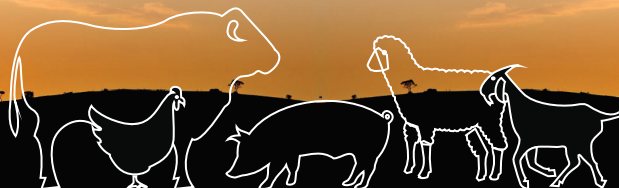


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THE EFFECTS OF FEEDING PIGS WITH CONJUGATED LINOLEIC ACID ON MEAT QUALITY

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Invited paper

Abstract: Conjugated linoleic acid (CLA) is a mixture of the geometric isomers of linoleic acid. Studies have found that feeding animals with the addition of CLA has various positive effects, such as improved feed conversion, reduction of the amount of fat tissue and improvement of the quality of the resulting meat. The reason for the great interest in this compound is also its potential health effects on humans, such as anticancer effect, enhancing the immune system, reducing the occurrence of cardiovascular diseases and the like. Feeding pigs with the addition of CLA is gaining importance in recent years, because it affects the reduction of the thickness and share of subcutaneous fat tissue and the increase of lean meat/meat yield of the carcass without negative impacts on the quality of the resulting meat. Increased content of intramuscular fat, i.e. marbling in meat and fatty-acid changes in the composition of meat have been determined. Increasing the content of saturated fatty acids in the fat and muscle tissue improves technological characteristics and oxidative stability of meat. In addition, due to the incorporation of CLA isomers in the tissues of animals and its health benefits, this type of diet can be a method for obtaining the pork with the features "of functional foods." However, all these positive effects have not been confirmed in each of the experiments focusing on feeding pigs with CLA, because there are many factors that affect its efficiency.

Key words: conjugated linoleic acid, pig, meat quality

Introduction

The composition of the slaughter carcasses is influenced by the breed, gender, diet and housing conditions (*Wagner et al., 1999*), and these factors have an effect on the composition and quality of the resulting meat. Besides them, the quality of meat is also affected by the conditions in animal transport, time spent in

the depot sacrifices, the stunning and primary processing, and cooling conditions and storage (*Stanišić et al., 2012b*).

In recent years, the efforts of science and practice were focused on the reduction of body fat in carcasses of fattening animals, especially pigs (*Stanišić et al., 2011a*). As a result, first of all of the improvement of the genetic basis of modern high-quality pig breeds and their crosses, as well as progress in the field of animal nutrition, the thickness of fatty tissue at the back part is reduced to below 10 mm, and the content of muscle tissue in carcass often exceeds 60%. Pigs today are bred/selected to have an efficient feed conversion and to better convert the food into muscle tissue (*Schinckel et al., 2001*). However, this increase of meatiness had a negative impact on the quality of the resulting meat. There was a negative correlation between the increased share of muscle tissue and some sensory and technological parameters of meat quality, especially subcutaneous fat, as one of the main ingredients for the production of quality meat products (*Stanišić et al., 2011b, 2012a*).

In practice, there are many ways and means which may be used to balance the growth of muscle and fat tissue in the body of animals and produce meat of desired quality. Latest developments in biotechnology have enabled scientists to influence the growth and distribution of certain tissues of animals through modification of their metabolism. Metabolic modifiers, such as β -agonists, anabolic steroids, somatotropin, vitamin E and trace elements and lipids with metabolic effect, such as conjugated linoleic acid, are used in order to improve performance and meatiness of fattening animals.

