SHORT COMMUNICATION

Relationships between the daily intake of unsaturated plant lipids and the contents of major milk fatty acids in dairy goats

Andrés L. Martínez Marín1, Nieves Núñez Sánchez1, Ana I. Garzón Sigler1, Francisco Peña Blanco1 and Miguel Angel de la Fuente2

1 Universidad de Córdoba, Departamento de Producción Animal. Ctra. Madrid-Cádiz, km 396. Campus de Rabanales, 14071 Córdoba, Spain
2 Instituto de Investigación en Ciencias de la Alimentación (CSIC-UAM). Nicolás Cabrera, 9, 28049 Madrid, Spain

Abstract

A meta-regression of the effects of the amount of plant lipids consumed by dairy goats on the contents of some milk fat fatty acids (FA) was carried out. Fourteen peer-reviewed published papers reporting 17 experiments were used in the study. Those experiments compared control diets without added fat with diets that included plant lipids rich in unsaturated FA, summing up to 64 treatments. The results showed that increasing daily intake of plant lipids linearly reduced the contents of all medium chain saturated FA in milk fat. Moreover, it was observed that the longer the chain of the milk saturated FA, the greater the negative effect of the plant lipid intake on their contents. On the other hand, the contents of stearic acid and the sum of oleic, linoleic and α-linolenic acids in milk fat linearly increased as daily plant lipid intake rose. The results obtained corroborate previous reports on the effects of feeding dairy goats with increasing amounts of unsaturated plant lipids on milk FA profile.

Additional key words: meta-regression; dairy; fat; diet; ruminants.

Abbreviations used: DM (dry matter); FA (fatty acid); RMSE (root mean square error); UFA (unsaturated fatty acid).

Citation: Martínez Marín, A. L.; Núñez Sánchez, N.; Garzón Sigler, A. I.; Peña Blanco, F.; de la Fuente, M. A. (2015). Short communication: Relationships between the daily intake of unsaturated plant lipids and the contents of major milk fatty acids in dairy goats. Spanish Journal of Agricultural Research, Volume 13, Issue 2, e06SC03, 5 pages. http://dx.doi.org/10.5424/sjar/2015132-6509.

Received: 10 Jul 2014. Accepted: 16 Mar 2015

Some papers have reported the effect of adding different amounts of unsaturated plant lipids to dairy goat diets on their milk fat fatty acid (FA) contents (Mir et al., 1999; Schmidely et al., 2005; Nudda et al., 2006; Martínez Marin et al., 2012), but few of them have quantitatively related the observed responses to the daily lipid intake. Martínez Marín et al. (2012) observed significant linear effects on the contents of the major FA in milk fat when fed dairy goats with increasing amounts (0, 32, 48 or 66 g/d) of three differently unsaturated plant lipids; however, they did not derive quantitative relationships. On the other hand, the amounts of unsaturated plant lipids supplied to dairy goats in the literature cover a much wider range, i.e. 15 to 207 g/d (Nudda et al., 2006; Ollier et al., 2009). Therefore, a meta-analysis of published research data could help to relate the observed responses of milk fat FA contents to the amount of unsaturated plant lipids fed to dairy goats, as an alternative to the scarcity of papers on the matter. Meta-analysis is a statistical tool used to objectively review prior published results even if the researches differ in their methodology. Meta-regression is a type of meta-analysis that allows integrating quantitative findings to formulate models that best explains the observations (St-Pierre, 2001; Sauvant et al., 2008).

The aim of this paper was to put published research data on feeding dairy goats with unsaturated plant lipids to meta-regression, in order to quantify the relationship between the amount of lipids consumed and the contents of some FA in milk fat.

Research papers on the inclusion of unsaturated plant lipids in dairy goat diets were searched in bibliographic databases (ISI Web of Science, Scopus, PubMed, Google Scholar) using the following keywords: fats, goats, milk fat and fatty acids. Studies were selected if...
they compared a control diet with no added fat with one or more treatments that included one unprotected fat source rich in unsaturated FA. A total of 14 peer-reviewed research papers with 17 experiments and 64 treatments fulfilled the requirements (Suppl. Table S1 [pdf online]).

