

## META-ANALYSIS OF DIETARY SUNFLOWER OIL SUPPLEMENTATION FOR DAIRY GOATS: PERFORMANCE, MILK COMPOSITION, AND MILK FATTY ACID PROFILE

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### ABSTRACT

Milk and dairy products are important foods that contribute to daily nutrient requirements and improve consumers' health. The objectives of this study were to critically review and quantify, using meta-analysis and meta-regression, the effects of supplementation with sunflower oil (SFO) on dry matter intake (DMI), milk yield (MY), components and fatty acids (FAs) profile in dairy goats. A total of 154 papers were reviewed. Nine articles (10 experiments) met the eligibility criteria and were used in the analysis. The effect size for all parameters was calculated as raw mean difference (RMD) and standardized mean difference (SMD). Heterogeneity was determined using  $I^2$  statistics, while meta-regression was used to examine factors influencing heterogeneity. Responses to SFO supplementation were heterogeneous for all variables studied. However, SFO decreased DMI (RMD = -0.050 kg / d;  $p = 0.007$ ) and increased milk fat percent (MFP;  $p < 0.001$ ) and milk lactose percent (MLP;  $p < 0.001$ ), but the effect size was not significant for MY. The inclusion of SFO in dairy goats rations enhanced C18:1 cis-9 (RMD = +2.22 g / 100 g FA;  $p < 0.001$ ), C18:1 trans-11 (RMD = +2.77 g / 100 g FA;  $p < 0.001$ ), C18:2 cis-9 trans-11 (RMD = +0.261 g / 100 g FA;  $p < 0.001$ ), C18:3 n-3 (RMD = +0.078 g / 100 g FA;  $p = 0.002$ ) and MUFA (RMD = +7.16 g / 100 g FA;  $p = 0.002$ ) and PUFA (RMD = +1.49 g / 100 g FA;  $p < 0.0001$ ), and diminished SFA (RMD = -7.53 g / 100 g FA;  $p = 0.008$ ). Overall, the meta-analysis data indicated that dietary SFO supplementation in dairy goats has a positive effect on desirable milk components for human consumption. However, a cost-effectiveness analysis is needed to provide accurate recommendations to farmers and the dairy goat industry.

**Keywords:** Fatty acid profile, goat feeding, sunflower oil, vegetable oil.

## INTRODUCTION

Currently, total goat milk production is estimated at 18.7 million tons (1.9% of the global milk production) and projected to increase by 53% by 2030, reaching 28.6 million tons (Plata-Pérez et al., 2022; Saran Netto et al., 2022). Goat milk and dairy products are considered great sources of high-quality nutrients, especially proteins and fats (Vargas-Bello-Pérez et al., 2021; Salles et al., 2019). Nowadays, there is a trend for safe, natural, and healthy dairy foods, derived from an increasing consumer awareness about the connection between diet and health, and at times resulting in willingness to pay a premium price for such food products (Vargas-Bello-Pérez et al., 2021; Vargas-Bello-Pérez et al., 2022). In this sense, research on milk fat is still oriented to the improvement of its nutritional value, with particular attention paid to: i) reducing saturated fatty acids (SFA, commonly known as a group of fats with health-related issues); ii) increasing the desirable fatty acids (FA), such as branched-chain fatty acids (BCFA) and ruminant trans-fatty acids (TFA), especially vaccenic, rumenic, and  $\alpha$ -linolenic acids; and iii) enhancing the omega-6 (n6) to omega-3 (n3) ratio for optimizing human health (e.g. anticarcinogenic, antiatherogenic, and immune modulator effects) (Plata-Pérez et al., 2022; Vargas-Bello-Pérez et al., 2022; Chilliard et al., 2014; Bionaz et al., 2020).

In terms of costs, time, and responses, the most effective strategy to modulate milk FA toward a healthier profile is through dietary changes (Vargas-Bello-Pérez et al., 2020). In this regard, supplementation of goat feed with high linoleic vegetable oil, such as sunflower oil (SFO), has been addressed as a good nutritional strategy for enhancing the beneficial FA (such as  $\alpha$ -linoleic acid and n-3 PUFA) in milk and dairy products (Saran Netto et al., 2022; Razzaghi et al., 2015). However, literature regarding SFO supplementation on goat performance, milk yield, and FA profile has been inconsistent, which can be related to the differences in the experimental conditions (e.g., supplementation level, basal diets, and processing of oilseeds as well as animal variables) (Vargas-Bello-Pérez et al., 2021).

Meta-analysis offers a standardized statistical framework for estimating the mean effect size of an intervention or exposure factor from individual experiments that are too small or underpowered to demonstrate a statistically significant association. It also allows for the examination of between-study variability or heterogeneity of treatment effects (Sutton, 2008; Lean et al., 2009). The present study aimed to conduct an analytic

review to quantitatively summarize the global effect of SFO supplementing on dry matter intake (DMI), milk yield (MY), milk composition, and FA profile in dairy goats through a meta-analysis on a database built from independent studies.

## MATERIALS AND METHODS

### Search of the published literature

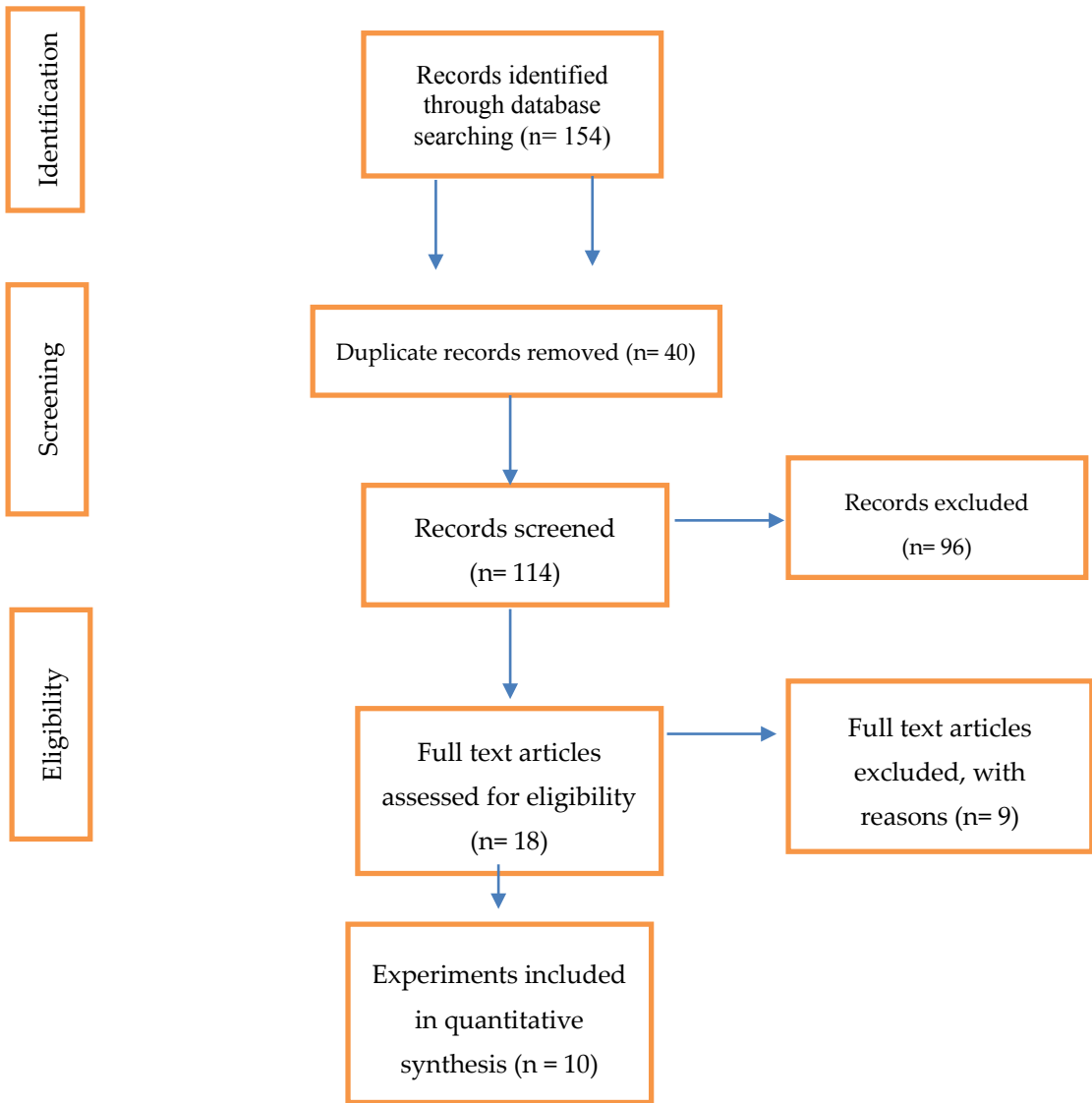
To investigate the impact of sunflower oil (SFO) on the productive performance and milk FA profile of dairy goats, an extensive literature search was performed in English publications from 2000 to 2022. The literature search included two search engines, the ISI Web of Knowledge (<http://wokinfo.com>) and Google Scholar (<http://scholar.google.com>). The keywords provided to the field experts included sunflower, fatty acids, milk, and dairy goat. For Google Scholar, several thousand hits were collected, and results were sorted in order of relevance. The screening of papers stopped after at least 50 records after the last relevant record was identified. No restrictions were imposed on the selection of journals based on impact factor or quartile ranking.

