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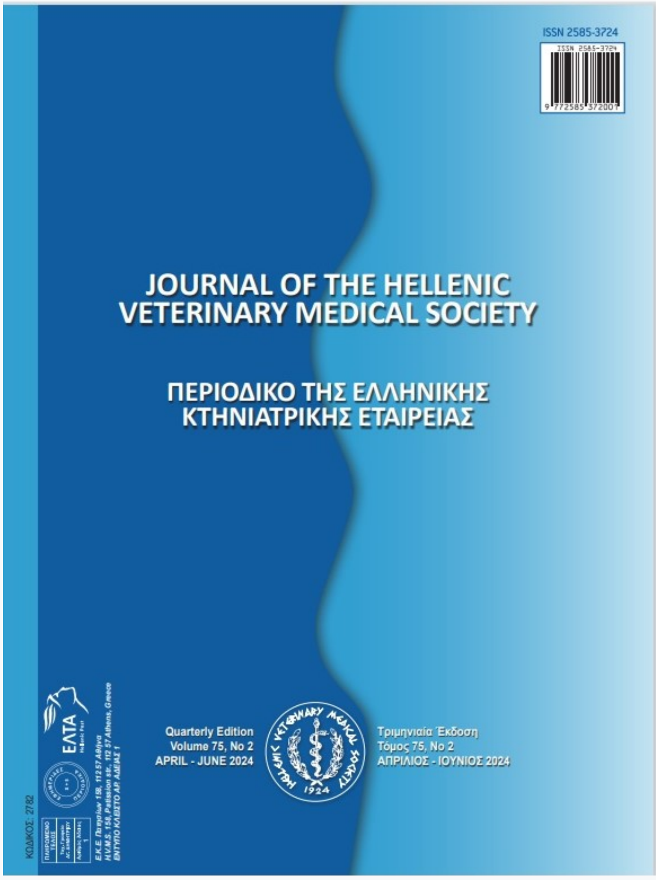

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Conjugated linoleic acid in meat

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ABSTRACT: Conjugated linoleic acid (CLA) consists of a group of geometric and positional isomers of linoleic acid. According to various studies, CLA has been reported to exhibit beneficial effects against cancer, atherosclerosis, diabetes, obesity, cardiovascular malfunction, or enhanced immune function. The most important isomers of CLA found in meat are cis-9, trans-11 CLA and the trans-10, cis-12 CLA. Naturally occurring CLA originates mainly from bacterial isomerisation as well as biohydrogenation of polyunsaturated fatty acids (PUFA) in the rumen of ruminant animals and endogenous formation from desaturation of trans-fatty acids in the tissues of monogastric as well as ruminant animals. Several factors such as seasonal variations, animal breeds, management or diet are influencing the CLA content in meat. In ruminants, the CLA content in meat is usually higher than this of monogatric animals orincluding chicken. The CLA content in beef and lamb is usually above 1 mg/g fat, while in pork, horse and chicken meat is usually found lower than 1 mg/g fat. The storage (refrigeration or freezing) as well thermal processing such as cooking seems to have no effect on CLA content of meat.

***Keywords*:** Conjugated linoleic acid; meat; meat products; fatty acids; meat storage

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INTRODUCTION

Conjugated linoleic acid (CLA) is a group of isomers of linoleic acid (*cis*-9, *cis*-12, Octadecadienoic acid), with two double bonds in positions 9 and 12 and *cis* configuration (*cis*-9, *cis*-12). In linoleic acid the two double bonds are separated with two single bonds, while in CLA the two double bonds are separated with a single bond. In CLA, the two double bonds of CLA are placed in positions 9 and 11, or 10 and 12, but double bonds have also been found in other positions. These double bonds can be found in either *trans* or *cis* configuration and form positional varieties as well as geometrical positional varieties depending on the conformations (cis / *cis*; *cis* / *trans*; *trans* / *trans* or *trans* / *cis*), which results in the formation of several CLA isomers (Griinari and Bauman,1999).

The predominant CLA isomers found in foods are the *cis*-9, *trans*-11 CLA isomer and the *trans*-10, *cis*-12 CLA isomer (Chikwanha et al. 2018). The *cis*-9, *trans*-11 CLA and the *trans*-10, *cis*-12 CLA accounts for 85-90% among the total CLA isomers in ruminant meat products (Dhiman et al. 2005). However, the major CLA isomer is *cis*-9, *trans*-11 CLA found in higher amounts (almost 80%) than *trans*-10, *cis*-12 CLA in many ruminant food products (Griinari and Bauman,1999).