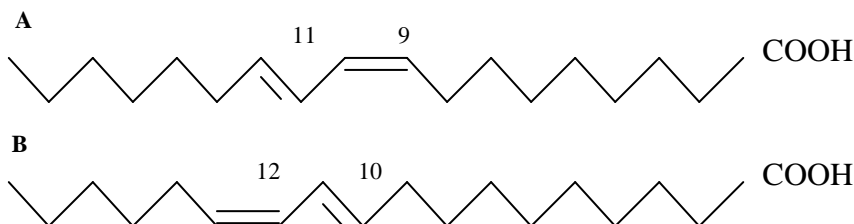
Conjugated linoleic acid (CLA) was first identified in 1935 after an analysis of cow's milk fat using UV spectrophotometer (*Booth et al., 1935*). However, the nutritional potential of CLA was recognized only in 1978, when Dr. Michael Pariza and his associates isolated a substance from roasted beef which showed mutagenic effects (*Pariza et al., 1979*). Later research has shown that this "mutagen" has anti-carcinogenic effects and that it was in fact $cis\Delta^9$, $trans\Delta^{11}$ conjugated linoleic acid derivative (*Pariza and Hargaves, 1985; Ha et al., 1987*). Since then, a number of experiments was performed to study its functional and structural aspects.

CLA is found naturally in milk and fat tissue of ruminants, and formed in the rumen through bacterial biohydrogenation from linoleic acid, however, only a small amount of CLA (about 0.6 mg/g fat) is in pork meat (*Chin et al., 1992*). The only way to increase its share in the pork is adding chemically synthesized CLA in the diet (*Dugan et al., 1997*). Feeding pigs diet with the addition of CLA is gaining importance in recent years because it improves the characteristics of the carcass (*Stanišić et al., 2013*) and quality of pig meat, and at the same time, it is a way of obtaining meat and meat products enriched with CLA (*Lauridsen et al., 2005*).

Synthesis and mechanism of action of CLA

Conjugated linoleic acid (CLA) is a mixture of geometric isomers of $\text{cis}\Delta^9$, $\text{cis}\Delta^{12}$ – octadecadienoic (linoleic acid - 18:2 n-6) acid, for which many positive effects were identified, such as anticancer effect, improved feed conversion and reduction of the amount of fat and increase of the share of muscle tissue in the body of animals (*Pariza and Hargaves, 1985*). Name “conjugated” is derived from the fact that the double bonds in the molecule are separated by two carbon atoms between which there is a single bond. According to the location of the double bond and trans/cis combinations, there are 16 different forms of CLA isomers (*Pastoreli et al., 2005*).

CLA is naturally occurring through the bacterial biohydrogenation of linoleic acid (18: 2 n-6) in the rumen of ruminants by means of several kinds of microorganisms (*Grinari et al., 2000*). Consequently, food derived from ruminants (meat and milk) is the main source of CLA in the human diet (*Chin et al., 1992*). Although the food contains a large number of isomers of CLA, the most common are $\text{cis}\Delta^9$, $\text{trans}\Delta^{11}$ (cis-9, trans-11) and $\text{trans}\Delta^{10}$, $\text{cis}\Delta^{12}$ (trans-10, cis-12) (Picture 1).



Picture 1. Structure of $\text{cis}\Delta^9, \text{trans}\Delta^{11}$ – conjugated linoleic acid (A) and $\text{trans}\Delta^{10}, \text{cis}\Delta^{12}$ - conjugated linoleic acid (B)

There are several mechanisms of action of CLA, but the majority are still not fully explained, and one reason for this, as stated by *Yang et al. (2000)*, is that the CLA oxidizes faster than linoleic acid, which would mean that the conjugated double bond is more susceptible to oxidation than unconjugated. Finding that CLA is more susceptible to oxidation, i.e. that it can be oxidized prior to its utilization, suggests that its effects can be reduced, and this may be one of the reasons for variations in the results of its effects between different experiments.

