Dietary Conjugated Linoleic Acid Positively Affects Immunologic Variables in Lactating Sows and Piglets[1,2]

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ABSTRACT We studied the effects of conjugated linoleic acid (CLA) on metabolic and immunologic variables in lactating sows and piglets. Gestating sows ($n = 16$) were assigned to 1 of 2 weight- and parity-matched groups supplemented with 0% (C) or 0.5% (T) of a CLA preparation containing 50% CLA isomers. Supplementation started in late pregnancy and continued throughout lactation. At weaning, 80 piglets, half from each group of sows, were assigned to 0% CLA (C) or 0.5% CLA (T). Thus, there were four groups of 20 piglets: C-C, C-T, T-T, and T-C. Body weight and the number of piglets per litter at birth and weaning, and the chemical composition of colostrum did not differ among the groups. CLA affected the fatty acid composition of colostrum fat; palmitoleic and γ-linolenic acid were significantly lower compared with controls, whereas eicosenoic and eicosatrienoic acids were significantly higher. Feeding CLA increased ($P < 0.05$) colostrum IgG in sows. Sows fed CLA had higher ($P < 0.05$) serum leptin, IgG, and lysozyme. Nursing piglets from CLA-fed sows had significantly higher ($P < 0.01$) serum lysozyme and IgG. Consumption of CLA did not affect postweaning growth. Postweaning piglets fed CLA (T-T, C-T) had a higher IgG titer at 25 d ($P < 0.05$) and 35 d ($P < 0.01$) after weaning. Serum lysozyme was also higher at 25 d ($P < 0.05$) in CLA-fed piglets (T-T, C-T). At 35 d, serum α-1 acylglicoprotein was lower ($P < 0.05$) in piglets fed CLA. Dietary CLA had a positive effect on immunologic variables in lactating sows and piglets. J. Nutr. 134: 817–824, 2004.

KEY WORDS: • *conjugated linoleic acid* • *immune response* • *sows* • *piglets*

Conjugated linoleic acid (CLA)[4] is the collective name for a group of geometric and positional isomers of linoleic acid (18:2) in which the double bonds are separated by a single carbon-carbon bond instead of a methylene group.

CLA has been shown to have many favorable biological effects, as recently reviewed by Pariza et al. (1). Numerous direct effects have been attributed to CLA on enzymatic activities involved in lipid metabolism, producing changes in milk fatty acid composition and markedly depressing the total content and yield of milk fat (2). In addition, dietary CLA isomers are excreted in colostrum and milk and are therefore available to the suckling piglets (3).

Numerous studies also reported that CLA had immune-modulating effects in several experimental models (4–6). We previously reported that dietary CLA enhanced serum IgG and lysozyme production, but had no effect on serum α-1 acylglycoprotein (AGP) levels in weaned piglets fed CLA for 4 wk (5). This finding is in agreement with the work of Sugano et al. (7) in which rats fed diets supplemented with 0.5 and 1% CLA had increased serum IgG. In addition Rooke et al. (8) suggested that IgG synthesis by piglets is positively correlated with the amount of maternal IgG absorbed, thus reinforcing the importance of an adequate IgG intake from colostrum. Krakowski et al. (9) reported that high immunologic values of colostrum determine the immunity of piglets not only in the suckling period, but also after weaning.

CLA was also reported to reduce the catabolic responses induced by immune stimulation, mediated by cytokines and regulated by prostaglandin E_2 (PGE_2) synthesis, without adversely affecting immune function (10). Therefore, reducing the immune response may provide more available energy for anabolic processes in skeletal muscle.

However, studies addressing the relationship between CLA supplementation and immune response are still lacking in lactating sow and piglets. Thus, the aims of the present study were to determine whether dietary CLA supplementation during late gestation and lactation influences reproductive per-

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[4] Abbreviations used: AGP, α-1 acylglicoprotein; C, control; CC, control sows-control piglets; CLA, conjugated linoleic acid; CT, control sows-treated piglets; EFA, essential fatty acids; IGF-1, insulin-like growth factor-1; IL, interleukin; MUFA, monounsaturated fatty acid; NEFA, nonesterified fatty acid; PGE_2, prostaglandin E_2; PPAR, peroxisome proliferator-activated receptor; PW, postweaning diet period; T, treated; TC, treated sows-control piglets; TNF, tumor necrosis factor; TT, treated sows-treated piglets.