Treatments were coded either as NOLIP when the diet contained no added fat, or as LIP if the diet included extra fat, which could be supplied as oils and oilseeds, either rich in oleic or linoleic or α-linolenic acids. Percentages of added fat in each treatment, dry matter (DM) intake and several other factors related to the fat source studied, the animals and the diets used, and the experimental design were recorded and stored in a Microsoft Excel spreadsheet. Variables studied were the milk fat contents of short and medium chain saturated FA (from C4:0 to C16:0), stearic acid, and the sum of oleic, linoleic and α-linolenic acids (C18 UFA). Milk fat contents of neither oleic, linoleic and α-linolenic acids, accounted for separately from each other, nor the fat contents of short and medium chain saturated FA, stearic acid, and the sum of oleic, linoleic and α-linolenic acids in the plant lipids supplied (Martínez Marín et al., 2013b) and this factor could not be considered in the present study, due to the low number of published experiments.

MIXED procedure of SAS 9.1 (SAS Institute Inc., Cary, NC, USA) was used in the statistical analyses. Firstly, we compared NOLIP and LIP diets to rule out diet composition as a source of interference. The linear mixed model used was (Sauvant et al., 2008):

$$Y_{ijk} = \mu + S_i + T_j + ST_{ij} + e_{ijk},$$

where $Y_{ijk}$ is the dependent variable, $\mu$ is the overall mean, $S_i$ is the random effect of the $i^{th}$ experiment, $T_j$ is the fixed effect of the $j^{th}$ level of treatment (NOLIP and LIP), $ST_{ij}$ is the random interaction between the $i^{th}$ experiment and the $j^{th}$ level of treatment, and $e_{ijk}$ is the residual error. Next, a regression analysis (meta-regression) was conducted to study the relationship between the daily intake of supplementary fat and the FA contents in milk fat. The graphical analysis of data showed that the relationships between the FA contents in milk fat and the daily fat intake, if it existed, were linear, so only this type of relationship was investigated. For each equation obtained, the root mean square error (RMSE) and the coefficient of determination ($R^2$) were calculated. The linear mixed model used was (Sauvant et al., 2008):

$$Y_j = B_0 + S_i + B_jX_j + b_jX_j + e_{ij},$$

where $Y_j$ is the dependent variable, $B_0$ is the overall intercept, $S_i$ is the random effect of the $i^{th}$ study, $B_j$ is the overall regression coefficient of $Y$ on $X$, $X_j$ is the value of the continuous predictor variable (fat intake, fixed effect), $b_j$ is the random effect of the experiment on the regression coefficient of $Y$ on $X$, and $e_{ij}$ is the residual error. The data were weighed with the square root of the number of animals used in each treatment using the WEIGHT statement.

Twenty two out of 64 treatments corresponded to control diets without added fat, and 16, 16 and 10 corresponded to diets enriched in oleic acid, linoleic acid and α-linolenic acid, respectively. Supplementary lipids were fed as free oils (30 treatments) and oilseeds (12 treatments). Supplementary fat was (mean ± SD and range) 3.43 ± 1.420 [0.80-6.50] % dry matter, or 74.3 ± 40.40 [15.0-207.0] g/d, or 1.30 ± 0.630 [0.25-3.45] g/kg body weight. Experiments carried out with dairy goats to investigate the effects of unsaturated plant lipid supplementation on milk fat FA contents are limited in number, compared with those carried out with dairy cows. Because of the few number of available treatments in our meta-regression, it was not possible to study the effects of the type of plant lipids supplied (i.e. rich in oleic, linoleic or α-linolenic acids) or the fat sources used (oils vs. oilseeds). On the other hand, the interference due to diet composition, if any, should be minimal because there were no differences between NOLIP and LIP treatments, except for the extra fat added (Table 1).

