Technical Note

Digestibility and voluntary intake of vine leaves (Vitis vinifera L.) by sheep

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Abstract

Twelve castrated adult male (62±3 kg body weight) Manchego sheep were used in a digestion trial. Diets consisted of vine leaves or vetch-oat hay alone, and vetch-oat hay and vine leaves in a ratio of 1:1. Voluntary intake of vine by-products by eight females Manchego sheep was also measured. The CP level was greater in vetch-oat hay (73 g kg⁻¹ DM) than vine leaves (68 g kg⁻¹ DM) and the vine leaves had a higher level of neutral-detergent insoluble crude protein (NDF-CP, 38 g kg⁻¹ DM) than vetch-oat hay (17 g kg⁻¹ DM). NDF was less in vine leaves (319 g kg⁻¹ DM) than in vetch-oat hay (566 g kg⁻¹ DM). However, the lignin content of cell walls in vine leaves (146 g kg⁻¹ NDF) was higher than hay (127 g kg⁻¹ NDF). The content of condensed tannins of the vine leaves (50 g kg⁻¹ DM) was 10 times higher than that of the vetch-oat hay. DMD and OMD of vine leaves was 42.2 and 47.2%, respectively. Apparent digestibility of CP in vine leaves was negative (−25.8%) and was lower (P<0.001) than CP digestibility of vetch-oat hay (48.9%). Digestibilities of NDF and ADF of vine leaves were 3.8 and −0.6%, respectively. These values were lower (P<0.001) than those obtained for vetch-oat hay (53.9 and 54.7%, respectively). DM and DOM intake of vine leaves was 1010.7 and 415.3 g per day, respectively. With the exception of urea, there were no differences (P>0.05) between sheep fed leaves and those fed vetch-oat hay in measured plasma metabolites. These results suggest that the low digestibility of the vine leaves is due to several factors, such as the high level of condensed tannin content and low protein digestibility.

Keywords: Vine leaves; Vetch-oat hay; Condensed tannins; Digestibility; Voluntary intake; Blood metabolites

1. Introduction

In Mediterranean countries, sheep nutrition is based on natural grazing and supplementary feeding consisting mainly of roughage, by-products and concentrates. The roughage usually used is hay mainly consisting of annual sown legumes and small grain cereals such as common vetch-oats. Caballero and García (1996) concluded that common vetch is the legume used by most farmers (90%) and oat is the main species used as a companion crop (50%) in areas of Central Spain.

Grapes are widely grown in the Mediterranean basin, producing considerable quantities of by-products. Grapevines consist of branches and leaves from annual pruning of vineyards. Traditionally, this residue is grazed by sheep or goats after the harvest.
The most important feature of the by-product of grapes is their low energy value reflecting a high cell wall content and low digestibility of the cell wall fraction. This is associated with a high content of lignin and other antinutritional factors, such as tannins (Alvira et al., 1983).

Tannins, a naturally-occurring polyphenolic compound in vine by-products, are present either in hydrolyzable or condensed forms (Butler, 1989). They form complexes with proteins, polysaccharides, amino acids, fatty acids and nucleic acids (Flores et al., 1994). Both the composition and the extent of polymerization of tannins are important factors in determining their ability to form such complexes. In addition, there is an inverse relationship between high tannin level in forage and palatability, digestibility and voluntary intake. In the present study, the digestibility and intake of vine tree leaves were measured when fed alone and in a mixed ration with vetch-oat hay, and the blood metabolic profile in sheep consuming vine leaves was examined.

2. Materials and methods

Vetch-oat (Vicia sativa L.–Avena sativa L.) hay harvested in mid May at early-seed and full-bloom stages for vetch and oat, respectively, was used. The botanical fractions of the hay were 65% of oat and 35% of vetch. Vine tree (Vitis vinifera L.) leaves from annual pruning (performed in October) after harvest were used. Stems were discarded leaving only the leaves as fresh feed.

Twelve castrated adult male (62±3 kg body weight) Manchego sheep were used in the digestion trial. A completely randomized design was used and the animals were assigned to three diets consisting of vine leaf or vetch-oat hay alone, and vetch-oat hay and vine leaf in a ratio of 1:1. The hay was cut into 2–5 cm lengths. Animals were in individual digestibility cages. The digestibility trial lasted 21 days (14 days for acclimatization and 7 days for the experiment); feces were collected and daily intake controlled. Feed was offered at an intake level close to maintenance (40–45 g DM kg\(^{-1}\) LW\(^{0.75}\) per day) as recommended by Van Es and Van Der Meer (1980).