In ruminants, CLA is either formed in the rumen by the enzymatic action of bacteria on polyunsaturated fatty acids or endogenously in the tissues and mammary gland by the enzymatic activity of *Δ*-desaturase on C18:1 *trans*-11 Vaccenic acid. Several beneficial health effects have been attributed to CLA consumption of CLA. Thus, CLA exerts anticancer activity, acts against atherosclerosis, diabetes, obesity, inflammation, colitis, or cardiovascular malfunction (Benjamin et al., 2015; Yang et al., 2015).

The present review provides published data on the presence of CLA in meat of ruminants or monogastric animals from works conducted in many countries in recent years.

CLA in MEAT

The CLA levels in meat depend on several factors such as animal species, animal breed, diet, rearing conditions, pasture grazing, seasonality of the year, etc. Among them, the most important factor is the animal's diet.

1. Feeding factors

1.1 Ruminants

Since pasture grass is rich in monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA) as well as the fatty acids of linoleic acid (C18:2 *cis*-9, *cis*-12), α-linolenic acid (C18:3 *cis*-9, *cis*-12, *cis*-15) and γ- linolenic acid (C18:3 *cis*-6, *cis*-6, *cis*-12), the precursors of the *cis*-9, *trans*-11 CLA synthesis in rumen of pasture grazing ruminants, is leading to increased CLA levels in meat (Realini et al., 2004; Zervas and Tsiplakou, 2011; Fiorentini et al., 2018). Continental crossbred steers fed under pasture grazing or conventional feed presented *cis*-9, *trans*-11 CLA levels of 1.08% and 0.37% (Fatty acids methyl esters) FAME, respectively (French et al., 2000).

The intramuscular fat of Aberdeen Angus crossbred steers finished at pasture showed higher CLA levels as compared to those fed under conventional feeding conditions (Patino et al., 2014). Nuernberg et al. (2005a) reported that exclusive pasture grazing of calves (Holstein and Simmental breeds) resulted in increased *cis*-9, *trans*-11 CLA in intramuscular fat ranging from 0.50% to 0.75% FAME compared to corresponding calves reared on concentrate diet. The proportion of *cis*-9, *trans*-11 CLA in intramuscular fat was clearly higher in lambs (Black Head x Gotland breeds) reared on pasture than in those fed on concentrate feed in the stall (1.9 mg/g versus 1.11 mg/g FAME) (Nuernberg et al., 2005b). Santos-Silva et al. (2002) found that pasture grazing lambs had a higher proportion of *cis*-9, *trans*-11 CLA in intramuscular fat than those fed in the farm with concentrate feeding (0.87% versus 0.24% FAME).

Feeding ruminants with diets rich in polyunsaturated fatty acids (PUFA) and conjugated linoleic acid (CLA), such as oilseeds (e.g., sunflower, linseed, cardamom), vegetable oils (e.g., sunflower oil, soybean oil), or fish oils, has been demonstrated as an effective method for increasing CLA levels in the intramuscular fat of these animals (Schmid et al., 2006; González et al., 2014). For instance, a study showed that supplementing fattening calves (Brown Swiss) with sunflower seeds (3%) led to a significant rise in CLA content in their intramuscular fat, reaching levels of 7.8 mg/g fat, compared to 5.6 mg/g fat in the control group (Scheedera et al., 2001). Mapiye et al. (2013) reported that the addition of sunflower seeds (18.4%) to the supplementary feeds of calves resulted in higher CLA content in intramuscular fat of 0.79

% FAME versus 0.67% FAME of control group. The addition of cardamom nuts (5%) to the supplementary feeds of fattening calves resulted in an increase in the content of *cis*-9, *trans*-11CLA in intramuscular fat (1.10% FAME) (Bottger et al., 2000). Wachira et al. (2002) reported that feeding lambs of Suffolk, Soay and Friesland breeds with feeds supplemented with linseed (10%) resulted in an increase in PUFA in intramuscular fat (*longissimus dorsi* muscles), with *cis*-9, *trans*-11CLA levels ranging between 10, 14 and 16 mg/g fatty acids, respectively. The positive effect of flaxseed feed supplementation on the increase in CLA in the intramuscular fat of lambs was also reported by Noci et al. (2011). Meat from Nellore steers fed with concentrated feeds supplemented with soybean, sunflower, linseed oil (3.50%) or a control diet presented *cis*-9, *trans*-11 CLA levels of 0.47, 0.43, 0.39 and 0.25% FAME, respectively (Costa et al., 2020). Nellore bulls aged 24 months and fed with sunflower cake (0, 9.0, 18.0 and 27% dry matter) for 105 days showed *cis*-9, *trans*-11 CLA levels in *longissimus* muscles of 0.58, 0.70, 0.73 and 0.82% FAME, respectively (Oliveira et al., 2019). Suffolk lambs (4 months old) fed a concentrated diet supplemented with radiata pine bark polyphenolic extract at 0, 1 and 2% dry matter for 35 days resulted in *trans* 10, *cis* 12 CLA levels of 0.58, 0.72 and 0.90% FAME, respectively (Vera et al., 2023).

