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Effects of clays used as oil adsorbents in lamb diets on fatty acid composition of abomasal digesta and meat

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ABSTRACT

This experiment was designed to test the hypothesis that using clays as vegetable oil adsorbent results in a partial protection of polyunsaturated fatty acids from rumen biohydrogenation (BH). Complementarily, we also hypothesized that the addition of clays to high-concentrate high-oil diets attenuates the *trans*-10 shift of rumen BH pathways. Therefore, we compared the effect of using bentonite and/or vermiculite as dietary oil (60 g/kg of added sunflower and linseed oil blend, 1:2 v/v) adsorbent on the fatty acid (FA) composition of lamb's abomasal digesta and meat. For that, we used 40 Merino Branco ram lambs, randomly allocated to 20 slatted boxes. The four established diets resulted from a completely randomized design: C (no clay), B (30 g/kg bentonite), V (30 g/kg vermiculite) and BV (15 g/kg bentonite plus 15 g/kg vermiculite). Pen was considered as the experimental unit. None of the diets affected ($P > 0.05$) animal performance or meat quality traits although BV treatment resulted in higher proportions of high price joints in the carcass ($P = 0.024$) and muscle ($P = 0.036$) and lower proportions of kidney knob channel fat ($P = 0.014$) and total dissectible carcass fat ($P = 0.046$). No differences among diets were observed on rumen volatile FA and protozoa counts. Only minor effects of diets on FA composition of abomasal digesta were observed. Diet B reduced *t*11-18:1 ($P < 0.05$) while diet BV reduced ($P < 0.05$) *t*10-18:1 and tended ($P = 0.054$) to increase 18:0. Total BH intermediates tended ($P = 0.060$) to be reduced by diet BV while diet B clearly increased ($P < 0.001$) the *t*10-18:1/*t*11-18:1 ratio. No effect was observed in BH of 18:2n-6 or 18:3n-3. Treatments had no effect on total meat lipids and the FA profile of meat presented was very similar among treatments. Meat samples from animals from all diets presented high contents of *t*10-18:1 (≈11 g/100 g of total FA) and *t*10,*c*15-18:2 (≈2.0 g/100 g of total FA), whereas the contents of *t*11-18:1 (≈0.9 g/100 g of total FA) and *c*9,*t*11-18:2 (≈0.2 g/100 g of total FA) were quite low. No effect was observed in 18:2n-6, 18:3n-3 and long chain PUFA contents in meat. Our results show that both bentonite and/or vermiculite used as vegetable oil adsorbents in high concentrate based diets were ineffective to protect PUFA from rumen BH as well as to prevent the *trans*-10 shift.

Abbreviations: ADF, acid detergent fibre; B, diet with 30 g/kg of bentonite; BH, biohydrogenation; BV, diet with 15 g/kg of bentonite and 1.5 g/kg of vermiculite; C, control diet; DM, dry matter; FA, fatty acid; NDF, neutral detergent fibre; PUFA, polyunsaturated fatty acid; V, diet with 30 g/kg of vermiculite; VFA, volatile fatty acids.

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1. Introduction

Edible fats from ruminants are characterized by being rich in saturated and *trans* fatty acids (FA) that have been associated with increased risk of cardiovascular diseases (FAO, 2010), despite the fact that individual saturated and *trans* FA might differ in its biological effects (Gebauer et al., 2011). Therefore, developing strategies to decrease saturated FA in ruminant products is an important research target. Modification of ruminant's fat composition through nutrition is possible but is strongly limited by the fact that rumen microbial ecosystem extensively isomerises and hydrogenates dietary polyunsaturated FA (PUFA). During rumen biohydrogenation (BH), conjugated linoleic acid isomers and *trans* octadecenoates are formed. Some of these BH intermediates, like rumenic acid (*c*9,*t*11-18:2) but also vaccenic acid (*t*11-18:1) possess an anti-carcinogenic effect on several animal and cell culture models (Gebauer et al., 2011; Lim et al., 2014). The most effective way to increase meat *c*9,*t*11-18:2 content (the main conjugated linoleic acid isomer) is by supplementing forage based diets with unsaturated vegetable oils (Bessa et al., 2005; Bessa et al., 2015). In those conditions, dietary PUFA are extensively isomerised and partially hydrogenated, with a large accumulation of *t*11-18:1 that, after being absorbed in the small intestine, is extensively converted to *c*9,*t*11-18:2 by the action of delta-9 desaturase enzyme (Bessa et al., 2015). However, intensive animal production often uses diets based on compound feeds rich in starch, leading to modifications in BH pathways that result in an accumulation of *t*10-18:1 instead of *t*11-18:1 (i.e., *trans*-10 shift) (Aldai et al., 2013). As the *t*10-18:1 isomer cannot be endogenously converted into conjugated linoleic acid isomers, the establishment of the *trans*-10 shift results in a large accumulation of *t*10-18:1 and in a low content of *c*9,*t*11-18:2 in tissues (Bessa et al., 2015). Furthermore, contrasting with *t*11-18:1, the *t*10-18:1 isomer is potentially harmful to consumer's health (Aldai et al., 2013; Mapiye et al., 2015). Thus, strategies to mitigate the production of *t*10-18:1 in animals fed diets based on compound feeds need to be developed.

