal nitrogen, blood glucose, blood urea, blood total protein, and blood non-esterified fatty acids (NEFA) of free-ranging goats that were exposed to different levels of CTs.

We tested the hypothesis that CTs impose nutritional limits to growth performance of goats by reducing nutrient absorption and increasing faecal nitrogen. Based on this hypothesis, we predicted CTs to reduce ADG, or levels of blood protein and blood glucose. We further predicted CTs to increase faecal nitrogen, and levels of blood urea and blood NEFA of free-ranging goats.

## 2. Material and methods

### 2.1. Study area

Fieldwork was conducted during dry (June to August 2012) and wet (January to April 2013) seasons at the Roodeplaart Experimental Farm of the Agricultural Research Council (ARC) in South Africa (25°20'–25°40'S; 28°17'–28°25'E). The climate in the study area is semi-arid with a mean annual precipitation of 646 mm and an average daily temperature of 29 °C (Panagos et al., 1998). The natural vegetation of the farm (2067 ha) is classified as a Marikana Thornveld by Mucina and Rutherford (2006). The dominant tree species whose foliage or fruits were consumed by goats during this study included five species of *Vachellia* (formerly named: *Acacia*), three species of *Searsia* (formerly named: *Rhus*), two species of *Combretum*, *Dichrostachys cinerea*, *Ziziphium mucronata*, *Gymnosporia buxifolia*, *Scolopia zeyheri*, *Ehretia rigida*, *Carissa bispinosa*, *Dombeya rotundifolia*, *Pappea capensis*, *Grewia flava*, *Berchemia zeyheri*, and some *Euclea* species. A number of shrubs were consumed, the main of which were *Lippia rehmannii* and *Tarconanthus camphoratus*. A dwarf shrub *Aloe greatheadii* var. *davyana* was also present in the study area (see Table 1). Plant nomenclature and further details on the phenology and morphology of these plant species can be found in Coates Palgrave (1977,1985) and Kyalangalilwa et al. (2013).

### 2.2. Experimental design

We used 45 yearling, indigenous female goats (South African veld breed) ranging from 10 to 18 months old with an initial live weight of 14.9 kg (standard deviation  $\pm$  3.7). Due to some logistical difficulties, different animals were used for different seasons. In both seasons, all animals were deprived of drinking water overnight, weighed one day before the experiment and were allocated to three treatment groups such that all groups had an equal number of animals (N = 15) and a similar mean within a 95% confidence interval live weight per group. Every morning between 07:00 and 08:00, the same 15 animals were orally dosed with 20 g DM of PEG 6000 dissolved in 50 ml of water whereas another 15 were dosed with 50 ml of water plus 20 g DM of CT (extracted from mimosa bark) and the last 15 received only 50 ml of water (control). The mimosa extract, was obtained from the bark of the Black Wattle (*acacia mearnsii*) trees and contained a minimum of 66% condensed tannin. This extract was obtained from Mimosa Extract Company whose business is to extract tannins shortly after stripping the bark using a counter current principle in auto claws under temperatures above 100 degrees Celsius. Mimosa tannins have an average molecular weight of 1250 units. More information can be obtained on: <http://mimosa-sa.com/production-process-mimosa-extract/> (accessed on 28 February 2018). The three treatment groups were maintained

throughout the experiment during each season. Three grazing paddocks of similar size ( $\pm$  1.7 ha) were fenced off and stocked with 15 animals (i.e. 5 from each treatment group) from 08:00 in the morning until 15:30 in the afternoon daily. All animals were treated for internal and external parasites before the experiment and had *ad libitum* access to water throughout the experiment. From 08:00 all animals were allowed to forage freely in their respective paddocks until 15:30 when they were corralled to avoid predation and theft. All animals were fasted overnight.

### 2.3. Data collection

The experiment lasted for 45 days in dry season and 65 days in wet season. Since different animals were used for different seasons, and live weight, faecal nitrogen and blood measurements were conducted at different time intervals for different seasons, no direct comparisons were made between the two seasons. During dry season, all animals were weighed in the morning on days: 0, 12, 22, 38 and 45 of the experiment, and they were weighed on days: 0, 7, 21, 27, 34, 40, 45 and 65 during wet season. Average daily gains per animal were calculated as the difference between initial and final live weights divided by the duration of the experiment (in days) in each season.

Faecal samples were collected on days: 0, 25, 45 and 65 from all animals in wet season and on days 0, 25 and 45 in dry season. Animals were fitted with nylon faecal collection bags in the afternoon (16:30) and faecal samples collected from the harness bags at 7:00 the following morning. Collected faeces were oven dried at 40 °C until completely dry. The faecal samples were milled and analysed for nitrogen by the micro Kjeldahl method (AOAC, 1997).

Blood samples were collected from all animals on days 0, 31 and 45 during dry season, and on days 0, 25, 45 and 65 during wet season. Sampling was done via a jugular venepuncture with an evacuated tube system three times during dry season and four times in wet season. This sampling was done at 07:00 in the morning after the animals had fasted overnight. The blood variables studied were chosen for various nutrition related reasons: Firstly, total protein reflects availability of protein and the decline in its concentration indicate protein deficiency (Ndlovu et al., 2007). Secondly, high blood urea levels indicate a high protein intake or excessive mobilization of muscle (Chimonyo et al., 2002). In ruminants a decrease in the blood urea concentration is related to low dietary protein intake due to recycling of urea from blood back to the rumen when dietary protein is low (Kohn et al., 2005). Thirdly, insufficient nutrient intake can reduce circulatory glucose and cholesterol levels. Lastly, NEFAs are released into the circulation as a direct result of lipid catabolism (Ndlovu et al., 2007). Ethylene diamine tetra acetic acid (EDTA) tubes were used to collect blood for glucose determination while clot activator tubes were used to collect blood for determining the concentrations of serum urea, total protein and non-esterified fatty acids (NEFA). All blood metabolites were assayed by the Cobas Integra 400/700/800 analyser using only the standard methods. Blood sampling was conducted by a trained veterinary assistant under permit number: APIEC11/039 provided by the Animal Ethics Committee of the ARC, South Africa.

### 2.4. Data analysis

A General Linear Model (GLM) with treatment as a fixed factor, paddock (i.e. where the animals foraged) as random factor, and ADG as a dependent variable was used. GLMs with repeated measures were used to test for differences in live weight, faecal nitrogen, blood glucose, blood urea, blood total protein and blood NEFA. In each model, time (i.e., day of measurement) was used as the within subject variable and treatment as the between-subject factor. Since different animals were used in dry and wet seasons and since measurements were conducted at different intervals for the two seasons, separate models were used for different seasons. In all models, unstandardized residuals

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