In the voluntary intake trial, eight female Manchego sheep with an initial body weight of 46±3 kg were used. Animals were kept in individual pens and fed with a ration consisting of vine leaves with barley grain-supplement at a level of 200 g DM per animal per day. The intake trial lasted 44 days (14 days for acclimatization and 30 days for the experiment), during that time daily intake was measured. Feeds were offered ad libitum daily to sheep in sufficient quantities to enable them to refuse at least 200 g DM kg\(^{-1}\) DM of feed offered. The amount of feed offered was adjusted daily on the basis of the intake of the previous day to achieve the target refusal rate. Feed refusals were collected before feeding. In both trials sheep were fed once daily at 10:00 hours and had free access to water and a mineral block. All the animals used in the experiments were vaccinated and drenched.

Blood samples (10 ml) were taken from each sheep used in the digestibility and intake experiments. In addition, a control group fed with vetch-oat hay was used for the sheep intake trial. Blood was taken at 09:00 hours during the last 2 days of each experiment from the jugular vein by a syringe, and the plasma was separated and stored at −20°C.

Feces were collected daily from each animal and dried in a forced air oven at 60°C for 48 h and ground through a 1 mm screen. Daily fecal samples (20% of the total) together with a sample of the feed offered, were stored for subsequent analyses.

The feces and feed samples were analyzed for dry matter (DM), ash, crude protein (CP) and ether extract (EE) according to AOAC (1984) methods. To determine cell components, neutral detergent fiber (NDF), acid detergent fiber (ADF) and permanganate lignin the methods proposed by Van Soest et al. (1991) were followed. Acid-detergent insoluble nitrogen (ADIN) and the neutral-detergent insoluble nitrogen (NDIN) were measured (Licitra et al., 1996), and these values were given in terms of crude protein (ADF-CP and NDF-CP, respectively). The vanillin–HCl (Broadhurst and Jones, 1978) method was used for determination of condensed tannins (TC).

The plasma was analyzed at the Clinical Pathology Laboratory of Murcia Veterinary University (Murcia, Spain), by procedures conventionally used for diagnosing domestic animal hepatic and kidney damage and general metabolic disorders (Kaneko, 1989). Components measured were total protein, albumin, cholesterol, glucose, urea and creatinine. In addition, the plasma enzymes aspartate aminotransferase
(AST), alkaline phosphatase (ALP) and gamma glutamyltranspeptidase (GGT) were measured.

All data were subjected to one way analysis of variance (ANOVA) (Steel and Torrie, 1980). Differences among means were tested using Duncan’s new multiple range test.

3. Results and discussion

CP content of vine leaves was 68 g kg\(^{-1}\) (73 g kg\(^{-1}\) for hay). These values agree with the values reported by Rebolé et al. (1988) for samples of grapevines (branches and leaves), but CP content of vine leaves is higher than those obtained by Rebolé (1994) for vine branches, either fresh (60 g kg\(^{-1}\)) or ensiled (50 g kg\(^{-1}\)). The geographic area or the season in which it is collected influence the chemical composition of vine by-product (Alvira et al., 1983).

NDF and ADF levels were 319 and 254 g kg\(^{-1}\) for vine leaf (566 and 364 g kg\(^{-1}\) for hay). The proportion of lignin content of cell wall in the vine leaf (146 g kg\(^{-1}\) NDF) was higher than in the hay (127 g kg\(^{-1}\) NDF). The content of condensed tannins of the vine leaves (50 g kg\(^{-1}\)) was 10 times higher than that of the vetch-oat hay (5 g kg\(^{-1}\)). Similar values were found by Rebolé (1994) for fresh vine branches.

