

EFFECTS OF ESSENTIAL OILS ON DRY MATTER INTAKE AND THE MILK PRODUCTION AND COMPOSITION IN LACTATING RUMINANTS: A REVIEW

EFEITOS DOS ÓLEOS ESSENCIAIS SOBRE O CONSUMO DE MATÉRIA SECA, PRODUÇÃO E COMPOSIÇÃO DO LEITE DE RUMINANTES: UMA REVISÃO

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ABSTRACT

This review gives an overview of several experiments conducted in vivo on the effects of plants rich in essential oils (EO) or of EO extracts on dry matter intake, milk production and composition. Based on in vivo studies on the effects of EO or whole plants rich in EO in lactating ruminants, it was possible to conclude that: i) milk yield was often positively affected after the first part of the lactation, in long-term studies, at high dosages of EO, and in sheep and goats rather than in dairy cows; and ii) milk composition was marginally affected, except for milk fatty acid composition, for which all studies on lactating goats and ewes, but none of those on cattle. In general, it appears that small ruminants are more responsive to the action of EO.

KEYWORDS: Milk composition, milk production, plant extracts.

RESUMO

Esta revisão fornece uma visão geral de vários experimentos realizados in vivo sobre os efeitos ricas em óleos essenciais (OE) ou em extratos de OE no consumo de matéria seca, produção e composição do leite de ruminantes. Com base nos estudos in vivo sobre os efeitos dos OE ou das plantas ricas em OE nos ruminantes, foi possível concluir que: i) a produção de leite foi frequentemente afetada positivamente depois da primeira fase de lactação em estudos de longa duração e com altas doses de OE, principalmente em ovinos e caprinos do que em bovinos de leite; e ii) a composição do leite foi pouco alterada, porém em todos os estudos com cabras e ovelhas em lactação houve alteração na composição dos ácidos graxos, enquanto que não houve nenhuma alteração nos estudos com bovinos. Em geral, parece que os caprinos e ovinos são mais sensíveis à ação dos OE.

PALAVRAS-CHAVE: Extratos de plantas, composição de leite e produção de leite.

INTRODUCTION

For a long time nutritionists have worked on the modification of rumen environment as a way of improving feed efficiency (BENCHAAAR et al., 2008a). Among the various compounds and additives studied to modify rumen environment and improve feed utilization efficiency, plant extracts have gained widespread interest. They have been also considered the only alternative to antibiotics (WALLACE, 2004). This Introduction will consider, in particular, the effects of plants rich in essential oils (EO) and of their extracts on dry matter intake, milk production and composition in lactating ruminants.

Essential oils are secondary metabolites of very diverse composition, usually isolated by stem distillation or solvent extraction, made up mainly by volatile terpenoids and phenylpropanoids (CALSAMIGLIA et al., 2007; BENCHAAAR et al., 2008a; PATRA, 2011). The composition and amount of EO of plant extracts vary with plant parts (e.g. leaf, root, stem, fruit peel or pulp, flower or seed) (DORMAN & DEANS, 2000) and plant species (BEZIC et al., 2005) and they are strongly affected by genetics, age and environmental factors (COSENTINO et al., 1999). Various properties and modes of action have been associated to EO (CALSAMIGLIA et al., 2007): i) antioxidant action, with scavenging of free radicals, inhibition of peroxidation of membrane lipids, stimulation of antioxidants enzymes, ii) activity against bacteria, mainly gram +, because EO occupy space in their hydrophobic cell membrane, fluidizing it and allowing leakage of ions, thus forcing bacteria to spend energy for ionic pumps and decreasing the energy available for their growth. In some cases (e.g. EO carvacrol and thymol), the hydrophilic external cell membrane of gram – bacteria can be disrupted, iii) denaturation and coagulation of cell protein constituents, and iv) inactivation of nucleic acids and proteins.

Essential oils has been fed to animals as: i) whole or part of plants containing EO. In this case nutrients other than EO are also supplied, content of EO is variable and rumen utilization and fermentation is slower than when oil extracts

are used, ii) oil extracts from specific plants, with variable content of EO and potentially high interaction with rumen microorganisms, iii) specific mixes of selected EO, normally sold as commercial products. They have constant and specific content of EO, with potentially high interaction with rumen microorganisms, and iv) single compounds, with known concentration and potentially high interaction with rumen microorganisms.