### Inclusion and exclusion criteria

Fig.1 shows a PRISMA flow diagram (Moher et al., 2009) of the data collected for the meta-analysis. Out of 154 published articles, duplicate articles (n = 40), review articles (n = 12), articles related to the effect of dietary fat or oilseed sources rich in UFA in other livestock species, and articles related to other sunflower products (such as seed, meal, and cake; n = 84) were excluded. Of the remaining 18 articles, 9 papers were excluded because they focused in a grazing system (n = 3), or sunflower oil was mixed with other oils or additives (n = 3), lacked a control group (n = 1), failed to report diet and chemical compositions (n = 2). Therefore, the 9 articles (including 10 experiments) identified for this meta-analysis met the main criterion, i.e., the effects of SFO on milk FA profile and productive performance of dairy goats. Two reviewers meticulously assessed all accessible articles based on predetermined inclusion and exclusion criteria. Any discrepancies or disagreements that arose during the screening process were diligently resolved through consultation with a third reviewer at each stage. A list of the experiments included in the meta-analysis is provided in Table 1.

### Data extraction

Data extracted from each study, included authors' names and year of publication, DMI (kg/day), MY (kg/day), milk fat percentage (MFP), milk protein percentage (MPP), milk lactose



**Fig. 1.** The PRISMA flow diagram of the systematic review from initial search and screening to final selection of publications to be included in the meta-analysis.

percentage (MLP), and milk FA profile (g/100 g). Data including country, breed of goat, amount of SFO, forage basis, body weight (kg), duration of the experiment (day), number of animals per treatment and control groups, and standard error were also extracted. The standard deviation (SD) was recorded as the measure of variance. If SD was not reported, it was calculated by multiplying the reported SE of means by the square root of the sample size.

A limitation observed in this meta-analysis was the absence of reported data concerning all nutrient compositions of the diet and the FA

profiles of the experimental diets. However, available information, including forage intake, neutral detergent fiber (NDF) intake, ether extract (EE) intake, and crude protein (CP) intake, was considered as a variable to perform meta-regression for productive performance and milk FA profile. The collected data were meticulously transferred to Excel spreadsheets (version 2019, Microsoft Corp., Redmond, WA) and thoroughly reviewed by two animal science researchers to ensure accuracy of transcription from the manuscripts into the spreadsheets before conducting statistical analyses.

## Statistical analysis

### Descriptive statistics

Descriptive statistics for the chemical composition of the diets (NDF, EE, and CP) and productive parameters (MY, MFP, MPP, and MLP) were performed using Excel spreadsheets (version 2019, Microsoft Corp., Redmond, WA).

### Effect size and Forest plots

Statistical analysis was performed using Comprehensive Meta-Analysis (CMA) software version 4 (Biostat, USA) to calculate the effect size for MY, MPP, MFP, MLP, and milk FA profile in terms of raw mean difference (RMD) at a 95% confidence interval. The RMD is the difference between the treatment and control groups. Calculating RMD allows the expression of the effect size with the same unit as the measurement. In addition to calculating the raw mean difference (RMD), the standardized mean difference (SMD) was computed for each outcome, accompanied by a 95% confidence interval. The SMD indicates the mean difference between treatment and control groups, standardized based on the SD of treatment and control groups (Borenstein et al., 2011). The SMD is calculated using the following formula:

$$\text{SMD} = \frac{\bar{x}_e - \bar{x}_c}{S_p}$$

where  $\bar{x}_e$  is the experimental group mean,  $\bar{x}_c$  is the control group mean, and  $S_p$  is the pooled SD (Lean et al., 2009).

A random-effects model was adopted for the meta-analysis. The model has an underlying assumption that the distribution of effects exists, resulting in heterogeneity among study results (Borenstein et al., 2011). The significance of effect size estimates (RMD and SMD) was declared at  $p \leq 0.05$ . Forest plots were constructed to evaluate the effects of sunflower oil on MY and MFP. The effect size for forest plots was the RMD at a 95% confidence interval using the random-effects model.

### Heterogeneity

Statistical heterogeneity refers to the true effects in each study not being identical (Sutton et al., 2008). The existence of heterogeneity reflects underlying differences in the clinical diversity of the herds, differences in study design, and statistical variation (Lean et al., 2009). Identifying the presence and sources of the heterogeneity improves the understanding of the responses to the interventions used. The  $I^2$  statistic was employed to quantify the heterogeneity of results among the trials (Lean et al., 2009).

$$I^2(\%) = \frac{Q - (k - 1)}{Q} \times 100$$

where  $Q$  is the  $\chi^2$  heterogeneity statistic and  $k$  is the number of trials. An  $I^2$  value between 0 and 40% might not be important; 30 to 60% may represent moderate heterogeneity; 50 to 90% might represent substantial heterogeneity; and 75 to 100% might represent considerable heterogeneity (Higgins et al., 2019).

### Meta-regression

Meta-regression analyses were used to explore the source of heterogeneity of response, using the individual RMD for each study comparison as the outcome and the associated SE as the measure of variance. In this study, meta-regression analysis was used to evaluate heterogeneous sources for parameters with more than 10 comparisons. Meta-regression was estimated using the method of moments, commonly known as the DerSimonian and Laird method. This method of estimating the variance between studies is well-established (Borenstein et al., 2011).