The *trans* 10, *cis* 12 CLA was significantly higher in meat of Barki breed lambs fed with concentrated feed with flaxseed oil (3%) as compared to control group (El-Sabaawy et al., 2015). The supplementation of 0, 55, and 110 g of sunflower oil to feed of calves for a period of 142 days resulted in *cis*-9, *trans*-11 CLA levels in fat of *longissimus dorsi* muscles of 4.3, 6.3, and 9.1 mg/g, respectively (Noci et al., 2005). The *cis*-9, *trans*-11 CLA in intramuscular fat reached 0.8% of fatty acids in calves fed 0.6% linseed oil (Enser et al., 1999). However, the supplementation of palm kernel oil at various levels (1.3% - 5.2%) in concentrated feed of Santa Ines lambs for a feeding period of 96 days has no important effect on *cis*-9, *trans*-11 CLA levels (0.40 - 0.44% FAME) in fat of *Longissimus lumborum* muscle (Castro et al., 2022).

Goats (Black Beenal breed) aged 1 year were fed a concentrated diet (67 g/kg) enriched with sunflower oil and soybean oil for a period of 30 days, resulting in an augmentation of polyunsaturated fatty acids (PUFA) and cis-9, trans-11 CLA in their intramuscular fat (Roy et al., 2013). The study showed that the levels of cis-9, trans-11 CLA in the intramuscular fat (*Longissimus dorsi* muscle) of the goats were 0.38% for the control group, 0.79% for the soybean oil group, and 1.14% for the sunflower oil group, respectively. Maia et al. (2012) examined the addition of castor oil in concentrated diet (30g/Kg) of 2-month-old goats (Boer × Saanen breeds) for a feeding period of 56 days and found that the *cis*-9, *trans*-11 CLA and *trans*-10, *cis*-12 CLA levels in the intramuscular fat (*Longissimus dorsi* muscle) were 0.19% and 0.16% FAME, while in controls were 0.14% and 10% FAME, respectively.

Badee and Hidaka (2014) examined the supplementation of soybean oil (SBO) and sunflower oil (SFO) in two concentrations (1.8% and 3%) in concentrated feed to 6-month-old Awassi lambs for a feeding time of 62 days, using 4 groups of lambs according to the amount of oil, assigned as SBO1.8%, SBO3%, SFO1.8% and SFO3%. The *cis*-9, *trans*-11 CLA in the intramuscular fat of various muscles was 0.9, 1.1, 1.2, 1.0 and 1.3 mg/g FAME for the groups of lambs fed SBO1.8%, SBO3%, SFO1.8% and SFO3%, respectively. Manso et al. (2009) found that the addition of sunflower oil (4%) and palm oil (4%) to concentrated feed of 9-week-old Merino lambs for a feeding time of 2 months resulted in a small increase in *cis*-9, *trans*-11 CLA in intramuscular fat (0.45 mg/g FAME) versus (0.41 mg/g meat) of the control group, while palm oil 4% showed no substantial change. Awassi lambs (aged 3 months) fed with control concentrated diet or a diet with combined quebracho tannins (20 or 40 g/kg diet) or sunflower oil (20 or 40 g/kg diet) for a feeding time of 70 days, presented *cis*-9,*trans*-11 CLA in *Longissimus dorsi* muscle fat of 0.40 and 0.73 and 0.93 g/100g total FA, respectively (Kamel et al., 2018).