In the present study, the contents of C4:0 and C6:0 in goat milk fat were not reduced with increasing daily fat intake. Furthermore, the negative effects of plant lipid intake on the FA synthesized de novo in the mammary gland were clear from C8:0 upwards. Indeed, the slope of the C10:0 equation was ten-fold higher than that of the C8:0 equation (Table 2). Although there are not published meta-regressions that allow a direct comparison, in several experiments carried out with dairy cows (Mustafa et al., 2003; Collomb et al., 2004; Akraim et al., 2007; Liu et al., 2008; Chilliard et al., 2009) and ewes (Mele et al., 2006; Zhang et al., 2006; Gómez-Cortés et al., 2008; Hervás et al., 2008, Bodas et al., 2010) fed similar amounts of unsaturated plant

### Table 1. Diet composition (percent on dry matter basis)

| Treatments | NOLIP | SEM | LIP | SEM | $P$ |
|------------|-------|-----|-----|-----|-----|
| Forage     | 42.91 | 2.126 | 44.01 | 1.480 | 0.68 |
| Crude protein | 16.19 | 0.137 | 16.48 | 0.100 | 0.09 |
| Neutral detergent fibre | 35.51 | 0.660 | 34.92 | 0.460 | 0.47 |
| Added fat  | –     | 3.43 | 0.219 | 3.43 | 0.219 |

NOLIP: diets with no added plant lipids; LIP: diets added with plant lipids rich in unsaturated fatty acids. SEM: standard error of the mean.
unsaturated fat intake and goat milk fatty acid profile

The average contents of medium chain saturated FA in milk fat were lowered more than 60% in the treatments with the highest fat intakes compared with no added lipid ones. Since medium chain saturated FA consumption is considered a risk of cardiovascular disease in people (Ulbricht & Southgate, 1991), the marked reduction of their content in the milk fat is positive from the point of view of human health.

Linear increases in the milk fat contents of both C18:0 and C18 UFA due to the increments in daily fat intake (Table 2, Fig. 1), can be explained by the higher dietary supply of long chain unsaturated FA. Any unsaturated FA of 18 atoms of carbon present in the rumen can be a source of stearic acid through biohydrogenation, thus increasing the availability of preformed stearic acid to the udder. Again, the increase of C18 UFA contents in milk fat was likely due to dietary lipids (between 1 and 3 g/kg body weight), the negative effects on de novo synthesized FA are clearly observed from C6:0 and C8:0. Also, Glasser et al. (2008) found out that lipid supplementation to dairy cows, resulting in an increase of FA with 18 atoms of carbon in duodenal flow, linearly decreased the yield of C4:0 to C16:0 in milk. These differences suggest that dietary unsaturated plant lipids could have dissimilar effects on rumen digestion or lipid mammary metabolism in dairy cows, goats and ewes, as pointed out by Chilliard et al. (2003) and Sanz-Sampelayo et al. (2007).

On the other hand, Hansen et al. (1984), working in vitro with goat mammary cells, proposed that a greatly enhanced supply of long chain FA could inhibit the incorporation of de novo synthesized FA into triacylglycerol, owing to competition for the diacylglycerol acyltransferase, which in turn would decrease de novo synthesis of FA. Bernard et al. (2009a) found that the increase in long chain FA captured by the mammary gland reduced the activity of the enzymes involved in FA synthesis in the udder. Furthermore, it has been previously observed, working in vitro with mammary cells, that lowering the ratio acetyl-CoA carboxylase to FA synthetase modifies the pattern of synthesized FA towards those of shorter chain (Bau- man & Davies, 1974). In agreement with that, the values of the slopes were increasingly higher in the regression equations obtained in the present work for C8:0, C10:0, C12:0, C14:0 and C16:0, to such extent that the value of the slope in the C16:0 equation was four-fold higher than the average of the slopes in the C10:0, C12:0 and C14:0 equations (Table 2, Fig. 1), i.e. the longer the chain of the saturated milk FA, the greater the negative effect of the intake of high quantities of unsaturated plant lipids. On the other hand, from the equations in Table 2, it was calculated that the average contents of medium chain saturated FA in milk fat were lowered more than 60% in the treatments with the highest fat intakes compared with no added lipid ones. Since medium chain saturated FA consumption is considered a risk of cardiovascular disease in people (Ulbricht & Southgate, 1991), the marked reduction of their content in the milk fat is positive from the point of view of human health.