In this study, forage intake, neutral detergent fiber intake, ether extract intake, and crude protein intake variable were used as a covariate for data related to DMI, MY, composition, and milk FA profile. As mentioned before, some studies did not report complete data including the chemical composition of diet and dietary FA profiles. This limitation did not allow the use of some parameters as covariates in meta-regression due to the scarcity of available data.

### Publication bias

Although a meta-analysis will yield a mathematically accurate synthesis of the studies included in the analysis, if these studies are a biased sample of all relevant studies, then the mean effect computed by the meta-analysis will reflect this bias. This issue is generally known as publication bias. Egger's linear regression asymmetry was used to examine the presence of publication bias ( $p < 0.10$ ).

## RESULTS

### Review of the data

Table 1 shows the selected papers and the data extracted for the meta-analysis. For the meta-analysis, 10 experiments were included. The studies were performed in Europe (8), specifically in Spain, France, and Denmark; Iran (1); and Jordan (1). In 4 experiments, the forage was based on alfalfa hay and the rest corresponded to orchard grass hay, natural grassland hay, and maize silage. Alpine goats were utilized in four experiments from the

**Table 1. Summary of papers used for the meta-analysis.**

References	No. of comparisons	Country	Breeds	Amount of unflower	Forage basis
Arco-Pérez et al., 2017	2	Spain	Murciano-Granadina	20 (g/kg DM)	Alfalfa hay
Bernard et al., 2005	1	France	Alpine	36 (g/kg DM)	Orchard grass hay
Bernard et al., 2008 (Exp.1)	1	France	Alpine	55 (g/kg DM)	Natural grassland hay
Bernard et al., 2008 (Exp.2)	1	France	Alpine	61 (g/kg DM)	Maize silage
Marin et al., 2011	1	Spain	Malagueña	48 (g/d)	Alfalfa hay
Marin et al., 2012	3	Spain	Malagueña	30, 48, 66 (g/d)	Alfalfa hay
Ollier et al., 2009	1	France	Alpine	44 (mg/g DM)	Alfalfa hay
Razzaghi et al., 2014	1	Iran	Saanen	37 (g/kg DM)	Corn silage and alfalfa hay
Titi et al., 2011	2	Jordan	Shami	30, 50 (g/kg DM)	
Vargas Bello Perez et al., 2022	1	Denmark	Danish Landrace	40 (g/kg DM)	Alfalfa +grass hay and Clover haylage

**Table 2. Descriptive statistics of data used in meta-analysis.**

Variable <sup>a</sup>	Mean		SD		Min		Max	
	S <sup>b</sup>	U <sup>b</sup>	S	U	S	U	S	U
Chemical composition of the diets								
NDF (g/kg)	348.50	352.21	85.76	82.13	255.00	268.00	553.00	553.00
EE (g/kg)	60.24	26.78	11.82	4.29	42.00	20.00	82.00	32.00
CP (g/kg)	165.71	166.43	12.60	17.06	135.00	140.00	185.00	196.00
Productive parameters								
MY (kg/d)	2.09	2.19	1.10	0.98	0.92	1.03	4.26	4.21
MFP (%)	4.71	4.21	1.30	1.14	2.26	2.02	6.79	6.25
MPP (%)	3.43	3.33	0.51	0.43	2.82	2.61	4.50	4.00
MLP (%)	4.60	4.55	0.16	0.12	4.34	4.41	4.85	4.76

<sup>a</sup>NDF, Neutral detergent fibre; EE, Ether extract; CP, Crude protein; MY, Milk yield; MFP, Milk fat percentage; MPP, Milk protein percentage; MLP, Milk lactose percentage; <sup>b</sup>S, Fat supplemented rations; U, Unsupplemented fat rations

database. Table 2 presents the descriptive statistical analysis of the chemical composition of diets, milk yield, and milk composition. In summary, the control group demonstrated the minimum and maximum levels of EE in the diet recorded at 20 (g/kg DM) and 32 (g/kg DM), respectively. The group receiving sunflower oil exhibited the lowest and highest quantities of EE in the diet, recording 42 (g/kg DM) and 82 (g/kg DM), respectively. The mean quantity of dietary NDF was 352.21 (g/kg DM) and 348.50 (g/kg DM) in the control group and SFO group, respectively (Table 2). Regarding the mean content of dietary CP, both groups recorded the same value of 166.43 (g/kg DM).

#### Dry matter intake and milk production

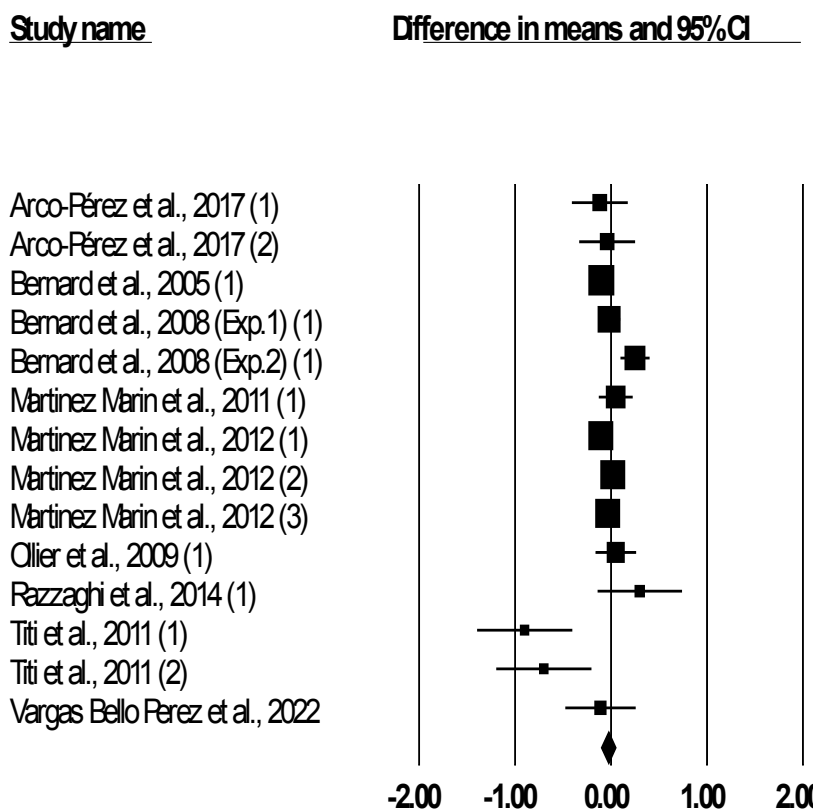
The supplementation of dietary SFO for dairy goats decreased the DMI ( $p=0.007$ ). However,

non-significant changes were observed for MY ( $p=0.356$ ; Table 3; Fig. 2). The heterogeneity ( $I^2$ ) for both DMI ( $p=0.014$ ) and MY ( $p<0.001$ ) was significant (Table 1). The Egger's test for DMI and MY showed that there is no publication bias (Table 3). Table 4 reports the meta-regression analysis for heterogeneous variables in the meta-analysis with differences between the SFO and control groups. DMI showed that NDF intake was a significant cause of heterogeneity between studies, suggesting that DMI will increase with increasing NDF intake. In terms of MY, the FOR intake, EE intake, and CP intake were significant causes of heterogeneity between studies. In this regard, a variability of response in MY was associated with SFO supplementation, which highlights a positive linear relationship between the EE intake and RMD of MY (Fig. 3).

