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Sunflower-seed oil, rapidly-degradable starch, and adiposity up-regulate leptin gene expression in lactating goats

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Abstract

We conducted experiments to evaluate the effects of lipid supplementation and the nature of starchy concentrate on the regulation of leptin synthesis in lactating goats. Multiparous goats in mid- to late lactation received diets based on different forages and containing plant oil or seeds rich in either 18:1*c*9, 18:2*n*-6 or 18:3*n*-3 corresponding to 3%–7% dry matter (DM) as lipid supplements, or diets based on concentrate as either rapidly or slowly degradable starch. The isoenergetic replacement of a part of the concentrate by either oleic sunflower-seed oil, formaldehyde-treated linseeds, or linseed oil did not modify leptinemia and the leptin mRNA concentration in adipose tissues, suggesting a lack of effect of 18:1*c*9, 18:3*n*-3, or their biohydrogenation products. Conversely, leptinemia and the leptin mRNA abundance were increased (by 20% and 140%, respectively, $P < 0.05$) in goats fed sunflower-seed oil under a grassland hay-based diet but not a maize silage-based diet, at similar energy intakes and adiposity. Thus, 18:2*n*-6 per se may up-regulate leptin gene expression, but the effect could be blunted by other fatty acids formed during the ruminal digestion of sunflower-seed oil when combined with maize silage. Consumption of rapidly but not slowly degradable starch increased (by 17%, $P < 0.05$) leptinemia. Moreover, during lactation, plasma leptin was positively correlated ($P < 0.05$) to adiposity parameters and negatively correlated to fiber intake. The results suggest that leptinemia responds poorly to nutritional factors in lactating goats, thus highlighting the physiological need to sustain hypoleptinemia during lactation.

Keywords: Leptin; Goat; Lactation; Dietary fatty acids

1. Introduction

Leptin is produced and secreted mainly by white adipose tissue in proportion to fat stores. Leptin was originally thought to act as the afferent signal in a feedback-loop-regulating adipose tissue mass by decreasing appetite and increasing energy expenditure. However, the ubiquitous distribution of leptin receptors in almost all tissues underlies the pleiotropism of leptin [1,2].

In ewes and cows, as in monogastric species, circulating leptin concentrations are positively related to daily energy intake in the short term and degree of adiposity and plane of nutrition in the long term [3]. We recently reported that in goats, lactation per se strongly decreased plasma leptin whatever the lactation stage, energy balance, milk production level, and pregnancy status [4]. We hypothesized that this hypoleptinemia could serve to increase productive efficiency and energy conservation during all lactation stages, not only for mammary function but also to promote the replenishment of body reserves [3]. However, to ascertain that hypoleptinemia is a key determinant of energy metabolism during lactation, potentially usable in animal production, it is necessary to

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determine whether leptinemia is regulated by extrinsic factors (nutrients, hormones) or intrinsic factors (adiposity) at either short or long term in lactating ruminants. To date, no relevant data have been published on goats, while the data available on lactating cows highlighted short-term regulations by extrinsic factors. Leptinemia during cow lactation was increased by jugular infusions of insulin [5,6], glucose [7], and propionate [8] and decreased by 3-day fasting or undernutrition [5]. Dietary lipids either increased [7], decreased [9], or had no effect [10–14] on cow plasma leptin depending on lipid type, energy balance, and lactation stage. Hence, to better understand the regulation of leptin gene expression during lactation in goats, the present study was designed with a 2-fold objective. First, we determined whether leptin gene expression in mid-lactating goats is modulated in the short-term by diets supplemented with starchy concentrate or diets containing plant oil or seeds rich in either 18:1*c*9, 18:2*n*-6, or 18:3*n*-3. Second, we examined the links between plasma leptin and body composition, blood, and milk parameters. These analyses allowed us to determine the major influencing factors involved in short- or long-term regulations of leptin gene expression during lactation in goats.