Bentonite and vermiculite are three-layer minerals with an expanding lattice belonging to the smectite group (Murray, 2006). The special properties of this group of clay minerals, such as high layer charge, medium to high cation exchange capacity and high surface area, are responsible for their high absorptive capacity and medium to high swelling capacity, making then valuable materials for a wide range of applications in industrial and farming systems. Bentonite has been used extensively to improve the binding quality of feed pellets, but also tentatively as rumen buffering agent to mitigate acidosis and milk fat depression (Bringe and Schultz, 1969). Vermiculite has been used as a fat carrier in order to mitigate the negative impact of fat supplements on rumen digestion (Tamminga et al., 1983; Jenkins and Palmquist, 1984).

More recently, Sinclair et al. (2005) reported that using vermiculite as a carrier for linseed oil resulted in partial protection against rumen BH. The putative effects of clays in protecting PUFA from rumen BH might occur due to a slower release of triacylglycerols or through PUFA saponification with Mg ions, released due to its high exchangeable ion capacity. Our team tested the effects of bentonite inclusion in high-forage high-oil lambís diets and did not detect any major effect on lamb meat FA profile, apart from a significant reduction of *t*10-18:1 (Jerónimo et al., 2010). In that experiment, besides using only bentonite, all diet ingredients were mixed together and thereafter pelleted. Thus, we hypothesized that, in order to achieve a protective effect against rumen BH, the dietary oil needs to be previously adsorbed into the clays and that vermiculite differs from bentonite regarding to their potential to protect the dietary oil from rumen BH.

The classic literature explored the effects of bentonite on mitigation of diet-induced milk fat depression in dairy cows (Bringe and Schultz, 1969; Rindsig et al., 1969). It is now well established, that diet-induced milk fat depression is closely linked to the occurrence of the *trans*-10 shift in the rumen (Bauman and Griinari, 2003). Moreover, our team had reported that bentonite reduced *t*10-18:1 concentration in lamb meat (Jerónimo et al., 2010), thus we hypothesized that inclusion of clays into high starch diets might mitigate the *trans*-10 shift that frequently occurs with those types of diets. Therefore, in the present experiment we compared the effect of using bentonite and/or vermiculite as dietary oil adsorbents on FA composition of lamb's abomasal digesta and meat.

2. Material and methods

2.1. Animals and diets

The trial was conducted at the facilities of Escola Superior Agrária de Coimbra, Portugal. Animal handling followed EU Directive 86/609/EEC concerning animal care. Forty Merino Branco ram lambs, with an average initial live weight of 12.2 ± 1.61 kg and ageing 60 ± 6.2 days (mean ± SD) were randomly assigned to 20 slatted pens. After one week of adaptation to the experimental conditions, in which lambs were dewormed with Ivomec® (Merial Portuguesa, Portugal), lambs stayed on trial for 45 days. Lambs were weighed at the beginning of the trial and then weekly before the morning feeding without fasting. During the trial, animals had fresh water always available and were fed once a day (at 09:00) with a diet containing 9 parts of a compound feed and 1 part of wheat straw. The four compound feeds used were named as C (no clay), B (30 g/kg of bentonite), V (30 g/kg of vermiculite) and BV (15 g/kg of bentonite + 15 g/kg of vermiculite) and were randomly allocated to the pens following a completely randomized design. Compound feed were produced in a commercial feed mill; the oil blend (linseed and sunflower oil, 2:1) was mixed with clays and then added to the compound feed in a vertical mixer before being pelleted (3 mm diameter, cold pelleting with no steam added). Ingredients and chemical composition of the feeds used in this trial are presented in Table 1. Straw and concentrate were given separately and the amounts offered and refused were recorded daily. Diets were provided ad libitum (110% of the consumption of the day before).