**Table 3. Effect size, heterogeneity, and publication bias for the effect of dietary sunflower oil on milk yield and milk composition in dairy goat.**

Outcome	No. of comparison	RMD (95% CI)		Heterogeneity		SMD (95% CI)	Publication bias Egger
		Random effect	P-value	$I^2$	P-value		
DMI	13	-0.050 (-0.086, -0.014)	0.007	52.39	0.014	-0.387 (-0.733, -0.041)	0.214
MY	14	-0.035 (-0.108, 0.039)	0.356	71.33	<0.001	-0.252 (-0.630, 0.127)	0.996
MFP	14	0.456 (0.234, 0.678)	<0.001	77.67	<0.001	0.874 (0.378, 1.369)	0.863
MPP	14	0.075 (-0.009, 0.159)	0.079	47.06	0.026	0.264 (-0.008, 0.535)	0.308
MLP	11	0.095 (0.044, 0.145)	<0.001	54.23	0.016	0.651 (0.110, 1.191)	0.014

DMI, dry matter intake; MY, milk yield; MFP, milk fat percentage; MPP, milk protein percentage; MLP, milk lactose percentage; RMD, raw mean difference; CI, confidence interval; SMD, standardized mean difference.



**Fig. 2. Forest plot of the effect of dietary sunflower oil on milk yield in dairy goat based on difference in means. The diamond at the bottom indicates the mean effect size, calculated according to a random-effect model. The size of the squares illustrates the weight of each study relative to the mean effect size. Smaller squares represent less weight. The horizontal bars represent the 95% confidence intervals for the study.**

### Milk composition

MFP ( $p < 0.001$ ) and MLP ( $p < 0.001$ ) increased with the addition of SFO in dairy goats' diet, with a tendency of increase for MPP ( $p = 0.079$ ). The forest plot shown in supplementary Fig. 4 reveals a significant effect of SFO supplementation on MFP. Indeed, the heterogeneity ( $I^2$ ) result was

also significant for MFP, MLP, and MLP (Table 3). Egger's test showed no publication bias for MFP and MPP, while a publication bias was detected for MLP ( $p = 0.014$ ; Table 3). MFP was shown to be heterogeneous and was further assessed by meta-regression in Table 4; however, none of the variables were found to be significant.

**Table 4. Summary of meta-regression analysis for the effect of dietary sunflower oil on milk yield and milk composition in dairy goat.**

Outcomes	Covariate	Slope	p-value	Intercept	p-value
Dry matter intake	FORI	0.207	0.588	-0.052	0.008
	NDFI	0.001	<0.001	-0.034	0.008
	EEI	-0.0002	0.732	-0.034	0.424
	CPI	-0.0002	0.802	-0.046	0.024
Milk yield	DMI	-0.153	0.791	-0.041	0.351
	FORI	1.097	0.069	-0.055	0.127
	NDFI	-0.0007	0.420	-0.052	0.237
	EEI	0.001	0.047	-0.163	0.029
Milk Fat percent	CPI	0.003	0.029	-0.037	0.292
	FORI	0.565	0.762	0.436	0.001
	NDFI	0.001	0.533	0.475	<0.001
	EEI	-0.004	0.147	0.793	0.002
Milk protein percent	CPI	-0.002	0.526	0.477	<0.001
	FORI	-1.444	0.096	0.132	0.015
	NDFI	0.0003	0.714	0.088	0.098
	EEI	-0.001	0.258	0.172	0.070
Milk lactose percent	CPI	-0.002	0.063	0.101	0.018
	FORI	0.666	0.036	0.060	0.045
	NDFI	-0.0002	0.643	0.085	0.005
	EEI	0.001	0.001	-0.008	0.849
	CPI	0.002	<0.001	0.067	0.001

FORI, Difference of forage intake in treatment and control diets; NDFI, Difference of Neutral Detergent Fiber intake in treatment and control diet; EEI, Difference of ether extract intake in treatment and control diet; CPI, Difference of crude protein intake in treatment and control diets; DMI, Difference of dry matter intake in treatment and control diet

Additionally, a variability of response was observed, associated with the dietary CP intake (CPI) and RMD of MPP, as shown in Fig. 5, where different levels of RMD of the MPP outcome showed a negative linear response with CPI levels. However, MLP showed that EE intake and CPI were the variables affecting heterogeneity.

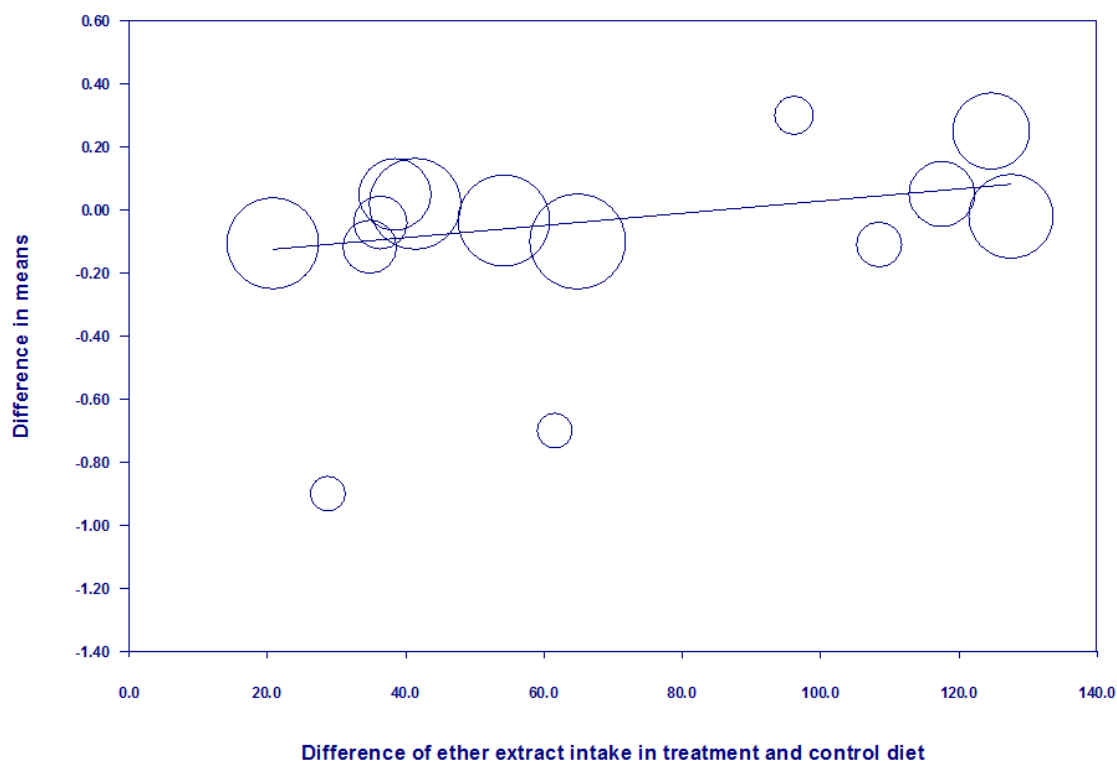
### Milk fatty acid profile

The results of the meta-analysis and meta-regression for the effect of supplementing SFO to the diets of dairy goats on the composition of milk FAs are reported in Tables 5 and 6, respectively. SFO decreased ( $p < 0.05$ ) the short-chain FAs (C8:0-C16:0), except for C4:0 and C6:0. The effect on odd-chain FAs was similar. Adding dietary SFO significantly increased C18:0 ( $p < 0.001$ ). Heterogeneity of short-chain and medium-chain (C4:0-C16:0), odd-chain (C15:0 and C17:0), and long-chain (C18:0) FAs was significant. C14:0, C15:0, and C16:0 showed that EE intake and CP intake were significant causes of heterogeneity between studies, whereas EE intake was a significant cause of heterogeneity for C10:0 and C17:0 (Table 6). These results showed that the

amount of C14:0, C15:0, and C16:0 decreased with increasing EE and CP intakes. In addition, the C10:0 and C17:0 decreased with increasing EE intake. Meta-regression for C 4:0, C 8:0, and C 18:0 revealed that none of the variables were causative for the heterogeneity observed (Table 6).