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formance, immune, metabolic, and hormonal variables in sows, and to assess the efficacy of CLA supplementation for improving growth and immune variables in piglets.

MATERIALS AND METHODS

Animals and diets. Landrace × Large White sows (n = 16) at 8 d before parturition were assigned to 1 of 2 groups matched for parity [control (C) = 3.83; treated (T) = 4.02] and body weight (mean 228 kg). One group (T) received a diet supplemented with 0.5% CLA (CLA ONE powder, PharmaNutrients). The control group (C) received an isoenergetic diet with 0.5% soybean oil as a replacement for CLA. The two diets were formulated to meet NRC requirements for nutrient standards (11). The CLA preparation contained 50% CLA isomers (equal quantities of *cis*-9, *trans*-11 and *trans*-10, *cis*-12 isomers) in the form of FFA. Upon farrowing, sows were fed their treatment diet twice daily (0800 and 1600 h). Sows were initially fed 1.5 kg, and this was increased daily by ~0.5 kg of feed until d 7 postpartum, depending on sows feed consumption and recovery after parturition. After d 7 postpartum, sows had free access to their diets until weaning. The composition of the lactation diets and the fatty acid profile of the lipid content of the diets are shown in **Tables 1** and **2**.

Sow weight was recorded at entry into the farrowing room and at the time of weaning. Body condition scores of sows were recorded at parturition and at weaning. The numbers of pigs born alive and stillborn were recorded. Daily feed intake was recorded for each sow.

At farrowing, litters were equalized within dietary treatments to the same number of piglets per litter (mean 8 piglets). Prestarter feed was freely available to the nursing piglets from 10 d of age until weaning. Litter weights were determined within 48 h of birth and at weaning. The prestarter feed was fortified to meet or exceed NRC requirements (11) for all nutrients and contained no antibiotic agent.

At 21 d of age, a total of 80 piglets, half females and half castrated males (6.5 kg live weight), were moved to cages and reared in an environmentally controlled room. All piglets remained in the same treatment group defined by their dam; they were then subdivided into two groups of 20 piglets each so that a total of 4 experimental groups resulted: CC (control sows-control piglets), CT (control sows-treated piglets), TC (treated sows-control piglets), and TT (treated sows-treated piglets).

Piglets from CT and TT groups were fed a starter diet supplemented with 0.5% CLA. No CLA supplementation was used for the starter diet fed to CC and TC piglets. The basal starter diet was fortified to meet or exceed NRC requirements (11) for all nutrients (14.23 MJ/kg, crude protein 21%, lysine 1.4% as fed). Antibiotics and other growth-promoting agents were not included in the diets. Piglets live weight were recorded at 15, 25, and 35 d post weaning. All procedures involving animals were in accordance with European Community guidelines (n. 86/609/CEE) and approved by the Italian Ministry of Health (L. n. 116/92).

Collection and analysis of colostrum samples. During parturition, ~30–40 mL of colostrum was collected from the functional glands of each sow after injection of 2 mL of oxytocin (Pitocina). The colostrum samples were immediately frozen at −20°C for later analysis. The colostrum samples were analyzed to determine chemical composition, fatty acid composition, and total IgG.

To determine dry matter content, colostrum samples were freeze-dried for 24 h. The ether extract was determined by diethyl ether extraction in a Soxhlet apparatus. Crude protein was determined by the Kjeldahl method (N × 6.25).

To determine the fatty acid composition, lipids were extracted with chloroform:methanol (2:1) as described by Folch et al. (12). The FFA were methylated with boron trifluoride:methanol according to Banni et al. (13). The methyl esters were separated on a Carlo Erba Instruments chromatograph (GC 6000 Vega series 2) equipped with a fused silica capillary column (0.25 mm i.d.× 100 m) containing a cyanopropyl siloxane (CP-Sil 88) stationary phase (0.25 μm film

TABLE 1

Ingredients and composition of sow lactation diets[1]

Ingredient	Control	CLA
	g/kg	
Corn meal	380	
Soybean meal	162	
Soft wheat bran	250	
Barley	160	
Calcium carbonate	15	
Dicalcium phosphate	8	
Vitamin mineral premix[2]	5	
Sodium cloride	4	
L-Lysine HCl	1	
Soybean oil[3]	15	10
CLA powder[4]	—	5

[1] The diets were formulated to contain 13 MJ digestible energy, 17% crude protein, 0.90% lysine, 0.62% methionine + cysteine, 0.66% threonine, and 0.22% tryptophan.