Many studies have been published on the use of EO, supplied in the various forms described above, on rumen fermentation and production performance, but most of them regard in vitro trials (CARDOZO et al., 2005, 2006; CASTILLEJOS et al., 2005; BUSQUET et al., 2006; BENCHAAAR et al., 2007a; CASTILLEJOS et al., 2008; PATRA et al., 2010; BHATTA et al., 2012). On this regard, extensive reviews have been published in the last years (CALSAMIGLIA et al., 2007; BENCHAAAR et al., 2008a; PATRA, 2011). Therefore, the main objective of this review is to give an updated overview of experiments conducted in vivo on the effects EO on dry matter intake, milk production and composition in lactating ruminants with a special emphasis on possible differences among ruminant species. The results of this review are synthesized in Tables 1 to 3.

EFFECTS OF ESSENTIAL OILS ON DRY MATTER INTAKE AND THE MILK PRODUCTION AND COMPOSITION IN LACTATING RUMINANTS

Different types of essential oil sources have been used in lactating ruminants (Tables 1 and 2). Several experiments used EO commercial complexes, whereas others used plant extracts or, only in one case, unprocessed leaves of plants containing EO Dairy cows Table 1 reports the main results of 9 studies carried out on lactating cows. The studies differed for the EO source, for the EO dosage, and for the forage to concentrate ratio of the diets. They also differed for lactation stage and production level of the cows.

Few studies observed differences in dry matter intake (DMI) (Table 1). Santos et al. (2010) observed a non-significant decrease (-1.43 kg/d;

P=0.13) and Tassoul and Shaver (2009) found a significant (-1.8 kg/d; P<0.05) decrease in DMI as a result of the inclusion of commercial EO complexes in the diets. In addition, Hristov et al. (2013) observed a linear decrease in DMI as the dosage of *Origanum vulgare* leaves increased in the diets. In contrast, Kung et al. (2008) observed an increase in DMI (+1.9 kg/d; P<0.05) as a result of the addition of a commercial EO complex. No obvious reasons could be found to explain these differences. In particular, for an EO commercial complex such as Crina (Crina S.A., Gland, Switzerland; see note 2 of Table 1 for its characteristics) DMI increased in one experiment, did not change in a second one and decreased in a third one (Table 1). It appears that the variation in DMI were not driven by the EO complex used.

Milk production was affected only in one experiment, where it tended to increase (+1.9 kg/d; P<0.16) when the EO commercial complex Crina was used (KUNG et al., 2008). The authors suggested that earlier studies did not affect milk yield because they had been carried out in early lactation, whereas their study was carried out in mid-lactation. The importance of the stage of lactation was in part confirmed by Tassoul and Shaver (2009) in a long-term experiment using Crina as a source of EO. They found that until the 7th week of lactation milk yield was numerically lower in the EO diet, but from the 8th to the 15th week of lactation milk yield became progressively numerically higher in the EO group. Unfortunately, they interrupted the experiment when the difference was increasing at a high rate. All the other experiments which used EO complexes did not report milk yield differences, but they were all carried out in early lactation (Table 1). The only study that compared stages of lactation (51 DIM vs. 247 DIM) was that of Hristov et al. (2013), who used 3 dosages of *Origanum vulgare* leaves. They did not observe any milk yield or milk composition differences associated with the treatments, except for a linear decrease in DMI as the dosage of *Origanum vulgare* increased. It is clear that the studies that used EO complexes might not be fully comparable with this study, which used leaves of a plant rich in EO. It should be pointed out that in the

study of Hristov et al. (2013), as in those of Santos et al. (2010) and Tassoul and Shaver (2009), there was a marked increase in feed efficiency utilization (kg milk/kg of feed) by the cows.

Regarding milk composition, in general the experiments did not show any effect of EO on milk fat and protein concentration (Table 1). Only Santos et al. (2010) found a significant, even though small, increase in milk fat concentration, whereas Tassoul and Shaver (2009) reported a significant but numerically small decrease of milk protein concentration. In general, EO did not cause variations in BCS or body weight (Table 1). Only Santos et al. (2010) found a significant increase in BCS for the cows which used an EO complex.