Linear increases in the milk fat contents of both C18:0 and C18 UFA due to the increments in daily fat intake (Table 2, Fig. 1), can be explained by the higher dietary supply of long chain unsaturated FA. Any unsaturated FA of 18 atoms of carbon present in the rumen can be a source of stearic acid through biohydrogenation, thus increasing the availability of preformed stearic acid to the udder. Again, the increase of C18 UFA contents in milk fat was likely due to dietary

| Fatty acids | Parameter estimates | RMSE | R² |
|------------|---------------------|------|----|
|            | Intercept | SEM  | p   | Slope | SEM  | p   |      |
| C4:0       | 2.12      | 0.217| <0.001| 0.0015 | 0.00033| <0.001| 0.60 | 0.80 |
| C6:0       | 2.42      | 0.174| <0.001| 0.00005| 0.00044| 0.250 | 0.26 | 0.95 |
| C8:0       | 2.85      | 0.201| <0.001| 0.00021| 0.00070| 0.006 | 0.41 | 0.91 |
| C10:0      | 10.14     | 0.324| <0.001| 0.00205| 0.00253| <0.001| 1.05 | 0.87 |
| C12:0      | 5.08      | 0.183| <0.001| 0.00185| 0.00182| <0.001| 0.66 | 0.86 |
| C14:0      | 11.24     | 0.253| <0.001| 0.00336| 0.00330| <0.001| 0.59 | 0.96 |
| C16:0      | 30.56     | 0.782| <0.001| 0.01053| 0.01196| <0.001| 2.75 | 0.90 |
| C18:0      | 6.93      | 0.297| <0.001| 0.00591| 0.00922| <0.001| 1.78 | 0.89 |
| C18 UFA    | 18.76     | 0.909| <0.001| 0.00567| 0.01020| <0.001| 2.60 | 0.76 |

C18 UFA: sum of oleic, linoleic and α-linolenic acids. SEM: standard error of the mean. RMSE: root mean square error. R²: coefficient of determination.
unsaturated FA that escaped unchanged from the rumen, although in the case of oleic acid, it could be also due to Δ-9 desaturation of stearic acid in the mammary gland (Chilliard & Ferlay, 2004). From the equations in Table 2, it can be calculated that the increments of C18:0 and C18 UFA contents in milk fat as daily fat intake increased did not account for the lessening of medium chain saturated FA contents, which indicates that the contents of other FA, probably cis and trans mono- and polyunsaturated FA from rumen metabolism of dietary FA, were also increased in milk fat in response to plant lipid supplementation (Chilliard et al., 2007).

In conclusion, the meta-regression carried out in this work allowed to obtain quantitative relationships between the amount of unsaturated plant lipids consumed by dairy goats and the changes in the contents of some milk FA. Our results corroborate that dietary unsaturated plant lipids have a more pronounced negative effect on medium chain saturated FA contents as their chain length increases. The milk fat contents of C18:0 and C18 UFA were increased as daily fat intake rose, but the increments observed did not account for the lessening of medium chain saturated FA contents.

References

Akraim F, Nicot MC, Juaneda P, Enjalbert F, 2007. Conjugated linolenic acid CLnA, conjugated linoleic acid CLA and other biohydrogenation intermediates in plasma and milk fat of cows fed raw or extruded linseed. Animal 1: 835-843. http://dx.doi.org/10.1017/S175173110700002X.

Andrade PVD, Schmidely PH, 2006a. Influence of percentage of concentrate in combination with rolled canola seeds on performance, rumen fermentation and milk fatty composition in dairy goats. Livest Sci 104: 77-90. http://dx.doi.org/10.1016/j.livsci.2006.03.010.

Andrade PVD, Schmidely PH, 2006b. Effect of duodenal infusion of trans10,cis12-CLA on milk performance and milk fatty acid profile in dairy goats fed high or low concentrate diet in combination with rolled canola seed. Reprod Nutr Develop 46: 31-48. http://dx.doi.org/10.1051/rnd:2005062.

Bauman DE, Davis CL, 1974 Biosynthesis of milk fat. In: Lactation: a comprehensive treatise. Vol. 2. (Larson BL, Smith VR, eds.). Academic Press, NY (USA), pp: 31-75.

Bernard L, Rouel J, Leroux C, Ferlay A, Faulconnier Y, Legrand P, Chilliard Y, 2005. Mammary lipid metabolism and milk fatty acid secretion in alpine goats fed vegetable lipids. J Dairy Sci 88: 1478-1489. http://dx.doi.org/10.3168/jds.S0022-0302(05)72816-2.