[2] Formulation of the complete vitamin premix (/kg diet): 1.2 mg all-*trans* retinol; 0.006 mg cholecalciferol; 9.9 mg vitamin E; 2.8 mg riboflavin; 1.3 mg vitamin B-6; 0.015 mg vitamin B-12; 0.2 mg menadione; 102 mg pantothenic acid; 10 mg niacin; 0.48 mg folic acid; 84 mg Fe as Fe-sulfate; 0.56 mg I as $Ca(IO)_3$; 0.2 mg Se as Na_2Se; 9.2 mg Cu as $CuSO_4$; 81 mg Zn as ZnO_2; 2.5 mg Mn as MnO_2; 196 g choline; and 0.99 mg biotin.

[3] The fatty acid composition of the soybean oil (g/100 g total fatty acids) was: palmitic acid, 12 g; stearic acid, 4.40 g; oleic acid, 22 g; linoleic acid, 54 g; and linolenic acid, 7.60 g.

[4] The fatty acid composition of CLA preparation (g/100 g total fatty acids) was: myristic acid, 0.35 g; palmitic acid, 3.83 g; stearic acid, 1.15 g; elaidic acid, 28.30 g; oleic acid, 0.52 g; linoleic acid, 1.75 g; linolenic acid, 0.10 g; CLA *cis* 9-*trans* 11, 32.52 g; CLA *trans* 10-*cis* 12, 31.39 g.

TABLE 2

Lipid concentration and fatty acid composition of lactation diets[1,2]

	Control	CLA
Lipid, *g/kg diet*	45.4	44.4
	g/100 g total fatty acids	
14:0	1.30	1.15
16:0	15.1	14.5
18:0	4.65	4.25
20:0	0.41	0.25
22:0	0.43	0.29
Σ SFA	21.89	20.44
16:1	1.97	1.64
18:1(n-9)	29.9	28.2
18:1(n-7)	2.90	2.22
20:1(n-9)	0.39	0.56
22:1(n-9)	0.38	0.45
Σ MUFA	35.54	33.07
18:2(n-6*t*)	0.15	0.10
18:2(n-6*c*)	38.38	36.09
18:3(n-3)	2.45	2.02
18:3(n-6)	0.56	0.42
20:3(n-6)	0.22	0.38
Σ PUFA	41.76	39.01
CLA-isomers[3]		
cis-9, *trans*-11	ND[3]	3.34
trans-10, *cis*-12	ND	3.17

[1] Only fatty acids that accounted for 0.1 g/100 g of total are presented.

[2] Fatty acids are designated by the number of carbon atoms followed by the number of double bonds. The position of the first double bond relative to the methyl (n) end of the molecule is also indicated.

[3] ND, not detected.

thickness) (Chrompack). Temperature conditions for the oven were as follows: 5 min at 140°C, then increased at 4°C/min from 140 to 200°C, held at this for 20 min and increased at 4°C/min to 225°C and held at this temperature for 30 min. The injector temperature was 270°C and the detector temperature, 300°C. The injection volume was 0.4–0.8 μL. The methyl esters were identified by comparison with the retention times of pure standards (Supelco 37 Component FAME mix). The CLA isomers, *cis*-9, *trans*-11 and *trans*-10, *cis*-12, were identified using CLA standards (Matreya). Colostrum IgG was determined using the method described below for serum IgG.

Collection of serum samples. Blood samples were collected before the morning feeding by anterior vena cava puncture of the sows at d 2, 10, and 20 of lactation. Six piglets from each litter, half males and half females, homogeneous for live weight, were selected and blood collected by anterior vena cava puncture at 2, 10, and 20 d of lactation.

During the postweaning phase, a total of 60 piglets (15 from each group) from the animals sampled before weaning were punctured by anterior vena cava puncture at d 15, 25, and 35 after weaning. The blood samples were collected into vacutainers, immediately placed on ice, and subsequently centrifuged at 3500 × *g* for 15 min. Serum was partitioned into aliquots and stored at −20°C for later analysis.