2. Materials and methods

2.1. Animals and diets

Five different experiments were performed on multiparous Alpine goats: 4 mid-lactation trials and 1 late-lactation trial (Table 1). These trials were originally performed to evaluate the effect of nutrition, and particularly lipid supplementation, on milk fat composition (Table 1) and lipid metabolism in the mammary gland [15–18].

Briefly, goats from trial 1 were fed an alfalfa hay-based diet with a 58:42 forage-to-concentrate ratio (dehydrated sugar beet pulp [47%], barley [29%], and soybean meal [24%]) (control diet), either with no additional lipid or supplemented with soybean seeds (rich in linoleic acid), representing an addition of 3.8% of dry matter (DM) as lipids for 3 weeks (Table 1, [15]). Goats from trials 2 to 5 were offered 3 experimental diets according to a 3 × 3 Latin square design with 21-d (trial 2) or 28-d (trials 3-5) experimental periods using 4 or 5 animals per group (Table 1). Each experimental period consisted of a 14-d (trial 2) or 21-d (trials 3-5) adaptation and a 7-d sampling period. Diets of trials 2 to 4 were composed of hay or maize silage (38%–58% of DM) offered ad libitum and a concentrate mixture (42%–62% of DM; dehydrated sugar beet pulp, barley, and soybean meal; Table 1) with no additional lipids or supplemented with plant seeds or oils providing an addition of 3.33% to 6.38% of DM as lipids. Trial 2 contained oleic sunflower-seed oil or formaldehyde-treated linseed (rich in oleic and linolenic acid, respectively [16]), whereas trials 3 and 4 contained sunflower-seed oil or linseed oil (rich in linoleic and linolenic acid, respectively [18]). In trial 5, 3 diets supplemented with 130 g/d sunflower-seed oil (ie. 7.4% to 8.9% of diet DM, Table 1) differed in the amount and nature of the concentrate, in particular the ruminal degradability of starch (Table 1 and [17]). Concentrate types and levels were: 0.82 kg/d of DM as corn grain (30%), dehydrated sugar beet pulp (6%), soybean meal (35%), and flattened wheat (29%) for the control goats; 1.28 kg/d of DM as corn grain (79%), dehydrated sugar beet pulp (7%), and soybean meal (14%) for goats fed slowly degradable starch; 1.32 kg/d of DM as dehydrated sugar beet pulp (6%), soybean meal (14%), and flattened wheat (80%) for goats fed rapidly degradable starch. Natural grassland hay was offered ad libitum (see reference [18] for its description and composition). Diets were offered as 2 equal meals at 8:30 AM and 4:30 PM.

The goats were housed in a metabolism unit in individual stalls with continuous access to water, and they were milked at 8:00 AM and 4:00 PM. Chemical composition of the milk and feed ingredients was determined using standard procedures outlined elsewhere [18]. Daily intakes and energy balances from each trial are reported in Table 1. All experimental procedures were approved by the Animal Care Committee of INRA in accordance with the Use of Vertebrates for Scientific Purposes Act 1985. At the end of the experiments, the goats were slaughtered.

2.2. Plasma measurements

Blood samples were collected into EDTA tubes (Venoject, C.M.L, Nemours, France) from the jugular vein at 7:30 AM the day before slaughter to determine plasma insulin (mean intra- and interassay coefficients of variation were 8.6% and 10.2% respectively; INSI-PR RIA kit, CIS bio International, Gif-sur-Yvette, France), leptin, and metabolites. Plasma leptin concentration was determined in duplicate according to the previously described disequilibrium double-antibody ovine-specific RIA validated for leptin determination in goat plasma [19]. The mean intra- and interassay coefficients of variation were 6.6% and 9.2%, respectively. Plasma concentrations of glucose, non-esterified fatty acids (NEFA), and beta-hydroxybutyrate (BHB) were determined enzymatically by the glucose dehydrogenase method (Glucose RTU kit; BioMérieux, Lyon, France),