Bernard L, Bonnet M, Leroux C, Shingfield KJ, Chilliard Y, 2009a. Effect of sunflower-seed oil and linseed oil on tissue lipid metabolism, gene expression, and milk fatty acid secretion in Alpine goats fed maize silage-based diets. J Dairy Sci 92: 6083-6094. http://dx.doi.org/10.3168/jds.2009-2048.

Bernard L, Shingfield KJ, Rouel J, Ferlay A, Chilliard Y, 2009b. Effect of plant oils in the diet on performance and milk fatty acid composition in goats fed diets based on grass hay or maize silage. Brit J Nutr 101: 213-224. http://dx.doi.org/10.1017/S0007114508006533.

Bodas R, Manso T, Mantecón AR, Juárez M, de la Fuente MA, Gómez-Cortés P, 2010. Comparison of the fatty acid profiles in cheeses from ewes fed diets supplemented with different plant oils. J Agric Food Chem 58: 10493-10502. http://dx.doi.org/10.1021/jf101760u.

Bouattour MA, Casals R, Albanell E, Such X, Caja G, 2008. Feeding soybean oil to dairy goats increases conjugated linoleic acid in milk. J Dairy Sci 91: 2399-2407. http://dx.doi.org/10.3168/jds.2007-0753.

Chilliard Y, Ferlay A, 2004. Dietary lipids and forages in interactions on cow and goat milk fatty acid composition and sensory properties. Reprod Nutr Dev 44: 467-492. http://dx.doi.org/10.1051/rnd:2004052.

Chilliard Y, Ferlay A, Rouel J, Lamberet G, 2003. A review of nutritional and physiological factors affecting goat milk lipid synthesis and lipolysis. J Dairy Sci 86: 1751-1770. http://dx.doi.org/10.3168/jds.S0022-0302(03)73761-8.

Chilliard Y, Glasser F, Ferlay A, Bernard L, Rouel J, Doreau M, 2007. Diet, rumen biohydrogenation and nutritional quality of cow and goat milk fat. Eur J Lipid Sci Technol 109: 828-855. http://dx.doi.org/10.1002/ejlt.200700080.

Chilliard Y, Martin C, Rouel J, Doreau M, 2009. Milk fatty acids in dairy cows fed whole crude linseed, extruded linseed, or linseed oil, and their relationship with methane output. J Dairy Sci 92: 5199-5211. http://dx.doi.org/10.3168/jds.2009-2375.

Colomb M, Sollberger H, Bütkofer U, Sieber R, Stoll W, Schaeren W, 2004. Impact of a basal diet of hay and fodder beet supplemented with rapeseed, linseed and sunflowerseed on the fatty acid composition of milk fat. Int Dairy J 14: 549-559. http://dx.doi.org/10.1016/j.idairyj.2003.11.004.

Glasser F, Ferlay A, Doreau M, Schmidely P, Sauviant D, Chilliard Y, 2008. Long-chain fatty acid metabolism in dairy cows: A meta-analysis of milk fatty acid yield in relation to duodenal flows and de novo synthesis. J Dairy Sci 91: 2771-2785. http://dx.doi.org/10.3168/jds.2007-0383.

Gómez-Cortés P, Frutos P, Mantecón AR, Juárez M, de la Fuente MA, Hervás G, 2008. Milk production, conjugated linoleic acid content, and in vitro ruminal fermentation in response to high levels of soybean oil in dairy ewe diet. J Dairy Sci 91: 1560-1569. http://dx.doi.org/10.3168/jds.2007-0722.

Hansen HO, Grunnet I, Knudsen J, 1984. Triacylglycerol synthesis in goat mammary gland. Biochem J 220: 521-527.

Hervás G, Luna P, Mantecón AR, Castañeres N, de la Fuente MA, Juárez M, Frutos P, 2008. Effect of diet supplementation with sunflower oil in milk production, fatty acid profile and ruminal fermentation in lactating dairy ewes. J Dairy Res 75: 399-405. http://dx.doi.org/10.1017/S0022029908003506.
Liu ZL, Yang DP, Chen P, Lin SB, Jiang XY, Zhao WS, Li JM, Dong WX. 2008. Effect of dietary sources of roasted oilseeds on blood parameters and milk fatty acid composition. Czech J Anim Sci 53: 219-226.