The addition of fish oil (0.6%) to the concentrated feed of Charolais steers for feeding time of 120 days resulted in an increase in *cis*-9, *trans*-11 CLA in the *Longissimus lumborom* muscle fat (5.7 mg per 100 g muscle versus 3.2 mg per 100 g muscle in control) (Enser et al., 1999). The addition of fish oil (60 g/Kg diet) to concentrated feed of lambs (Suffolk, Soay and Friesland breeds) aged 8 months had a positive effect on the increase in intramuscular *cis*-9, *trans*-11 CLA only in the Soay breed (1.78 mg/g fat versus 1.32 mg/g fat in), while no important *cis*-9, *trans*-11 CLA changes were found in the rest two breeds (Wachira et al., 2002). The partial replacement of soybean oil

(40 g/Kg diet) by fish oil at concentrations of 0, 2.5, 5 and 7.5 g/Kg diet in lambs (Santa Inês breed, aged 2 months) during a rearing period of 3 months showed an increase of *cis*-9, *trans*-11 CLA in intramuscular fat to 0.78, 1.11, 1.07, 1.22, 1.13 mg /g FAME, respectively (Ferreira et al., 2014). Charolais-sired crossbred were fed with concentrated feed with 58 g fish oil/Kg, 137 g sunflower oil/Kg, 46 g soyabean/Kg, 49 g cane molasses/Kg and 686 g pollard/Kg for 22 weeks, and post-slaughter the *cis*-9, *trans*-11 CLA levels in intramuscular fat were 61 and 71 mg/100 g muscle in control and supplemented diet, respectively (Moloney et al., 2022).

1.2 Monogastric animals

In ruminants, the fatty acids consumed with feed undergo changes due to microbial fermentations in the rumen in contrast to monogastric animals where the fatty acids contained in animal feed remain almost unchanged during their digestion and blood absorption. In order to increase the CLA levels in meat of monogastric animals, the feed should contain trans-fatty acids (such as trans-vaccenic acid) as a substrate for the endogenous synthesis of CLA in their tissues or CLA supplements should be added to their feed (Wang et al., 2021). CLA supplements commonly used in the diet of monogastric animals consist of a mixture of CLA isomers (mainly *cis*-9, *trans*-11 CLA and *trans*-10, *cis*-12 CLA) (Pinelli-Saavedra et al., 2019).

Supplementation of pig diets (Large White × Landrace breed) with 1% CLA or 1% sunflower oil resulted in *cis*-9, *trans*-11 CLA concentrations in pig muscle of 5.5 mg/g and 0.9 mg/g, respectively (Eggert et al., 2001). The concentrations of cis-9, trans-11 CLA in the pig *Longissimus dorsi* muscle increased to 0.1, 3.7, 10.1, and 11.6 mg/g of fat with, respectively, 0%, 1.0%, 2.5% or 5.0% CLA supplementation in the feed for a feeding period of four weeks (Joo et al., 2002). Dietary administration of 2% linseed extract with 1% CLA to pigs (Large White) for a period of gaining 100 Kg pig body weight (initial weight 60 Kg) resulted in *cis*-9, *trans*-11 CLA in intramuscular fat (*semimembranosus* muscle) of 0.94 mg/g fat as compared to none in the groups given only 1% and 2% linseed extracts (Bee et al., 2008).

The addition of 1% CLA isomers mixture (50% *cis*-9, *trans*-11 CLA and 50 % *trans*-10, *cis*-12 CLA) to the diet of pigs for a 6weeks feeding time, resulted in abdominal muscles fat *cis*-9, *trans*-11 CLA and *trans*-10, *cis*-12 CLA levels of 0.95 mg/g FAME and 0.45 mg/g FAME, respectively, while addition of 1% soybean oil in the diet showed no detectable CLA levels (Barnes et al., 2012). Morel et al. (2013) reported that the supplementation of pig diets with 4.4% fish oil and 1.1% linseed oil for 84 days feeding period resulted in *cis*-9, *trans*-11 CLA levels in *Longissimus dorsi* muscle fat of 1.04 mg/g FAME and 0.73 mg/g FAME, respectively. The addition of palm oil 2%, palm oil 2% and CLA 0.5%, palm oil 2% and CLA 1% in pig diet resulted in *cis*-9, *trans*-11 CLA in intramuscular fat (*Longissimus dorsi* muscle) of 0.076, 0.195 and 0.341 mg/g FAME, respectively (Intarapichet et al., 2008). The supplementation of CLA 0% (control), CLA 0.5% and CLA 1% in the basal feed of finishing pigs (Landrace - Yorkshire) for a feeding time of 30 days resulted in *Longissimus thoracis* muscle *cis*-9, *trans*-11 CLA levels of 0.031, 0146 and 0.211 mg/100 g tissue, respectively, or *trans*-10, *cis*-12 CLA levels of 0.012, 0.051 and 0.092 mg/100 g tissue, respectively (Pinelli-Saavedra et al., 2019).

