

## Ability of Different Types and Doses of Tannins to Modulate In Vitro Ruminant Biohydrogenation in Sheep

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Based on the potential benefits to consumer health, there is considerable interest in developing nutritional strategies for modulating the fatty acid (FA) composition of ruminant-derived products. Several reports have suggested that feeding tannins can favourably alter ruminal biohydrogenation (BH) of dietary unsaturated FA, enhancing accumulation of *trans*-11 18:1 in the rumen and thereby the content of *cis*-9 *trans*-11 conjugated linoleic acid (CLA) in milk or meat (Khiaosa-ard *et al* 2009, Vasta *et al* 2009). However, there is a large controversy surrounding the efficacy of these phenolic compounds, which has been mainly attributed to their type and amount in the diet (Kronberg *et al* 2007, Toral *et al* 2011, 2013). This assay was therefore conducted to study the effect of different types and doses of tannins with the aim of selecting an effective treatment to modulate the ruminal BH of dietary unsaturated FA.

Four commercial oenological extracts of tannins [2 hydrolysable (chestnut and oak) and 2 condensed (quebracho and grape)] were added (at 0, 2, 4, 6 or 8% of the DM incubated) to a diet supplemented with 2% DM of sunflower oil, and their effect on BH was examined using batch cultures of rumen microorganisms. Five ewes fed a TMR similar to that incubated (forage:concentrate ratio 50:50) were used as donors of ruminal inocula. Cultures were repeated on 3 different days (run = replicate), and freeze dried residues of in vitro ruminal digesta were analysed for FA composition (Toral *et al* 2012).

After 12 h incubations (when, according to preliminary data, effects were better detected), all tannins and doses, with the exception of quebracho8%, proved to be able to slightly reduce the concentration of 18:0 ( $P < 0.05$ ). Low and moderate doses increased concentrations of 18:2n-6 and 18:3n-3 (up to 90 and 71%, respectively;  $P < 0.01$ ). However, only the 6% and 8% doses augmented *cis*-9 18:1. Likewise, concentrations of *cis*-9 *trans*-11 18:2 were only favoured by quebracho6%, chestnut6%, grape8% and oak8% ( $P < 0.001$ ), the highest value representing an increase of 128% compared to the control. A tendency to a greater accumulation of *trans*-11 18:1, the target FA in the rumen according to Khiaosa-ard *et al* (2009) and Vasta *et al* (2009), was only detected with grape6% and 8%, chestnut6% and 8%, and oak2% (up to 15.5%;  $P < 0.10$ ). An irregular behaviour was observed for *trans*-10 18:1: some treatments increased its concentration but some others, such as quebracho, chestnut and oak at 2%, decreased it ( $P < 0.001$ ). All these changes are supposed to be due to the effects of tannin extracts on rumen microbiota, as supported by variations, in many cases, in odd and branched-chain FA concentrations. Overall, the results would suggest a general inhibition of the rumen microbial BH of dietary unsaturated FA rather than a specific inhibition of the conversion of *trans*-11 18:1 to 18:0, which is in line with observations by Kronberg *et al* (2007) when trying to lessen the ruminal reduction of 18:3n-3. Other in vitro assays (e.g., Khiaosa-ard *et al* 2009, Vasta *et al* 2009) showed a beneficial impact of tannins on BH at very high doses (up to 18% DM). Nevertheless, such high levels would be impractical under farm conditions.

The tannin  $\times$  dose treatment showing a more promising behaviour was oak2%. This combination increased total polyunsaturated FA, 18:3n-3, 18:2n-6 and *trans*-11 18:1, and decreased 18:0 and *trans*-10 18:1 concentrations. Further studies are now necessary to make sure that these effects are extended to in vivo conditions and that the treatment does not impair ruminal fermentation or animal performance.

To conclude, the examined oenological tannin extracts seem to be able to modulate in vitro ruminal biohydrogenation of dietary unsaturated FA. On the other hand, the high dose required in most cases suggests that their efficacy would be rather limited in terms of animal feeding. Further research would still be recommended for extracts exerting a desirable effect at low doses, such as that of oak tannins at 2% of the DM incubated.

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