Dietary supplementation of SFO for dairy goats led to an increase in the concentration of C18:1 cis-9 ( $p < 0.001$ ), C18:1 trans-11 ( $p < 0.001$ ), C18:2 cis-9 trans-11 ( $p < 0.001$ ), and C18:3 n-3 ( $p = 0.002$ ) in milk, whereas C18:3 n-6 ( $p < 0.001$ ) decreased. Unsaturated fatty acids (UFA) showed significant heterogeneity. Meta-regression performed on milk C18:3 n-3 showed that EE intake was the only variable affecting the heterogeneity observed (Table 6).

By examining the Egger's test for publication bias, no bias in the publication for milk FAs was observed, except for C6:0, C18:1 trans-11, C18:2 cis-9 trans-11, and C18:3 n-3. The inclusion of SFO in the diet decreased the concentration of SFA ( $p=0.008$ ) and increased total MUFA ( $p=0.002$ ) and PUFA ( $p<0.001$ ) in milk. Egger's test found no publication bias for total SFA, MUFA, and PUFA ( $p<0.1$ ; Table 5). The heterogeneity of total



**Fig. 3.** Scatter plot of the meta-regression of difference of ether extract intake in treatment and control diet. The size of the circles represents the weight given to each individual study in the meta-analysis.

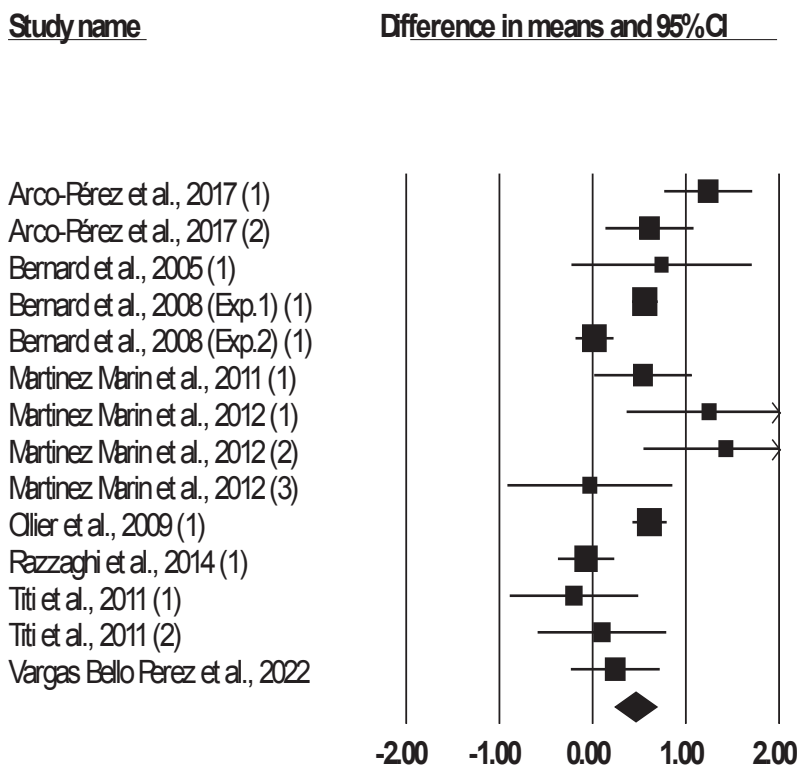
SFA, MUFA, and PUFA was also significant ( $Q$  and  $P$ ; Table 5).

## DISCUSSION

In this meta-analysis, less DMI was observed following dietary supplementation of SFO for dairy goats. This is in agreement with the literature as there are reports of decreases in DMI when fat is supplemented to the diets of lactating dairy cattle (Mahdavi et al., 2019; Rabiee et al., 2012). Similarly, a recent meta-analysis (Lashkari et al., 2024) of 25 published studies reported a concave effect of PUFA-rich vegetable oil supplementation on DMI of early lactating dairy cows. Reduced DMI for PUFA is also in line with other meta-analyses, which have demonstrated that increasing the crude fat content in cattle diets leads to decreased DMI (Rabiee et al., 2012; Glasser et al., 2008). The mechanism by which vegetable oils including SFO supplementation influence DM intake has not been fully elucidated in dairy cows (Saran Netto et al., 2022) or dairy goats (Nudda et al., 2020). However, Allen (2000) demonstrated that reduced DMI with fat supplementation may be attributed to

diminished rumen fermentation, lowered levels of cholecystokinin in the gut, and a slower rate of fatty acid metabolism in the liver. Although the typical recommendation for lipid inclusion in ruminant diets is up to 6–7% of dietary DM (NRC, 2001), it is well documented that higher levels can adversely affect rumen fermentation, leading to decreased DMI and ruminal fiber digestion (Muñoz et al., 2021). In the papers used for the current meta-analysis, the EE content of SFO-supplemented diets ranged from 4.2 to 8.2% of DM, which may have influenced the observed results concerning lower DMI. Furthermore, it has been suggested that the reduced DMI is associated with the effects of fat on ruminal fermentation, intestinal hormone release, and regulatory mechanisms controlling DMI as well as the restriction of ruminant's ability to oxidize FAs, and a consequent tendency to shift the site of nutrient digestion from rumen to the intestines (Pantoja et al., 1994). Consistently, a meta-analysis on the effects of fat additions to the diets of dairy cattle suggested that hypophagic effects of fat increase with the proportion of unsaturated fatty acids (UFAs) at the duodenal level (Rabiee et al., 2012). In addition, it has been described that