The meat from two crossbred horse groups displayed varying levels of cis-9, trans-11 CLA. The first group, consisting of suckling foals raised under grazing conditions and slaughtered at 4 months of age, exhibited a CLA level of 0.407 mg/g meat. In contrast, the second group, comprising concentrate-finished foals slaughtered at 12 months of age, showed a significantly higher CLA level of 1.28 mg/g meat (Belaunzaran et al., 2018). Similarly, Hispano-Bretón horses aged between 6 and 8 months were reared in a commercial farm under grazing conditions until 11-13 months of age. After slaughtering, these horses were fed on a concentrated diet and straw *ad libitum* for 100-120 days before presenting a cis-9, trans-11 CLA level of 0.0535% FAME in both muscle and subcutaneous fat (Beldarrain et al., 2021). Corino et al. (2007) reported that the feeding of rabits (New Zealand White breed) aged 55 days with feed supplemented with 0 και 0.5% CLA for a feeding time of 37 days resulted in *cis*-9, *trans*-11 CLA levels in intramuscular fat of 0.4 mg/g fat and 2.2 mg/g fat, respectively. Feeding of Minxinan black rabbits (age 75 days) by using basic diet supplemented with 0 (control), 0.5%, 1.0%, 1.5% CLA isomers mixture (*cis*-9, *trans*-11 CLA 36.0%, *trans*-10, *cis*-12 CLA 41.7%, other isomers CLA 3.3%) for 55 days resulted in intramuscular both *cis*-9, *trans*-11 CLA levels of 0.03, 0.55, 1,07 and 1.77 % of total fatty acids, respectively (Liu et al., 2022). Other studies also revealed that feeding synthetic CLA to rabbits can enrich their meat with CLA (Marounek et al., 2007; Petracci et al., 2009).

1.3 Poultry

Du and Ahn, (2002) reported that broiler chickens (3 weeks old) fed a diet with 0%, 2.0% or 3.0% CLA for 5 weeks presented a linear increase in breast muscle *cis*-9, *trans*-11 CLA of 0, 32.8 and 53.7 mg CLA/g fat, respectively. Szymczyk et al. (2001) examined the addition of CLA (0%, 0.5%, 1.0% and 1.5%) to the diet of chickens (Arbor Acres) for a feeding period of 35 days and found that *cis*-9, *trans*-11 CLA levels in breast muscle were 0.2, 8.9, 52.5, and 93.5 mg/g fat, respectively.

In their study, Kawahara et al. (2009) conducted an experiment where 28-day-old chickens (Abor Acres) were fed mixtures of safflower oil and CLA (20 g/kg) in the diet. The mixtures were prepared in three different ratios: 2:0, 1:1, and 0:2 (weight:weight) and were given to the chickens for a period of 28 days. The researchers observed that the intramuscular total CLA isomers in the breast of the birds were 3.40, 24.30, and 47.70 mg/g FAME, respectively, for the three different ratios. They also noted that the major isomer of CLA present in the breast muscle tissue was cis-9, trans-11 CLA. On the other hand, Herzallah (2013) conducted a study to investigate the impact of dietary administration of probiotic lactic acid bacteria (LAB) on the level of CLA formation in chicken breast muscle tissue. The LAB were *Lactobacillus plantarum*, *Lactobacillus lactis*, *Lactobacillus casei* and *Lactobacillus fermentum* and a mixture of four *Lactobacillus reuteri* strains of (1 ml with bacterial populations of 10^6 CFU/ml) in 1-day-old chicks for a feeding time of four weeks. The results showed that only the *L. reuteri* strains increased the breast *cis*-9, *trans*-11 CLA from initial 0.3 mg/g to final 1.88 mg/g FAME. Chicks (Ross, 1day old) or ducks (Pekin ducks, 1day old) fed with a diet with 0.0, 0.1 and 0.2 % CLA for 35 and 49 days, respectively, presented breast *cis*-9, *trans*-11 CLA of 24.3, 38.0 and 68.1 mg/g fat, and 0.0, 41.1 and 68.1 mg/g fat for the chicken and duck diet groups, respectively (Halle et al., 2012).

Szymczyk and Szczurek (2016) studied the effect of pomegranate seed oil and linseed oil supplementation in the diet on meat fatty acid profile and performance of broiler chickens (Ross 308) aged 22 days for a feeding time of 35 days. They found that the supplementation of pomegranate seed oil (0.0, 0.5, 1.0 and 1.5%) in the diet showed *cis*-9, *trans*-11 CLA levels in meat of 0.74, 1.87, 2.78 and 4.47 g/100 g of fatty acids, respectively, while linseed oil (0.0 and 2%) in the diet resulted in *cis*-9, *trans*-11 CLA levels in meat of 2.65 and 2.28 g/100 g of fatty acids, respectively. The supplementation of grapeseed oil at 0%, 1,5% and 2% levels in the diet of broilers Ross 308 (21days age) for a feeding period of 22 days resulted in *cis*-9, *trans*-11 CLA levels in the thigh muscle of 0.84, 0.88 and 0.95 mg/g of fat, respectively (Bialek et al., 2018).