Analyses

Blood metabolites in sows. Metabolites were determined by enzymatic spectrophotometric assays [glucose, Sigma; nonessential fatty acids (NEFA), Wako Chemicals] and hormones by RIA [insulin, Porcine insulin, Linco Research; thyroxin, T4 Mab, ICN Pharmaceuticals; insulin-like growth factor-1 (IGF-1), Diagnostic System Laboratories]. Because a specific procedure for swine leptin is not available, we used a commercially available multispecies RIA using antibody against human leptin (Multispecies, Linco Research); values obtained were expressed as human equivalent immunoreactive leptin. The concentrations of immunoreactive leptin, found by serial dilution (25–100 mL) of sows' plasma, were similar to those of the supplied reference curve of human leptin concentrations.

IgG in serum and colostrum. IgG was determined by the radial immunodiffusion method of Mancini et al. (14) using a commercial kit (Bethyl Laboratories).

Lysozyme in serum. The lysozyme assay employed *Micrococcus lysodeikticus* cells as substrate for lysozyme using the method of Osserman and Lawlor (15). Briefly, 25 mg of dried *Micrococcus lysodeikticus* (M-3770; Sigma), was mixed with 0.5 mL of physiologic saline and 50 mL of agarose gel. The mixture was poured into dishes containing 44 wells filled with 10 μL of standard lysozyme solution (L-6876; Sigma) at concentrations 10, 5, 2.5, 1.25, or 0.625 mg/L. Residual wells were filled with 50 μL of serum and incubated in dishes at 24°C for 18 h. Lysozyme concentrations were determined by comparing clear zone diameters of samples with those produced by the standard lysozyme solutions.

α-1 Acylglycoprotein (AGP). AGP was determined by the radial immunodiffusion method described by Mancini et al. (14) using a commercial kit including specific antibody (Cardiotech Services).

Statistical analysis. Statistical analysis of the data were performed with the General Linear Model (GLM) procedure of SPSS (SPSS/PC+ Statistics 11.0.), and the residual error was used to test the main effect of dietary treatment. Data are presented as means ± pooled SEM. Immunoglobulin, lysozyme, AGP, hormones, and serum metabolites of sows and nursery piglets were assessed by repeated-measures ANOVA. Growth performances and immunologic data of postweaning piglets were analyzed as 2 × 2 × 2 randomized ANOVA with sex, lactation (L), and postweaning diet period (PW) as effects. The individual pig values were considered to be the experimental unit of all response variables. Differences between means were tested by the Student-Newman-Keul's *t* test. Differences were considered significant at $P < 0.05$.

RESULTS

Performance of sows and piglets. The sow groups did not differ in terms of feed intake, weight loss, or body condition scores at parturition and at the end of the lactating period (**Table 3**). The weights of individual piglets at 2 and 10 d after birth and at weaning did not differ (Table 3). The body weights of postweaning piglets were not affected by the diet fed to the dams during lactation (L) or by the PW. The sexes also did not differ (**Table 4**).

Colostrum. CLA dietary supplementation during late gestation did not affect the overall chemical composition of colostrum. The fatty acid composition of colostrum was affected by CLA supplementation (**Table 5**). As expected, the proportion of CLA isomers was higher in colostrum from treated sows than from controls ($P < 0.01$). CLA supplementation increased the concentration of eicosenoic acid ($P < 0.05$) and eicosatrienoic acid ($P < 0.02$). Palmitoleic acid ($P < 0.01$) and γ-linolenic acid ($P < 0.05$) were lower in the colostrum of treated sows. The ratio of palmitoleic acid to palmitic acid was lower in the T than in the C group ($P < 0.05$).

Blood metabolites of sows. Serum leptin was increased in CLA-fed sows ($P < 0.05$) (**Table 6**). The other serum variables investigated were not influenced by dietary CLA supplementation. There were significant differences in leptin ($P < 0.01$), NEFA ($P < 0.01$), and IGF-1($P < 0.01$) on different sampling days (Table 6). Thyroxin, insulin, and glucose were not affected by CLA supplementation or by sampling day. A significant treatment × sampling day interaction for leptin and glucose was observed ($P < 0.05$).