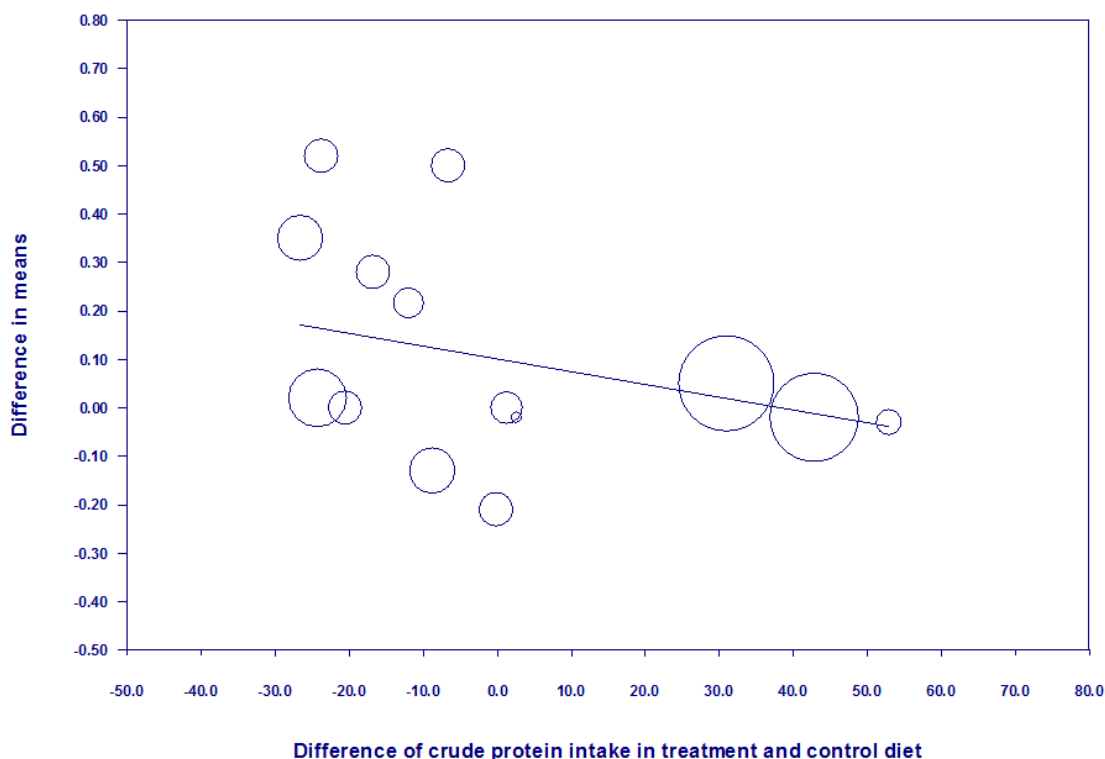


**Fig. 4.** Forest plot of the effect of dietary sunflower oil on milk fat percentage in dairy goat based on difference in means. The diamond at the bottom indicates the mean effect size, calculated according to a random-effect model. The size of the squares illustrates the weight of each study relative to the mean effect size. Smaller squares represent less weight. The horizontal bars represent the 95% confidence intervals for the study.

the DMI reduction might be related to plasma concentrations of certain FAs resulting from fat metabolism (Saran Netto et al., 2022). The FAs that seem to be involved in the DMI reduction mechanism are C18:2 n-6 and C18:1 n-9, which account for 63.42 and 23.64% of the fatty acid profile of SFO, respectively (Pantoja et al., 1994; Plata-Pérez et al., 2021). Furthermore, Bradford et al. (2008) indicated that an increase in plasma glucagon-like peptide-1 and cholecystokinin could be involved in the reduction of DMI when vegetable oil rich in PUFA is added to the diets of dairy cattle.

The inclusion of SFO did not affect MY. The absence of a noticeable effect of lower DMI in cows fed with PUFA on MY can be the result of increased energy density in the fat-supplemented diets to counterbalance the reduction in DMI (Lashkari et al., 2024). Consistent with the findings of our study, a recent meta-analysis conducted by Gallardo and Teixeira (2023) demonstrated that diets high in UFAs did not exert an influence on milk production. However, it is a well-known fact that some variables including sources and type of

fat, supplementation level, and lactation stage play a key role in the overall effect of dietary fat on MY (Plata-Pérez et al., 2021). Moreover, in the investigations included in the current meta-analysis, diets with and without SFO supplements were generally isoenergetic. Therefore, the diets differed only for the energy source (fat vs. starch) and not for the energy density (Vargas-Bello-Pérez et al., 2021). This result in MY was confirmed by a meta-analysis and meta-regression carried out to compare the dietary oil seeds for dairy cattle on MY and components (Rabiee et al., 2012). An additional explanation for the unaffected MY, even in the context of reduced DMI, may lie in the greater efficiency of milk fat production from dietary FAs as compared to de novo FA synthesis process (Palmquist, 1984). Furthermore, diets supplemented with PUFA tend to have a lower heat increment –the amount of energy lost as heat during the digestive process– per unit of energy consumed compared to other energy sources (Ingvarsen, 2006). Current results suggest that MFP, MPP, and MLP increased with the addition of SFO in dairy goat's diet, supporting the fact



**Fig. 5.** Scatter plot of the meta-regression of difference of crude protein intake in treatment and control diet. The size of the circles represents the weight given to each individual study in the meta-analysis.

that, in general, oilseeds have no effect on MY but enhance milk fat secretion and induce variable effects on milk protein concentrations in goats (Plata-Pérez et al., 2021; Rabiee et al., 2012). Additionally, the increase in MFP with SFO follows the net increase in the FAs brought to the mammary gland due to the lipid supplement in the diet (Bernard et al., 2007). Muñoz et al. (2021) found that the supplementation of whole oilseeds in dairy cows resulted in an increased milk fat concentration, which was partially attributed to an increased availability of preformed FAs for uptake by the mammary gland, due to a higher supply of exogenous FAs provided by the oilseed supplementation. However, it should be noted that research has identified variations in energy partitioning among different dairy ruminant species as a result of dietary fat supplementation (Chilliard et al., 2000). This suggests that these animals allocate energy from their diets in a different fashion depending on the species, potentially influencing how fat supplementation affects their metabolism and production outcomes (Fernández et al., 2020; Lunesu et al., 2021). Compared with other ruminants, goats are more resilient to lipid supplements as they have

less sensitivity to the anti lipogenic effects of some trans-FA isomers during mammary lipogenesis (Chilliard et al., 2014; Vargas-Bello-Pérez et al., 2022), and this might be an explanation for the higher MFP in SFO-supplemented goats over milk fat. Moreover, our results point to the fact that the proportion of dietary fiber was adequate in the experimental trials included in the meta-analysis to promote the formation of acetate and butyrate, which are the main precursors of the FA synthesized in the mammary gland (Chilliard et al., 2014; Vargas-Bello-Pérez et al., 2020). However, the effects of SFO inclusion on MFP were inconsistent. In general, it is accepted that feeding ruminants with PUFA could lead to inhibition of de novo synthesis of milk fat, resulting in decreased MFP (Bionaz et al., 2020; Vargas-Bello-Pérez et al., 2021). Some studies have explained the changes in MFP by the traditional glycogen/insulin theory (Bionaz et al., 2020). However, the most acceptable theory was reported by Baumgard et al. (2000), who described that the inhibition of milk fat synthesis, as well as its modified FA composition, may be related to substances produced in the rumen such as the PUFAs. In this study, MFP tended

**Table 5. Effect size, heterogeneity, and publication bias for the effect of dietary sunflower oil on milk fatty acid profiles in dairy goat.**