Other factors

Differences in CLA concentrations have been observed between different muscles of the same animal, among different breeds as well as between animals of the same breed. European x British and Wagyu cattle-breeds, fed the same diets, presented *cis*-9, *trans*-11 CLA levels in the intercostal muscles of 1.7 and 1.8 mg/g fat, respectively (Mir et al., 2002). Wachira et al. (2002), compared 3 sheep breeds reared with the same diet and found a significantly higher percentage of *cis*-9, *trans*-11 CLA in their *longissimus dorsi* muscles in the Soay breed compared to the Suffolk and Friesland breeds. Alfaia et al. (2007) investigated the effect of slaughter season and muscle type on intramuscular CLA percentage in purebred (Arouquesa) calves reared in traditional pasture-grazed veal fattening units in Portugal. The animals were slaughtered in June and October and the CLA in the *Longissimus dorsi* and *semitendinosus* muscles had differences, particularly in minor CLA isomers (*trans* -12, *trans* -14, *trans* -11 and *cis* -11, *trans* -13 CLA), while differences in CLA levels were also observed in different seasons. Pestana et al. (2012) studied the effect of slaughter season and muscle type on intramuscular CLA levels in purebred (Mirandesa) calves fed under grazing pasture conditions. They observed that the *Longissimus lumborum* muscles had significantly higher CLA levels than the *semitendinosus* muscles, with *cis*-9, *trans*-11 CLA levels of 86.03 vs. 75.61 mg/g muscle in spring and 86.90 vs. 84.03 mg/g muscle in autumn, respectively.

The intramuscular CLA (total CLA isomers) in *Longissimus lumborum* muscle of lambs sheep breeds Altamurana, Bagnolese, Gentile di Puglia, Laticauda, Leccese reared in South Italy under the same feeding conditions were 3.18, 3.07, 1.51, 1.21, 2.96 g/100 g FAME, respectively (Giliberti et al., 2021). Difference in CLA levels in *Longissimus thoraci* muscle of five different Iberian sheep breeds and reared under intensive and semi-extensive rearing conditions were reported by Cadavez et al. (2020). In *Longissimus dorsi* muscle fat of adult Malpura (Indian breed) sheep and lambs the *cis*-9, *trans*-11 CLA levels were 1.06 and 0.19% FAME, respectively, while the *trans*-10, *cis*-12 CLA levels were 0.02% and 0.07% FAME, respectively (Bhatt et al., 2020). According to a study of Gravador et al. (2018), the *cis*-9, *trans*-11 CLA levels in *Longissimus thoracis et lumborum* muscle of lambs were affected by gender, age and breeds in Scottish Blackface or Texel × Scottish Blackface sheep breeds.

The *cis*-9, *trans*-11 CLA in subcutaneous adipose tissue was 4.92, 5.22, 6.42, and 2.68 mg/g fat for the bovine breeds of Salers bulls, Pirenaica bulls, Pirenaica heifers and Holstein-Friesian cows in Spain, respectively (Gamarra et al., 2018). The intramuscular fat (*Longissimus thoracis* muscle) of Italian Simmental Goudali crossbreed young bulls and pure Goudali breed presented *cis*-9, *trans*-11 CLA of 0.23 and 0.16 g/100 g meat, respectively (Ojong et al., 2017).

CLA levels were higher in thigh muscle than breast muscle of chicken (Isa Brown Strain) (Franczyk-Zarów et al., 2017). The *cis*-9, *trans*-11 CLA levels in fat of ducks were affected by duck breeds (Peking duck and Cherry Valley), as well as the age (Fesler and Peterson, 2013).

Juárez et al. (2009a) examined the intramuscular fatty acid of two horse breeds of Burguete and Hispano-Breton and found that *cis*-9, *trans*-11 CLA concentrations were 0.46 mg/g FAME and 0.39 mg/g FAME, respectively. An earlier study showed that CLA in horse muscle was 0.60 mg/g fat (Dufey 1999).