Immunologic variables of sows and suckling piglets. Colostrum IgG levels were significantly ($P < 0.05$) higher in T sows than C sows (**Table 7**). Serum IgG concentrations in lactating sows (Table 7) were greater ($P < 0.01$) in those fed CLA than in control sows. Lysozyme serum concentrations were greater ($P < 0.05$) in CLA-fed than control sows. No differences in AGP levels were found between dietary treatments and sampling days. Serum IgG and lysozyme increased significantly during the lactation period ($P < 0.01$).

Piglets from treated sows had higher serum concentrations of IgG ($P < 0.001$) and lysozyme ($P < 0.01$) than controls during the suckling period (**Table 8**). AGP serum concentrations did not differ. Serum IgG and AGP decreased during the lactation period ($P < 0.001$), whereas serum lysozyme in-

TABLE 3

Effect of CLA supplementation during gestation and lactation on the performance of sows and piglets[1]

	Treatment		
Item	Control	CLA	SEM
Sows' weight, *kg*			
At farrowing (−8 d)	224.5	233.28	13.08
At weaning (21 d)	201.25	202.28	12.49
Weight loss, *kg*	23.25	31.00	4.78
Feed intake, *kg/d*	3.85	3.63	0.23
Piglets/litter, *n*			
Total born	8.25	9.15	1.24
Born alive	8.12	8.57	1.27
Stillborn	0.12	0.57	0.21
Weaned	7.5	7.7	0.31
Piglets' weight, *kg*			
At birth	1.79	1.77	0.12
10 d	3.47	3.25	0.19
At weaning	6.17	5.33	0.34

[1] Values are means ± pooled SEM, $n = 8$.

TABLE 4

Body weights of piglets born to CLA-treated and control sows and fed diets with (T-T, C-T) and without (C-C, T-C) CLA supplementation in the postweaning period[1]

	Diet					*P*-value[2]		
Item	T-T	T-C	C-C	C-T	SEM	L	PW	L × PW
Piglets weight, *kg*								
At weaning	6.03	6.61	6.44	6.97	0.33	0.157	0.929	0.111
After weaning								
15 d	8.69	9.27	8.03	9.42	0.50	0.709	0.461	0.104
25 d	11.55	11.92	12.93	13.73	0.71	0.504	0.696	0.396
35 d	16.77	16.54	17.33	18.78	1.00	0.329	0.926	0.815

[1] Values are means ± pooled SEM, $n = 20$.
[2] Effects of lactation (L) and the PW or the interaction between lactation and the starter diet period (L × PW).

creased ($P < 0.001$) with the age of the piglets. The sexes did not differ.

Immunologic variables of postweaning piglets. Serum IgG, lysozyme, and AGP concentrations in postweaning piglets were not significantly affected by the diet fed to the dams during lactation (L) (**Table 9**). However, irrespective of the dietary fat supplied to the dams during the period of lactation, piglets fed the diet supplemented with CLA (T-T; C-T) had higher IgG concentration at 25 ($P < 0.05$) and 35 ($P < 0.01$) d postweaning than controls (C-C; T-C). Lysozyme levels were higher in treated piglets (T-T; C-T) at 25 d postweaning ($P < 0.05$) and AGP serum concentrations were lower in piglets fed CLA (T-T; C-T) at 35 ($P < 0.05$) d postweaning compared with controls (C-C; T-C). The sexes did not differ.

TABLE 5

Chemical composition and fatty acid composition of colostrum of sows fed the control and CLA-supplemented diets[1]