Several researchers found that CLA affects the increase in energy consumption, because increased oxygen consumption of animals fed CLA was established in the trials (*West et al., 2000*), which represents one of the mechanisms that influence the reduction of fat tissue in the body of animals. Another mechanism for reducing the amount of fat tissue under the influence of CLA is the

impact on reduction of the size and/or number of fat cells in the fat (*Pastoreli et al., 2005*). This can be achieved by inhibition of the enzyme lipoprotein lipase in adipocytes, inhibiting the activity of the enzyme of the sterol-CoA desaturase. Since the lipoprotein lipase is a key enzyme which regulates the deposition of fat in the body, its inhibition will induce the reduction of adipogenesis, which was confirmed for the effect of the trans-10, cis-12 isomers of CLA, but not for cis-9, trans-11 isomer (*Park et al., 1997*). Sterol-CoA desaturase is an enzyme which regulates the conversion of saturated to mono-unsaturated fatty acids, which are the main constituents of the subcutaneous fat tissue, so its inhibition can influence the reduction of the quantity of fat tissue (*Park et al., 2000; Lin et al., 2004*). As a consequence of the above, the addition of CLA in the diet of pigs can decrease the accessibility of fatty acids for the synthesis of triglycerides and reduce the amount of fat, as found by several authors (*Pastoreli et al., 2005*).

Feeding animals with CLA affects the change of fatty acid composition of fat in muscle and adipose tissue. On the basis of investigations that the CLA reduces the content of monounsaturated fatty acids (MUFA) in animal fats, *Evans et al. (2000)* have found that the trans-10, cis-12 CLA isomer is responsible for this decrease. However, the mechanism of action of CLA on reducing the MUFA and increasing the saturated fatty acids (SFA) has not been fully explained. *Lee et al. (1994)* assume that the effect of CLA on saturation of fatty acids manifests through the inhibition of the enzyme activity, Δ -9 sterol-CoA desaturase. This theory was confirmed by *Pariza et al. (2000)*.

Some of the effects of feeding pigs with CLA, which have been documented in scientific publications, are: improved feed conversion, reducing the thickness of subcutaneous fat, increased marbling of meat, increased firmness of body fat, improved meat color, change in the fatty acid composition of meat and reduced susceptibility to oxidation. However, all these positive effects are not confirmed in every experiment which included feeding pigs with CLA, because there are many factors that affect its efficiency.

The effect of CLA on quality of pig meat

CLA is thought to positively influence the quality of pork. Studies have found that the thickness of subcutaneous back fat decreases on average by about 1.2 mm, but the amount of intramuscular fat increases by about 7%, in pigs fed CLA (*Dunshee et al., 2005*). This increase of the content of intramuscular fat or marbling of the meat would mean that the feeding pig with CLA improves the sensory quality of the meat. However, most authors did not identify significant effect of CLA on sensory scores of tenderness, juiciness and flavor of roasted meat (*Thiel et al., 1998; Dugan et al., 1999; Thiel-Cooper et al., 2001; Wiegand et al.,*

2002; Larsen *et al.*, 2009), which are positively correlated with the content of intramuscular fat.

In several studies positive effects of feeding animals with CLA on the parameters of technological quality of meat are revealed. In this regard, water holding capacity of meat from pigs fed CLA is either enhanced (*O'Quinn et al.*, 1998; *Joo et al.*, 2002; *Szymczyk*, 2005) or remains unchanged (*Thiel-Cooper et al.*, 1999; *Wiegand et al.*, 1999).

Influence of feeding pigs with CLA on instrumental meat color differs between studies. *Wiegand et al.* (2001) report a significant increase in L* and a* and decrease in b* values of meat color from pigs fed CLA. On the other hand, some researchers did not determine the impact of CLA on the change of the meat color (*O'Quinn et al.*, 1998; *Dugan et al.*, 1999; *Martin et al.*, 2007; *Barnes et al.*, 2012).

As regards the pH value, *Wiegand et al.* (2001) report that pigs fed with the addition of CLA had a lower pH value of the meat, as measured 3 hours post-mortem, while the pH value 24 hours after slaughter was not different compared to the control. Other researchers suggest that CLA has no influence on the final pH value (*Thiel-Cooper et al.*, 1999; *Dugan et al.*, 1999; *Wiegand et al.*, 2001; *Eggert et al.*, 2001).