Overall, it appears that in most cases EO did not induce noticeable effects on production performance of lactating cows. A possible positive effect on milk production after the peak of lactation needs to be confirmed with other experiments.

Dairy Sheep

Few publications report studies on the utilization of EO on lactating ewes (Table 2). Two studies were based on the supply of EO extracts to dairy ewes: Giannenas et al. (2011) tested two dosages of the EO complex Crina, whereas Chiofalo et al. (2012) tested two dosages of EO extracts of *Rosmarinus officinalis*. In both cases, treatments affected milk yield but did not affect DMI. In the study of Giannenas et al. (2011) milk yield was not significantly affected at the lowest dosage (0.075 g/d per head of EO complex) but markedly and progressively increased above this dosage (+20% and 35% for the dosages 0.15 and 0.217 g/d per head of EO complex, respectively; Table 2). Interestingly, in this study there was also a marked reduction of somatic cell count for all dosages considered, suggesting that this was one of the causes of the increase in milk yield. In the other study, Chiofalo et al. (2012) observed a 10% significant increase in milk yield at the highest dosage (1.2 g/d per head of EO extracts) (Table 2). In this study there were some effects on milk composition too and BCS increased in both treated groups.

The study of Manca et al. (2012b) supplied the vegetative components (leaves and small twigs) of three plants (*Melissa officinalis*, *Ocimum basilicum* and *Thymus vulgaris*) at 3 dosages (50, 125 and 200 g/d, DM basis) to lactating dairy ewes. The plants were mixed with a different proportion of corn and pea meal, to form isoproteic mixes, and supplied individually (450 g/d of DM) to lactating dairy ewes. The results showed that the aromatic plants did not affect DMI, except for *Melissa officinalis*, which caused a 33% reduction of the intake of the mix when used at the highest dosage. This was probably caused by the very fibrous and ligneous structure of this plant. In addition, there were no effects of aromatic plants on milk yield and composition, except for a linear decrease of milk urea as the dosage of *Thymus vulgaris* increased and a decrease of milk lactose as the dosage of *Rosmarinus officinalis* increase (Table 2). However, the plants induced significant effects on milk fatty acid composition, as discussed later in this review.

Dairy Goats

Four studies were found in which EO were supplied to lactating goats. One was based on the supply of a blend of monoterpenes at two dosages to Alpine and Saanen goats and reported no effects on DMI, milk yield and its composition (MALECKY et al., 2009).

A second study (HEIDARIAN MIRI et al., 2013) supplied two dosages (at 1 and 2 g/L of rumen volume) of cumin seed extracts to lactating goats in early lactation. Milk yield was increased (+13%) by the lowest dosage (1 g/L of rumen volume). No effects were observed on milk fat and protein concentration, even though an increase in conjugated linoleic acid was observed for both dosages. A third study (BOUTOIAL et al., 2013) provided two dosages of rosemary (*Rosmarinus officinalis* spp.) to lactating Murciano-Granadina goats from parturition to seven months of lactation. Milk fat, protein and somatic cells did not change ($P>0.05$), with means of about 5.51%, 3.37% and 2.74 log SCC mL⁻¹, respectively.

In the fourth study, three oil extracts (oil

of cinnamon, garlic and ginger) rich in EO were compared with the control group (KHOLIF et al., 2012). Milk yield was significantly increased by all oils (+24%, +16% and +19% for cinnamon, garlic and ginger oils, respectively). In all treated groups, milk fat markedly decreased and milk protein increased even more strongly. The Authors attributed the decrease of milk fat concentration to both a dilution effect and a decrease of the rumen acetate to propionate ratio, and the increase in milk protein concentration to a possible increase in microbial synthesis group (KHOLIF et al., 2012).

EFFECTS OF ESSENTIAL OILS ON MILK FATTY ACIDS

Benchaar et al. (2007b) found on dairy cattle that the profile of milk fatty acids (FA) of cows was not influenced by supplementation with 750 mg per day of a mixture of EO compounds.