	Control	CLA	SEM
Chemical composition, *g/100 g*			
Dry matter	27.19	27.61	0.163
Crude Protein	16.50	15.92	0.734
Ether extract	6.25	5.84	0.347
Fatty acid composition, *g/100 g*			
14:0	4.08	3.66	0.352
15:0	0.35	0.50	0.190
16:0	16.70	16.85	0.927
17:0	0.41	0.45	0.066
18:0	6.30	6.34	0.267
21:0	0.21	0.24	0.043
22:0	0.16	0.55	0.079
Σ SFA	27.93	28.40	0.996
14:1	0.43	0.53	0.196
16:1	3.10^{A}	2.07^{B}	0.209
17:1	0.38	0.27	0.011
18:1(n-7)	1.87	1.65	0.260
18:1(n-9)	37.01	36.16	1.076
20:1	0.50	0.57	0.118
22:1(n-9)	0.42^{a}	0.85^{b}	0.109
Σ MUFA	43.30	41.98	1.326
18:2(n-6c)	22.89	21.10	1.252
18:3(n-6)	0.24^{a}	0.10^{b}	0.033
18:3(n-3)	1.90	1.45	0.163
20:3(n-6)	0.33^{a}	0.91^{b}	0.118
20:4(n-6)	2.04	2.11	0.282
CLA isomers			
cis-9, *trans*-11	ND^{A}	0.43^{B}	0.080
trans-10, *cis*-12	ND^{A}	0.42^{B}	0.102
Σ PUFA	27.95	25.55	1.162
16:1/16:0	0.18^{a}	0.13^{b}	0.015
18:1(n-9)/18:0	5.91	5.78	0.124

[1] Values are means ± pooled SEM, $n = 8$; those in a row with superscripts without a common letter differ: lowercase, $P < 0.05$; uppercase, $P < 0.01$. ND, not detected.

DISCUSSION

Performance. Published findings on the growth performance effects of feeding CLA to piglets agree with our results. Bee (16) and Harrel et al. (17) did not show any improvement in growth rate of neonatal piglets born to dams fed supplemental CLA. The milk yield might have not been decreased by CLA supplementation because the growth performance of those litters depended mainly on the sow's milk production.

The growth performance of piglets after weaning was unaffected by dietary CLA supplementation, which agrees with the observations of Ostrowska et al. (18) and Greene et al. (19) in pigs fed CLA. In contrast to our results, Bee (16) reported that irrespective of the starter diet fed in the 35-d postweaning period, pigs born to and reared by sows fed 2% CLA had greater feed intake, daily weight gain, and final body and warm carcass weights than pigs reared by sows fed the control diet. These differences might be due to the high level of CLA supplementation used by Bee (16).

Composition of colostrum. CLA supplementation was reported to profoundly affect lipid metabolism (20) and to increase the CLA content of milk fat in dairy cows, thus altering the fatty acid composition of the milk and lowering the yield of milk fat (2). Studies on lactating sows also found reductions in total milk fat at weaning. Poulos et al. (21) and Harrel et al. (17) found that milk fat was lowered by 17 and 36% after the addition of 0.5 and 1% CLA to the diet, respectively, and supplemented from d 40 of gestation to weaning, or from d 5 postfarrowing to weaning, respectively.

In the present study, the fat content of colostrum from CLA-fed sows was close to 7% lower than in colostrum from control sows ($P = 0.76$). The lack of effect of CLA on the overall fat content of colostrum in our study might be explained by the short length of CLA supplementation (8 d) before farrowing and the low level of supplementation.

We did find, however, that CLA supplementation affected the fatty acid composition of sows' colostrum. This is consistent with the known effects of dietary CLA on the fatty acid composition of fat, with increased SFA, decreased monounsaturated fatty acids (MUFA), and increased total CLA iso-

TABLE 6

Hormones and metabolites at 2, 10, and 20 d postpartum in serum of sows fed control and CLA-supplemented diets[1]

	Lactation day				*P*-values[2]		
	2	10	20	SEM	Tr	T	T × Tr
Leptin, *μg/L*							
Control	2.08	2.27	1.34	0.153	0.051	0.004	0.055
CLA	2.27	2.55	2.25	0.191			
Insulin, *μmol/L*							
Control	15.35	40.39	35.84	5.48	0.116	0.111	0.125
CLA	21.35	39.37	27.41	6.67			
Glucose, *mmol/L*							
Control	3.31	2.88	3.86	0.181	0.101	0.070	0.029
CLA	3.17	2.91	2.84	0.199			
NEFA, *μmol/L*							
Control	327.96	121.32	178.06	40.43	0.679	0.006	0.650
CLA	349.60	186.87	136.21	45.52			
Thyroxine, *μg/L*							
Control	16.64	23.16	24.40	2.50	0.413	0.253	0.167
CLA	24.05	25.13	22.49	2.78			
IGF-1, *μg/L*							
Control	86.38	190.44	153.77	28.51	0.546	0.001	0.779
CLA	74.20	155.80	135.08	23.70			