Feeding pigs with CLA affects a significant change in the fatty acid composition of fat and muscle tissue, which leads to the incorporation of CLA isomers in subcutaneous fat tissue and to a lesser extent in intramuscular fat (*Eggert et al.*, 2001; *Ramsay et al.*, 2001; *Wiegand et al.*, 2001; *Joo et al.*, 2002). The amount of isomers that is absorbed on this occasion and incorporated into the tissues depends on their concentration in food (*Ostrowska et al.*, 2003). The effect of dietary CLA on fatty acid composition of fat and muscle tissue is largely dependent on the type of fats/oils that are used as food and that are replaced by CLA, because some differences in the composition of fatty acids derive from the differences in the composition of the dietary fat/oil used in control and experimental groups. In this regard, *Dunshee et al.* (2005) suggest that in the interpretation of the effects of dietary CLA on fat composition, the fatty acid composition of the food used in fattening should be taken into account. Despite this fact, there are certain changes in the fatty acid composition of pork, attributed to the functioning of the CLA. For example, *Ostrowska et al.* (2003) found that the proportion of palmitic (16:0) and palmitoleic (16:1) acids, in the intramuscular and subcutaneous fat tissue increase linearly with an increase in the level of CLA in food. Some researchers indicate a considerable reduction in the shares of oleic (18:1), linoleic (18:2), linolenic (18:3), and arachidonic (20:4) acids in pigs fed CLA (*Chin et al.*, 1994; *Ramsay et al.*, 2001; *Joo et al.*, 2002; *Sun et al.*, 2004). As a result of the aforesaid changes in the fatty acid composition, there is an increase in the share of SFA, and reduction in the share of unsaturated fatty acids (*Joo et al.*,

2002; Wiegand *et al.*, 2002; Tishendorf *et al.*, 2002; Szymczyk, 2005; Lauridsen *et al.*, 2005), also an increase in the iodine number/value of the fat (Eggert *et al.*, 2001), and thus increase of the hardness of fat tissue and improvement of its technological characteristics (Eggert *et al.*, 2001; Thiel-Cooper *et al.*, 2001).

Table 1. Summary of published reports on the effect of CLA on meat quality of pigs

Reference	% CLA	Breed ¹	Sex	Weight range (kg)	IM ²	Fatty acid		Colour (CIE) ⁵	WBC ⁶	pH
						SFA ³	UFA ⁴			
<i>Dugan et al. (1997)</i>	2	L x VJ	♀ i ♂	62-106	↑	-	-	nc	nc	nc
<i>Ostrowska et al. (1999)</i>	0,7-5,5	VJ x L	♀	57-107	-	-	-	-	-	-
<i>Thiel-Cooper et al. (2001)</i>	0,07-0,6	(JxL)x(DxH)	♂	26-114	↓	↑	↓	-	-	-
<i>Ramsay et al. (2001)</i>	0,17-1,34	J x L	♀ i ♂	20-55	-	↑	↓	-	-	-
<i>Wiegand et al. (2002)</i>	0,75	(JxL)x(DxH)	♂	28-115	↑	↑	-	↑ b*	-	-
<i>Joo et al. (2002)</i>	1-5	L x VJ x D	♀	64-99	↑	↑	↓	-	↑	-
<i>Szymczyk (2005)</i>	0,1-0,6	L x VJ	♀ i ♂	61-108	↓	↑	↓	↑ L*	↑	nc
<i>Lauridsen et al. (2005)</i>	0,3	L x J x D	♂	40-130	nc	↑	↓	-	nc	nc
<i>Martin et al. (2007)</i>	0,6-1,2	VJ	♀	70-107	↑	↑	↓	nc	-	nc
<i>Barnes et al. (2012)</i>	0,6	PIC 380	♂	53-103	-	↑	↓	nc	-	-

nc – No change; ↑ – Increase; ↓ – Decrease.