Outcome	No. of comparison	RMD (95% CI)		Heterogeneity		SMD (95% CI)	Publication bias Egger
		Random effect	P-value	I <sup>2</sup>	P-value		
C4	14	0.070 (-0.049, 0.188)	0.249	78.61	<0.001	0.362 (-0.050, 0.774)	0.712
C6	14	0.132 (-0.013, 0.278)	0.075	94.95	<0.001	1.107 (0.171, 2.042)	0.008
C8	14	-0.221 (-0.405, -0.037)	0.018	85.63	<0.001	-0.698 (-1.324, -0.072)	0.761
C10	14	-1.988 (-2.737, -1.240)	<0.001	91.01	<0.001	-1.613 (-2.369, -0.857)	0.923
C12	14	-1.217 (-1.875, -0.558)	<0.001	97.42	<0.001	-2.186 (-3.268, -1.104)	0.492
C14	14	-1.753 (-2.599, -0.908)	<0.001	96.30	<0.001	-2.203 (-3.243, -1.163)	0.112
C15	14	-0.233 (-0.339, -0.128)	<0.001	94.40	<0.001	-1.871 (-2.785, -0.957)	0.199
C16	14	-4.889 (-7.577, -2.202)	<0.001	97.98	<0.001	-2.340 (-3.518, -1.163)	0.272
C17	14	-0.118 (-0.183, -0.052)	<0.001	95.12	<0.001	-1.694 (-2.453, -0.934)	0.302
C18	14	3.030 (1.411, 4.649)	<0.001	96.54	<0.001	2.046 (1.023, 3.069)	0.554
C18:1 cis9	12	2.225 (0.170, 4.279)	<0.001	97.10	<0.001	0.963 (0.090, 1.836)	0.103
C18:1 trans 11	10	2.774 (1.544, 4.005)	<0.001	97.64	<0.001	2.840 (1.641, 4.039)	0.064
C18:2 cis9 trans 11	10	0.261 (0.162, 0.360)	<0.001	90.80	<0.001	1.373 (0.800, 1.946)	0.011
C18:3 n3	12	0.078 (-0.127, -0.030)	0.002	84.25	<0.001	-0.677 (-1.047, -0.307)	0.032
C18:3 n6	10	-0.010 (-0.15, -0.005)	<0.001	62.86	0.004	-0.744 (-0.340, -0.148)	0.559
SFA	10	-7.530 (-13.095, -1.965)	0.008	97.70	<0.001	-2.214 (-3.603, -0.826)	0.329
MUFA	10	7.163 (2.564, 11.762)	0.002	97.35	<0.001	2.371 (1.044, 3.698)	0.377
PUFA	10	1.492 (0.701, 2.283)	<0.001	94.26	<0.001	1.493 (0.654, 2.333)	0.102

RMD, raw mean difference; CI, confidence interval; SMD, standardized mean difference

to be enhanced by dietary SFO, which could be explained by the fact fat supplementation did not alter energy intake, which is one of the most important nutritional factors affecting MPP (Chilliard et al., 2014; Vargas-Bello-Pérez et al., 2020; Bionaz et al., 2020). However, Oliveira et al. (2021) reported that vegetable oil supplementation did not affect MPP. Similarly, a recent meta-analysis conducted by Gallardo & Teixeira, (2023) highlights that the diets rich in PUFA did not alter MPP. The varying reports in the literature concerning the impacts of vegetable oils on milk protein underscore the necessity for a deeper comprehension of the bioactive functions of those PUFAs in the metabolism and physiology of ruminants. This in-depth understanding could help elucidate the complex interactions between dietary components and milk composition.

Our data revealed that MLP was also enhanced with SFO supplementation in dairy goats. In general, MLP is one of the most consistent components, being less affected by diet type. However, some studies reported increased MLP with vegetable oil supplementation, being associated with an increase in glucose, which is the main precursor of milk lactose (Mahdavi et al., 2019). In addition, the meta-regression results indicated that CPI influences both MPP and MLP

heterogeneity. This suggests that with increasing CPI, both MPP and MLP increase in goats receiving SFO. However, these results should be interpreted with caution because they were based on very few studies.

It has been well documented that dietary inclusion of vegetable oils for dairy small ruminants can serve as an effective strategy to enhance energy intake and improve the milk FA profile, especially with low UFA diets primarily composed of hay or silage (Gómez-Cortés et al., 2011; Nudda et al., 2014; Nudda et al., 2020). Dietary vegetable oil supplements have been shown to alter milk FA composition and enhance the nutritional quality of milk, which varies according to the composition of the basal diet and type of oil (Rabiee et al., 2012; Vargas-Bello-Pérez et al., 2022). The main aim of the current meta-analysis was to quantify changes in milk FA profile in goats fed SFO. In terms of effect size, dietary inclusion of SFO led to an increase in the concentration of C18:0, C18:1 t-11, C18:1 c-9, C18:2 c-9, t-11, C18:2 n-6, and C18:3 n-3, but to a decrease in the short-chain FAs (C8:0-C16:0). In addition, SFO was accompanied by a lower concentration of SFA and higher PUFA. This result agrees with previous studies on vegetable oils for dairy goats (Rabiee et al., 2012; Vargas-

**Table 6. Summary of meta-regression analysis for the effect of dietary sunflower oil on milk fatty acid profiles in dairy goat.**

Outcomes	Covariate	Slope	<i>p</i> - value	Intercept	<i>p</i> - value
C4	FORI	0.659	0.497	0.047	0.503
	NDFI	-0.002	0.142	0.043	0.540
	EI	0.002	0.146	-0.084	0.479
	CPI	0.003	0.139	0.049	0.395
C6	FORI	2.047	0.139	0.050	0.592
	NDFI	-0.00006	0.729	0.103	0.242
	EI	0.003	0.133	-0.156	0.420
	CPI	0.008	0.027	0.071	0.457
C8	FORI	-0.577	0.737	-0.196	0.113
	NDFI	0.0002	0.909	-0.214	0.046
	EI	-0.002	0.368	-0.057	0.777
	CPI	-0.002	0.482	-0.201	0.058
C10	FORI	-4.013	0.551	-1.875	<0.001
	NDFI	0.006	0.405	-1.887	<0.001
	EI	-0.019	0.028	-0.575	0.427
	CPI	-0.024	0.109	-1.864	<0.001
C12	FORI	-4.969	<0.001	-1.33	<0.001
	NDFI	0.004	0.583	-1.159	0.001
	EI	-0.014	0.048	-0.266	0.635
	CPI	-0.025	0.023	-1.163	<0.001
C14	FORI	-10.859	0.167	-1.467	0.003
	NDFI	0.013	0.169	-1.573	0.001
	EI	-0.031	<0.001	0.304	0.643
	CPI	-0.046	0.002	-1.655	<0.001
C15	FORI	-1.354	0.183	-0.195	0.003
	NDFI	0.0009	0.414	-0.220	<0.001
	EI	-0.004	<0.001	0.040	0.639
	CPI	-0.005	0.003	-0.215	<0.001
C16	FORI	-18.039	0.484	-4.410	0.007
	NDFI	0.044	0.101	-4.338	0.001
	EI	-0.064	0.022	-0.550	0.802
	CPI	-0.115	0.012	-4.666	<0.001
C17	FORI	-0.356	0.554	-0.107	0.006
	NDFI	0.0004	0.508	-0.112	<0.001
	EI	-0.001	0.071	-0.015	0.810
	CPI	-0.001	0.175	-0.112	0.001
C18	FORI	-0.393	0.979	3.033	0.002
	NDFI	-0.026	0.115	2.678	0.001
	EI	0.022	0.329	1.502	0.400
	CPI	0.032	0.374	2.960	<0.001
C18:1 cis9	FORI	-18.06	0.276	2.528	0.012
	NDFI	-0.014	0.481	2.171	0.023
	EI	0.024	0.545	0.803	0.758
	CPI	0.042	0.426	2.395	0.031
C18:3 n3	FORI	-0.344	0.370	-0.065	0.027
	NDFI	0.0003	0.617	-0.085	0.007
	EI	-0.001	0.001	0.036	0.383
	CPI	-0.001	0.070	-0.074	0.002