Similarly, Hristov et al. (2013) did not observe variations in milk fatty acids when supplementing the diet of lactating cows with three dosages of *Origanum vulgare* leaves.

In lactating Damascus goats, Kholif et al. (2012) found that supplementation with garlic oil, cinnamon oil or ginger oil increased unsaturated FA and conjugated linoleic acid (CLA), and that cinnamon oil also increased n3 linolenic acid.

Heidarian Miri et al. (2013) reported that cumin (*Cuminum cyminum*) seed extract fed at two doses (12.7 and 25.3 g/kg DMI) to lactating goats caused an increase in poly-unsaturated FA and CLA of the milk. In the study of Boutoial et al. (2013) on supplementation of rosemary (*Rosmarinus officinalis* spp.) to lactating Murciano-Granadina goats, polyunsaturated fatty acids (PUFA) increased ($P<0.05$) from 3.37 to 4.65 and then to 5.20 with increasing doses of 0, 10 and 20% of additive in the diet.

In the study of Manca et al. (2012a) on supplementation with vegetative parts of aromatic plants, the botanical species (*Melissa officinalis*, *Ocimum basilicum* and *Thymus vulgaris*) affected all milk FA groups (Table 3), except for the trans FA and the branched chain FA. In addition, as dose increased (50, 125 and 200 g/d, DM basis), branched

chain fatty acids increased, suggesting a supportive effect of the plants on microbial activity. There was also an increase in n3 poly-unsaturated FA and in the sum of conjugated linoleic acid isomers (Table 3) suggesting a reduction of biohydrogenation process, since these FA are intermediates of this process.

In the study of Boaventura Neto (2013) on supplementation with *Carum carvi* and *Coriandrum sativum* (seeds) and *Satureja montana* (vegetative parts) of aromatic plants, the botanical species affected only one class of milk FA. The long-chain FA (LCFA), whose proportions increased ($P=0.004$) in *Carum carvi* (+25%) and in *Satureja montana* (+17%) compared to the Control group. *Carum carvi* had also higher LCFA than *Coriandrum sativum*. Regarding the individual milk FA, the supply of aromatic plants caused a reduction ($P<0.05$) of the saturated FA 10:0, 12:0, 14:0 (neosynthesis), and an increase of the preformed 18:0. Nudda et al. (2013), in a study on goats, also showed a decreased in the same FA and suggested a benefit in milk FA profile in terms of human health, because, according to Tholstrup et al. (2003), these FA play an important positive role in the formation of blood cholesterol.

The atherogenic index (AI) is generally used as a measure of dietary fat quality. According to Addis et al. (2005), fat with a high AI value is harmful to human health. In this sense, our results showed a reduction ($P<0.05$) of about -18% of this index in *Carum carvi* and *Satureja montana* groups compared with the Control group. This pattern is similar to that observed by Addis et al. (2005) with sheep supplied with *Chrysanthemum coronarium* and by Nudda et al. (2013) with goats fed extruded linseed. In another study with lactating Sarda ewes, Boaventura Neto et al. (2014) when supplemented the diet with aromatic plants, *Carum carvi*, *Coriandrum sativum* and *Satureja montana* (125 g/head for each treatment), observed that Myristoleic, palmitoleic and linolenic acids were not affected ($P>0.05$), whereas the other FA were affected ($P<0.05$) by treatments. In most cases, the Control and *Satureja montana* groups had a similar pattern, and the *Carum carvi* and *Coriandrum*

sativum groups did not differ from each other. Lauric and myristic acids decreased by 26.2% and 14.3%, respectively, in the *Carum carvi* or *Coriandrum sativum* groups as compared with the Control or *Satureja montana* groups, which did not differ from each other. Stearic and oleic acids increased by 80.6% and 43.7%, respectively, in the *Carum carvi* and *Coriandrum sativum* groups as compared with the Control or *Satureja montana* groups. Eicosapentaenoic (EPA) increased by 50% in the *Satureja montana* group as compared with the other three groups, which did not differ from each other. Vaccenic acid increased by 95.2% and 163.9% in *Carum carvi* and *Coriandrum sativum* groups, respectively, as compared with the Control or *Satureja montana* groups, which did not differ from each other.