¹ LW – Large White; Y – Yorkshire; L – Landrace; D – Duroc; H – Hampshire;

² IM – Intramuscular fat; ³ SFA – Saturated fatty acids; ⁴ UFA – Unsaturated fatty acids;

⁵ CIE – CIEL*a*b* system (CIE, 1976); ⁶ WBC – Water binding capacity.

In examining the impact of CLA in the diet of pigs on the quality of loin *M. longissimus dorsi* muscle, *Joo et al. (2002)* report that the concentration of TBARS (tiobarbituric acid reactive substances) is lower in pigs fed CLA after 7 days of storage at 4°C. Those authors suggest that the difference in oxidative stability of fats may be the result of increasing the share of total saturated fatty acids and decreasing the share of polyunsaturated fatty acids. *Ha et al. (1990)* have found that the CLA acts as an antioxidant, of larger capacity than α -tocopherol, and compared it with the β -hydroxytoluene (BHT). However, recent research has put into question the antioxidant effect of CLA, because of its susceptibility to auto-oxidation (*Yang et al., 2000*).

Table 1 shows the summarized published data on the effects of feeding pigs with CLA on the quality of the resulting meat.

Conclusion

Conjugated linoleic acid (CLA) is a mixture of geometric isomers of $cis\Delta^9$, $cis\Delta^{12}$ – octadecadienoic (linoleic acid - 18:2 n-6) acid, for which many positive effects were identified, such as anticancer effect, improved feed conversion and reduction of the amount of fat and increase of the share of muscle tissue in the body of animals.

Feeding pigs with the addition of CLA is gaining importance in recent years, because it affects the reduction of thickness and the share of subcutaneous fat and increases the lean meat/meat yield in carcass without negative impacts on the quality of the resulting meat. Increasing the content of saturated fatty acids in the fat and muscle tissue improves technological characteristics and oxidative stability of meat. In addition, due to the incorporation of CLA isomers in the tissues of animals and its health benefits, this type of nutrition can be a method for obtaining the pork with the features "of functional foods." However, all these positive effects have not been confirmed in every experiment that included feeding pigs with CLA, because there are many factors that affect its efficiency.

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Uticaj ishrane svinja sa konjugovanom linolnom kiselinom na kvalitet mesa

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Rezime

Konjugovana linolna kiselina (CLA) predstavlja smešu geometrijskih izomera linolne kiseline. Istraživanja su utvrdila da ishrana životinja sa dodatkom CLA ima razne pozitivne efekte, kao što su poboljšana konverzija hrane, smanjenje količine masnog tkiva i poboljšanje kvaliteta dobijenog mesa. Razlog velikog interesovanja za ovo jedinjenje su i njeni potencijalni zdravstveni efekti na ljude, kao što su antikancerogeno dejstvo, poboljšanje rada imunog sistema, smanjenje nastanka kardiovaskularnih bolesti i sl.

Ishrana svinja sa dodatkom CLA dobija sve veći značaj poslednjih godina, jer utiče na smanjenje debljine i udela potkožnog masnog tkiva i na povećanje mesnatosti trupa, bez negativnih uticaja na kvalitet dobijenog mesa. Utvrđeno je povećanje sadržaja intramuskularne masti, tj. marmoriranosti mesa i promena masno-kiselinskog sastava mesa. Povećanje sadržaja zasićenih masnih kiselina u masnom i mišićnom tkivu poboljšava tehnološke karakteristike i oksidativnu stabilnost mesa. Dodatno, zbog inkorporacije CLA izomera u tkiva životinja i njenih zdravstvenih koristi, ovakva vrsta ishrane može biti i metod za dobijanje svinjskog mesa sa osobinama "funkcionalne hrane". Međutim, svi ovi pozitivni efekti nisu potvrđeni u svakom ogledu ishrane svinja sa CLA, jer postoje više faktora koji utiču na njenu efikasnost.

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