Maia FJ, Branco AF, Mourou GF, Coneglian SM, Dos Santos GT, Minella TF, Guimarães KC. 2006. Inclusão de fontes de óleo na dieta de cabras em lactação: produção, composição e perfil dos ácidos grasos do leite. R Bras Zootec 35: 1504-1513. http://dx.doi.org/10.1590/S1516-35982006000500033.

Martínez Marín AL, Gómez-Cortés P, Gómez Castro G, Juárez M, Pérez Alba L, Pérez Hernández M, de la Fuente MA. 2011. Animal performance and milk fatty acid profile of dairy goats fed diets with different unsaturated plant oils. J Dairy Sci 94: 5359-5368. http://dx.doi.org/10.3168/jds.2011-4569.

Martínez Marín AL, Gómez-Cortés P, Gómez Castro G, Juárez M, Pérez Alba L, Pérez Hernández M, de la Fuente MA. 2012. Effects of feeding increasing dietary levels of high oleic or regular sunflower or linseed oil on fatty acid profile of goat milk. J Dairy Sci 95: 1942-1955. http://dx.doi.org/10.3168/jds.2011-4303.

Martínez Marín AL, Pérez Hernández M, Pérez Alba LM, Carrion Pardo D, Gómez Castro AG, Garzón Sigler AI, 2013b. Effect of oils and seeds in ruminant diets on milk fat fatty acid profile: review. Rev Mex Cienc Pecu 4: 319-338.

Mele M, Bucchi A, Petachi F, Serra A, Banni S, Antongiovanni M, Secchiaroli P, 2006. Effect of forage/concentrate ratio and soybean oil supplementation on milk yield, and composition from Sarda ewes. Anim Res 55: 273-285. http://dx.doi.org/10.1051/anime:2006019.

Mele M, Serra A, Bucchi A, Conte G, Pollicarido A, Secchiaroli P, 2008. Effect of soybean oil supplementation on milk fatty acid composition from Saanen goats fed diets with different forage: concentrate ratios. Ital J Anim Sci 7: 297-311.

Mir Z, Goonewardene LA, Okine E, Jaeger S, Scheer HD, 1999. Effect of feeding canola oil on constituents, conjugated linoleic acid (CLA) and long chain fatty acids in goats milk. Small Rumin Res 33: 137-143. http://dx.doi.org/10.1016/S0921-4488(99)00016-4.

Mustafa AF, Chouinard PY, Christensen DA. 2003. Effects of feeding micronised flaxseed on yield and composition of milk from Holstein cows. J Sci Food Agric 83: 920-926. http://dx.doi.org/10.1002/jsfa.1430.

Nudda A, Battacone G, Usai MG, Fancellu S, Pulina G, 2006. Supplementation with extruded linseed cake affects concentrations of conjugated linoleic acid and vaccenic acid in goat milk. J Dairy Sci 89: 277-282. http://dx.doi.org/10.3168/jds.S0022-0302(06)72092-6.

Ollier S, Leroux C, De la Foye A, Bernard L, Rouel J, Chilliard Y, 2009. Whole intact rapeseeds or sunflower oil in high-forage or high-concentrate diets affects milk yield, milk composition, and mammary gene expression profile in goats. J Dairy Sci 92: 5544-5560. http://dx.doi.org/10.3168/jds.2009-2022.

Sanz-Sampelayo MR, Chilliard Y, Schmidely P, Boza J, 2007. Influence of type of diet on the fat constituents of goat and sheep milk. Small Rumin Res 68: 42-63. http://dx.doi.org/10.1016/j.smallrumres.2006.09.017.

Schmidely P, Morand-Fehr P, Sauvant D, St-Pierre NR, 2008. Meta-analyses of experimental data in animal nutrition. Animal 2: 1203-1214. http://dx.doi.org/10.1017/S1751731108002280.

Ulbricht TLV, Southgate DAT. 1991. Coronary heart disease: seven dietary factors. Lancet 338: 985-992. http://dx.doi.org/10.1016/0140-6736(91)91846-M.

Zhang RH, Mustafa AF, Zhao X, 2006. Effects of feeding oilseeds rich in linoleic and linolenic fatty acids to lactating ewes on cheese yield and on fatty acid composition of milk and cheese. Anim Feed Sci Technol 127: 220-233. http://dx.doi.org/10.1016/j.anifeedsci.2005.09.001.