SOME COMMENTS ON EFFECTS OF ESSENTIAL OILS ON MILK PRODUCTION AND COMPOSITION

The review of Khiaosa-ard and Zebeli (2013) did not find effects of EO on milk yield and composition in dairy cows, except for an increase in milk protein concentration and yield, whereas production data on sheep and beef cattle were not sufficient for a meta-analysis.

The limited effects on milk yield and composition in dairy cows on one side and the marked effects in lactating ewes and goats reported above could be due to real differences between animal species or due to the different treatments applied.

In dairy cattle, a possible positive effect on milk production after the peak of lactation was suggested by the two experiments which evaluated EO complexes in mid lactation in long term studies (KUNG et al., 2008; TASSOUL and SHAVER, 2009). In the case of lactating sheep and goats, in all experiments which used EO extracts or oils there was an increase in milk yield, at least in one dosage. This effect was particularly strong (+35%) for the experiment with the highest duration 5 months, (GIANNENAS et al., 2011). In most studies the doses used were comparably higher and the experiments were longer in sheep and goats than in the case of cows (Tables 1 and

2). Based on this information, it is possible to speculate that EO complexes are more effective after the beginning of lactation, i.e. periods of positive energy balance, in long term studies and at high dosages. Ruminants in early lactation are generally in negative energy balance and mobilize body fat and this might interact with the utilization of EO. Indeed, both in sheep (PULINA et al., 2006) and cows (CHILLIARD, 1993) the effects of the addition of fat in the diet were more evident in mid-late lactation than in early lactation. It would be interesting to study if the mechanisms involved in this phenomenon are similar between EO and supplemented fat, being both compounds water-insoluble.

The fact that small ruminants responded consistently better than cows could be also due to the higher rumen passage rate of liquids and solids which characterize ruminants of small body size compared to those of larger size (VAN SOEST, 1994; CANNAS et al., 2003). The high rumen passage rate of sheep and goats might have reduced the interaction of EO at rumen level and increased their intestinal digestion. This would imply that the effects of EO are more related to an enhancement of metabolic pathways of milk synthesis than to modifications of the rumen environment, as often postulated. This hypothesis is supported by the studies reported above on milk FA. It appears that in the few studies available on dairy cows milk fatty acids were not affected, by the supply of EO, whereas all studies on lactating goats and ewes observed an increase of the unsaturation of FA and of CLA, suggesting that EO can reduce the biohydrogenation process, potentially improving the nutraceutical value of their milk. This animal species difference could be, also for FA, a result of the high feed rumen passage rate of small ruminants in comparison to large ones, which could limit the ability or the need of rumen bacteria to complete the biohydrogenation process.

CONCLUSIONS

Based on in vivo studies on the effects of EO or of whole plants rich in EO on ruminants, it is possible to conclude that:

Milk yield was often positively affected after the first part of the lactation, in long term studies, at high dosages of EO, and in sheep and goats compared to dairy cows;

Milk composition was marginally affected, except for milk fatty acid composition, for which all studies on lactating goats and ewes, but none of those on cattle, observed an increase on the unsaturated FA and of CLA concentrations, with possible reductions of the rumen biohydrogenation processes;

In general, it appears that small ruminants are more responsive to the action of EO than cattle, possibly due to differences in the rumen feed and liquid passage rate, usually higher in small than in large ruminants.

Compared to in vitro studies, in which frequent measurements are possible, the variables of interest of in vivo studies are often measured few times, for the difficulties of taking measurements with alive animals. In addition, in vivo studies have to deal with the complex and difficult-to-control ruminal environment as well as with variations in plant species, plant parts, oils and extracts tested. Despite these limitations, in vivo studies are fundamental for the transfer of the knowledge acquired to production conditions. Therefore, in the future it would be important to conduct in vivo studies with more frequent samplings, possibly taken at various intervals from the first supply of EO, and monitoring how and to what extent the rumen environment and the animals can adapt to these compounds, so that the mode of action of these compounds could be better understood.

ACKNOWLEDGMENT

I would like to thank Dr. Ana Helena Dias Francesconi for editing the English of this paper.

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