Digestibilities of vine leaves, vetch-oat hay and vine leaves/vetch-oat hay (1:1) diets are given in Table 1. DMD and OMD of vine leaves were lower (\(P<0.001\)) than those of vetch-oat hay. According to these data, vine leaves could be considered low quality roughage (Ellis et al., 1988). However, DMD of vine leaves is higher than those obtained by Rebolé et al. (1988) for ensiled grapevines, due to the fact that this by-product contains leaves and branches. CP apparent digestibility of vine leaves was negative (\(-25.8\%)\) and was lower (\(P<0.001\)) than the CP digestibility of vetch-oat hay (48.9%). This result may be due to several factors. Firstly, the vine leaves had a higher level of NDF-CP (38 g kg\(^{-1}\), a 56% of CP is bound to cell wall) than that of vetch-oat hay (17 g kg\(^{-1}\)). Rebolé et al. (1988) reported 4 g kg\(^{-1}\) for NDF-CP for grapevines, equivalent to 61.5% of CP bound to the cell wall, and they found that digestibility of CP in NDF was negative (\(-16.4\%). Licitra et al. (1996) indicated that the nitrogen associated with NDF is normally cell wall-bound protein, which includes the slowly degradable protein and the indigestible nitrogen found in the acid-detergent residue. Secondly, it could be due to the presence of condensed tannins in vine leaf. These compounds form complexes with proteins and carbohydrates (Makkar et al., 1996), decreasing the available nitrogen and energy for rumen micro-organisms.

The low digestibilities of NDF and ADF of vine leaves might be due to the high content of lignin and tannin content in this by-product.

DM and DOM intake of vine leaves was 1010.7 and 415.3 g per day, respectively. The presence of tannin

<table>
<thead>
<tr>
<th>Item</th>
<th>Vine leaf:vetch-oat hay</th>
<th>S.E.M.(^d)</th>
<th>(P^e)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Apparent digestibility (%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dry matter</td>
<td>42.2(^ab)±1.7</td>
<td>51.1(^b)±0.7</td>
<td>58.4(^c)±0.4</td>
</tr>
<tr>
<td>Organic matter</td>
<td>47.2(^b)±1.7</td>
<td>56.5(^b)±0.7</td>
<td>61.2(^c)±0.6</td>
</tr>
<tr>
<td>Crude protein</td>
<td>(-25.8)^a±2.9</td>
<td>17.9(^b)±2.9</td>
<td>48.9(^c)±1.7</td>
</tr>
<tr>
<td>NDF</td>
<td>3.8(^b)±2.9</td>
<td>34.1(^b)±0.8</td>
<td>53.9(^c)±1.2</td>
</tr>
<tr>
<td>ADF</td>
<td>(-0.6)^a±6.4</td>
<td>31.9(^b)±0.4</td>
<td>54.7(^c)±1.1</td>
</tr>
<tr>
<td>Cellulose</td>
<td>47.1(^a)±5.4</td>
<td>39.3(^a)±2.9</td>
<td>52.6(^c)±1.9</td>
</tr>
<tr>
<td>Hemicellulose</td>
<td>19.7(^a)±15.1</td>
<td>500(^b)±6</td>
<td>566(^c)±11</td>
</tr>
</tbody>
</table>

\(^{a,b,c}\) Means in the same row with different superscripts are significantly different.
\(^d\) Standard error of means.
\(^e\) NS: \(P>0.05\); *: \(P<0.05\); ***: \(P<0.001\).
in a forage has been assumed to affect voluntary intake (McLeod, 1974). However, in this trial intake problems were not observed.

There were no differences between sheep fed with leaves and vetch-oat hay in the digestibility trials in any plasma metabolite, except urea (Table 2). In sheep fed with hay, the urea concentration (29.5 mg 100 ml$^{-1}$) was higher ($P<0.01$) than in sheep fed with leaves (8.9 mg 100 ml$^{-1}$), which is most likely related to the limitation in protein digestibility in sheep fed vine leaves. Similar results were found by Silanikove et al. (1996) where urea concentration was higher in goats fed with tannin-rich leaves than when fed wheat straw.

In the intake trial more plasma metabolic parameters showed significant differences than in the digestibility trial. A possible explanation for these higher differences could be due to the experimental period in intake trials of vine leaves (44 days), which was higher than in the digestibility trial (21 days). Specifically, plasma urea nitrogen and total protein decreased ($P<0.01$), and creatinine increased ($P<0.001$). This effect, during consumption of leaves, could be explained by negative nitrogen balance. The increase of creatinine observed can be related with renal failure, but the level found in the present study fell within the normal range for sheep (Kaneko, 1989). No significant changes in plasma enzymes AST and GGT were found. These enzymes are used to detect if tannin-related hepatotoxicity occurred (Zhu and Filippish, 1992).

It is concluded that the low digestibility of the vine leaves might be due to factors such as the high levels of lignin and condensed tannins, and low protein digestibility. However, this by-product was accepted by sheep and toxic effect was not evidenced in this study.

### References