CLA in retail meat

Limited research works has been published on the presence of CLA in the meat from production animals commercialized in the market. Limited research is available for the presence of CLA in retail meat. According to a previous study conducted in 1990-1991 in the USA (Chin et al., 1992), the *cis*-9, *trans*-11 CLA in the intramuscular fat of veal, beef and lamb ranged between 2.9 mg/g fat, 2.7 mg/g fat, 5.6 mg/g fat, respectively, while pig and chicken meat showed very low concentrations of CLA. However, a high concentration of *cis*-9, *trans*-11 CLA (2-2.5 mg/g fat) was determined in turkey meat. Schmid et al. (2006) summarizing research data from studies in Europe and the USA, and reported that the highest concentration of *cis*-9, *trans*-11 CLA found in lamb meat (4.3-19.0 mg/g fat), while a slightly lower concentration of *cis*-9, *trans*-11 CLA was in beef (1.2-10.0 mg/g fat). Cicognini et al. (2014) determined the CLA content in meat sold on the Italian market in 2011 and found that lamb meat contained the highest CLA level (9.8 mg/g fat), while beef ranged from 1.28 - 4.24 mg/g fat. For pork and horse meat the CLA levels were 0.67 and 0.34 mg/g fat, respectively. Thus, meat from ruminant animals appears to have the highest CLA concentration, as opposed as opposed to meat of monogastric animal origin.

Effect of storage and processing on CLA of meat

Martin et al. (2009) examined the effect of cold storage (4 ^{o}C) for 7 days of CLA in pork belly muscle meat (Large white x Landrace cross breed), from pigs fed a CLA-fortified diet (0%, 1% and 2%). According to their results, there were no important changes were observed in intramuscular *cis*-9, *trans*-11 CLA levels, throughout the 7 days of storage. Zanini et al. (2006) also reported that *cis*-9, *trans*-11 CLA in chicken breast and leg remained relatively unchanged during cold storage (4 ^{o}C for 7 days) and freezing (-20 ^{o}C for 100 days).

The CLA levels found in processed meat in studies conducted in various countries are shown in Table 3. Alfaia et al. (2010) studied the effect of heat treatment (boiling, baking, microwaving) on CLA and fatty acids of the intramuscular fat of the *Longissimus lumborum* muscle in calves (Alentejano breed). The CLA content in the fat of heat-treated muscles remained the same as in fresh meat, because these forms of heat treatment had negligible changes in CLA (*cis*-9*, trans*-11 CLA or *trans*-10, *cis*-12 CLA). Similarly, Shantha et al (1994) examined patties (beef) with different cooking methods (frying, baking, boiling and microwaving), and found that CLA concentrations in mg/g FAME did not differ greatly between cooked and fresh patties, although higher cooking temperatures enhanced total CLA concentration. Maranesi et al. (2005) also reported that boiling and microwaving had no significant effect on CLA content in lamb abdominal muscles. However, Martin et al. (2009), observed reductions in *cis*-9*, trans*-11 CLA and *trans*-10, *cis*-12 CLA in the muscles of pigs that had been supplemented with CLA enriched oil in their feed, and stored at 4 ^{o}C, in contrast to non-CLA-fortified controls. Schmid et al. (2006) reported that cooking or cold storage of meat does not have any decreasing effect on CLA concentration. Rant et al. (2019) reported that microwaving and roasting of lamb *Longissimus dorsi* muscle did not cause any im-

Table 3: CLA in processed meat products.