FORI: difference of forage intake in treatment and control diets; NDFI: difference of neutral detergent fiber intake in treatment and control diet; EI: difference of ether extract intake in treatment and control diet; CPI: difference of crude protein intake in treatment and control diets

Bello-Pérez et al., 2020). Similarly, several authors have reported a reduction in the de novo FA in the milk of dairy cows receiving diets enriched with vegetable oils (dos Santos Neto et al., 2021; Prom and Lock, 2021; Gallardo and Teixeira, 2023). Moreover, a significant decrease in contents of 12:0, 14:0, and 16:0 without changes in 4:0, 6:0, and 8:0 levels in milk fat is a frequent observation in goats fed supplemental SFO (Marín et al., 2011, 2012; Titi et al., 2011; Bernard et al., 2005). The inclusion of dietary oil sources rich in long-chain UFA for dairy cattle is often characterized by inhibition of the de novo synthesis of short- and medium-chain FAs in the mammary glands (Salles et al., 2019). This can be explained by one of the following situations: i) a lower volatile FAs (VFA) production in the rumen due to dietary UFA would decrease the level of FA synthesis in the mammary cell; or ii) the long-chain FA taken up by the mammary gland could inhibit enzymatic activities in the FA synthesis pathways in the mammary gland (Marín et al., 2012). The latter may be associated with the reduction of acetyl-CoA carboxylase activity as the mammary gland increases uptake and preferential incorporation of exogenous long-chain FAs derived from the diet or adipose tissue into milk fat (Titi et al., 2011; Chilliard et al., 2014). It has also been well established that the reduction in de novo FAs following the ingestion of vegetable oils can be attributed to the inhibitory effect of CLA in the mammary gland (Guo et al., 2024; Wang et al., 2023). Our study revealed a significant increase in the levels of cis-9, trans-11, C18:3 n-3, and C18:3 n-6 CLA. Concurrently, the decrease in de novo FAs in milk is associated with reduced synthesis of lauric (C12:0), myristic (C14:0), and, to a greater extent, palmitic acid (C16:0). These findings align with a recent meta-analysis that reported a decrease in 12- to 16-carbon FAs following vegetable-sourced PUFA supplementation (Gallardo and Teixeira, 2023). In addition, Mahdavi et al. (2019) found that the lower short-chain FAs following vegetable oils ingestion are attributed to the impaired ruminal fermentation of fiber, decreasing acetate formation, which is the main precursor for de novo milk fat synthesis of short-chain FAs in the mammary gland. Since short-chain FAs are considered hypercholesterolemia (Chilliard et al., 2014), producing milk with a reduced content of these FAs, the use of supplemental SFO could be interesting for the dairy industry. It is noteworthy that both the quantity and physical form of vegetable oils in the diet (Chilliard et al., 2003; Nudda et al., 2014; Nudda et al., 2020; Leduc et al., 2021), as well as their interactions with other dietary components and supplements (Cieslak et

al., 2010), may influence the milk fatty acid profile concentration in sheep and goats, potentially acting as a source of heterogeneity in the present study. Therefore, these factors should be taken into account in the development of feed strategies at both farm and industry levels, as well as in the research process, to decrease the noise effect by considering them as covariates when feasible.

Titi et al. (2011) reported that SFO supplementation decreases the desaturation ratio of C18:0 in the mammary gland, and thus increases the availability of either PUFA or trans-FAs as these FAs are putative inhibitors of the delta 9-desaturase. Moreover, Razzaghi et al. (2015) showed that the feeding SFO increases C18:0 and C18:1 at the expense of the short and medium-chain FAs, leading to both total and partial hydrogenation of the UFA taking place in the rumen, and probably a large extent to unidentified trans isomers of C18:1. It has also been demonstrated that supplementation of UFA for dairy cattle increases the content of trans-18:1 FAs in milk fat (Salles et al., 2019). The results of the present study are consistent with this pattern. The potential human health benefits have drawn the attention of researchers to the development of effective nutritional strategies to increase the CLA content of milk fat (Plata-Pérez et al., 2022; Bionaz et al. 2020). The C18:2 c-9, t-11 (rumenic acid) is also an important product of incomplete biohydrogenation of C18 PUFA; therefore, the higher concentration of this FA agrees with what was expected for goats supplemented with SFO (rich in  $\omega$ 6 PUFA) (Vargas-Bello-Pérez et al., 2022). Our results demonstrated that following the dietary addition of SFO, there are changes toward healthier goat milk from a human standpoint, as there are increases in some bioactive FA such as C18:1 c-9, which have been reported to prevent cancer, hypertension, atherosclerosis, and diabetes as well as enhancing immune function (Bernard et al., 2005; Salles et al., 2019; Vargas-Bello-Pérez et al., 2021). Milk and dairy products are the major sources of SFA in the diet in most developed countries. When SFO is included in the diet of dairy goats, SFA can be replaced with MUFA and PUFA in milk, offering a mechanism to lower SFA consumption in the human population (Saran et al., 2022; Vargas-Bello-Pérez et al., 2021). Overall, our results suggest that the amount of dietary SFO supplementation was conceived as a factor to promote a healthier milk FA profile, without affecting overall animal performance. While the present study did not examine the potential implications of utilizing transgenic SFO on milk quality, there is a need for further research into this area. In this sense, knowledge of the effects of transgenic SFO on the bioethics

and biosafety of ruminant products would contribute to the adoption of sustainable and responsible practices within the dairy industry. It is noteworthy mentioning that, although an  $F$  value of more than 50% could represent high heterogeneity, eliminating possible sources of heterogeneity would have led to study very few articles due to their low methodological quality. Therefore, caution must be paid when extrapolating and interpreting the obtained results.

## CONCLUSION

Dietary inclusions of sunflower oil (SFO) had marked effects on DMI, milk composition, and FA profile of goats. Responses to SFO supplementation were heterogeneous for all variables studied. However, MFP, MPP, and MLP increased, DMI decreased, and MY was not significantly affected by SFO feeding. Our meta-analysis data also indicated that SFO inclusion in dairy goats improved milk FA profile from a human health perspective. These changes resulted in reduced total SFA and increased contents of potential healthy FAs, such as natural trans (C18:1 t-11 and C18:2 c-9, t-11) and PUFA (C18:2 and C18:3) in milk, without detrimental effects on MY and milk composition. Moreover, SFO supplementation seems to be an effective way of decreasing the saturated/unsaturated ratio. Therefore, the dietary addition of oils rich in PUFA, like SFO, could be a convenient feeding strategy for dairy goats for the development of new value-added products. Furthermore, goats seem to tolerate the addition of UFA well, without detrimental effects on animal performance. However, a cost-effectiveness analysis is needed to provide accurate recommendations to farmers and the dairy goat industry.

## Conflict of interest

The authors declare that they have no conflicts of interest.

## Author contributions

Babak Darabighane, Navid Ghavipanje, Lizbeth E. Robles Jimenez, Manuel Gonzalez-Ronquillo, and Einar Vargas-Bello-Pérez had active participation in the bibliographic review. Babak Darabighane had active participation in the development of the methodology. Navid Ghavipanje, Lizbeth E Robles Jimenez, Manuel Gonzalez-Ronquillo, and Einar Vargas-Bello-Pérez had active participation in the discussion of the results. All authors reviewed and approved the final version of the article.

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