Meat Product	Type of meat	Country	Characteristics	*cis*-9, *trans*-11 CLA	*trans*-10, *cis*-12 CLA	Total CLA isomers	Reference
Sausage	beef	Turkey	Safflower 5%, Starter L. plantarum			3.37-3.51 mg CLA/g fat	Özer and Kılıc (2020).
Sausage	Beef (18%)	Poland	Addition of mixtures salt, sodium nitrite, dried acid whey (0.35-0.70%)	0.213 -0.220 %FAME	nd		Kononiuk,and Karwowska (2020).
Sausage	Fallow deer (18%)	Poland	Addition of mixtures salt, sodium nitrite, dried acid whey (0.35-0.70%)	0.100 -0.137 % FAME	0.010 -0.013 % FAME		Kononiuk,and Karwowska (2020).
Canned meat	Beef	Italy	Market	2.01 mg /g fat	0.01 mg /g fat		Cicognini et al. (2014)
Canned meat	Chicken; corned button	Australia	Market	1 mg/100g ; 135 mg/100g			Li et al. (2002)
Sausage	Goat meat/beef 50/50, 75/25 or 100 %	USA	Addition of rice bran 3%	0.63 % FAME; 0.79% FAME; 0.88% FAME; 0.77% FAME;	0.01 % FAME; 0 % FAME; 0.01% FAME; 0.03% FAME;		Malekian et al. (2016)
Sausage (Sucuks)	Beef (70 % lean meat and 22 % fat)	Turkey	Addition of 0.5 %, 1 %, 1.5 %, 2 %, 2.5 %, and 3 % CLA			Ranged between 2.97 - 3.15 % FAME	Özer and Kiliç (2015)
Cooked sausage	Beef 52,5 %, Pork 15%	Brazil	20% fat, 25,50,75,100% replacement of fat by celluse gel	1.3, 1.6, 1.4, 2.2, 2.5 % FAME			Almeida et al. (2014)
Corned beef	Meat from German Holstein bulls	Germany	Control diet with concentrate (2.5 kg) based on soybean meal (41%), wheat (40%), maize (10%) straw and minerals	3.74 mg/100 g	0.22 mg /100 g		Dannenberger et al. (2013).
Corned beef	Meat from German Holstein bulls	Germany	Experimental diet concentrate (2.5 kg) based on triticale (40%), wheat (28%), rapeseed cake (13%) and rapeseed oil (2%)	4.07 mg/100 g	0.17 mg/100 g		Dannenberger et al. (2013).
Tea sausage spread	Meat from German Holstein bulls	Germany	Control diet (mentioned above)	33.15 mg/100 g	2.04 mg/100 g		Dannenberger et al. (2013).
Tea sausage spread	Meat from German Holstein bulls	Germany	Experimental diet (mentioned above)	40.06 mg/100 g	1.09 mg/100 g		Dannenberger et al. (2013).
Scalded sausages	Meat from German Holstein bulls	Germany	Control diet (mentioned above)	18.48 mg/100 g	3.40 mg/100 g		Dannenberger et al. (2013).
Scalded sausages	Meat from German Holstein bulls	Germany	Experimental diet (mentioned above)	17.80 mg/100 g	2.25 mg/100 g		Dannenberger et al. (2013).
Salamin sausage Chorizo sausage Morcilla sausage Chorizo ahumado sausage	Not mentioned	Argentina	Salamin (dry cured sausage), Chorizo (raw sausage) Morcilla (blood sausage) Chorizo ahumado (smoked sausage)	0.03 % FA 0.19% FA 0.06% FA 0.03 % FA			Romero et al. (2013)
Sausage	Pork	Ireland	Pork from pigs of control diet and diet with 2 % CLA supplementation	(Control) 0.08 % FA; (Pig diet supplementation) 3.60 % FA			Juárez et al. (2009b)

Cooked sausage	Pork	Ireland	Pork from pigs fed with 0.9%, 1.8% and 3.6% Sunflower oil	1.0 mg/g, 2.3 mg/g, 5.4 mg/g,	0.60 mg/g, 1.4 mg/g, 3.6 mg/g	Marco et al. (2009).
Cooked sausage	Pork	Ireland	Pork from pigs fed with 0.9%, 1.8% and 3.6% CLA	2.9 mg/g, 5.2 mg/g, 7.1 mg/g,	1.8 mg/g, 3.5 mg/g, 5.0 mg/g	Marco et al. (2009).

portant changes in the content of *cis*-9*, trans*-11 CLA. Boiling or roasting of chicken thigh or breast had no significant effect on *cis*-9*, trans*-11 CLA and *trans*-10, *cis*-12 CLA levels (Franczyk-Zarów et al., 2017). The *cis*-9*, trans*-11 CLA levels in beef or lamb meat following cooking were also not affected (Purchas et al., 2015). Herdmann et al. (2010) reported that the *cis*-9*, trans*-11CLA of beef remained unchanged during the heat treatment for the production of corned beef.

CONCLUSIONS

The lipids derived from ruminants meat are rich in CLA isomers, and in particular in *cis*-9*, trans*-11 CLA with beneficial health properties. The CLA levels in meat are affected by several factors such as animal species, animal breed, diet, rearing conditions, pasture gazing or seasonality. The meat from ruminant animals appears to have the highest CLA concentration, compared to that of monogastric animal origin. Poultry and other monogastric animals feeds enriched with CLA supplements can increase the CLA levels in produced meat. Storage or heat treatment (boiling, baking, microwaving) of meat usually do not affect the CLA levels in meat.

Based on the information gathered and discussed in this review, future research should focus on the:

seasonality effect on CLA content in meat

Lipid oxidation on CLA content in meat

Mechanism of any anticancer properties of specific meat ingredients or meat rich in CLA

Bioavailability and matrix effect on CLA

CONFLICT OF INTEREST

None declared

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