[1] Values are means ± pooled SEM, $n = 8$.
[2] Effect of dietary treatment (Tr), sampling days (T), or that of the interaction between sampling days and treatment (T × Tr).

mers. Several studies reported similar changes in both the adipose tissue and milk of pigs fed CLA (22,23), whereas Bee (3) reported that dietary CLA supplementation during gestation significantly reduced MUFA and increased the SFA content in colostrum. The length of dietary CLA supplementation in our study was likely too short to cause an overall reduction in MUFA content and an increase in the SFA content of colostrum.

As expected, we detected CLA only in the colostrum of treated sows, in agreement with Bee (3). In addition CLA supplementation lowered the proportion of palmitoleic acid (16:1) likely by lowering the activity of Δ-9 desaturase. Bretillon et al. (24) and Lee et al. (25) reported that both the activity and gene expression of hepatic Δ-9 desaturase are inhibited by dietary CLA. In vitro studies showed that *trans*-10, *cis*-12 CLA is responsible for these effects (26). We also found that CLA supplementation significantly lowered the γ-linolenic acid [18:3(n-6)] content of colostrum, and this is consistent with the reports of Belury and Kempa-Steczko (27) that CLA may compete with linoleic acid for Δ-6 desaturase. The Δ-6 desaturation of linoleic acid to γ-linolenic acid is the rate-limiting step in the biosynthesis of (n-6) PUFA. Bretillon et al. (24) also observed a reduction in the activity of Δ-6 desaturase in rat liver microsomes after dietary supplementation of CLA.

The quality of dietary lipid supply in early childhood as a major determinant of growth and infant development, particularly essential fatty acids (EFA), is indispensable for cells, plasma membrane synthesis, and regulators of numerous cell and tissue functions (28). In the present study, CLA supplementation did not affect the percentage of EFA in colostrum, which did not differ between the two groups. Moreover, the

TABLE 7

IgG levels in colostrum and immunological variables of serum of sows fed control and CLA-supplemented diets[1]

	Lactation day					*P*-values[2]		
	0	2	10	20	SEM	Tr	T	T × Tr
Colostrum IgG, *g/L*								
Control	33.59				3.91	0.013	—	—
CLA	49.44							
Serum IgG, *g/L*								
Control	—	10.78	14.98	17.41	1.45	0.005	0.001	0.297
CLA	—	20.85	21.33	25.06	1.59			
Serum lysozyme, *mg/L*								
Control	—	0.996	1.398	1.462	0.19	0.025	0.002	0.462
CLA	—	1.866	2.013	2.256	0.21			
Serum AGP, *mg/L*								
Control	—	258.33	293.08	305.34	19.51	0.593	0.090	0.683
CLA	—	282.56	287.71	334.63	21.38			

[1] Values are means ± pooled SEM, $n = 8$.
[2] Effect of dietary treatment (Tr), sampling days (T), or that of the interaction between sampling days and treatment (T × Tr).

TABLE 8

Immunological variables in the serum of suckling piglets whose dams were fed the control or CLA diet[1]

	Lactation day				*P*-values[2]		
	2	10	20	SEM	Tr	T	T × Tr
Serum IgG, *g/L*							
Control	22.92	12.28	9.28	1.27	0.001	0.001	0.614
CLA	29.40	20.86	16.22	1.15			
Serum lysozyme, *mg/L*							
Control	0.47	0.50	1.29	0.25	0.001	0.001	0.068
CLA	0.65	0.71	2.19	0.17			
Serum AGP, *mg/L*							
Control	7712	1818	1074	19.38	0.127	0.001	0.101
CLA	7358	1880	1136	16.20			

[1] Values are means ± pooled SEM, $n = 8$.
[2] Effect of dietary treatment (Tr), sampling days (T), or that of the interaction between sampling days and treatment (T × Tr).

beneficial effects of CLA, transferred to infants via colostrum and milk, may help decrease the risks of several diseases including breast cancer, diabetes, and obesity. Including in these effects are those influencing the immune system, nutrient partitioning, and growth modulation (29).

Blood metabolites. The peptide hormone, leptin, has been implicated in feed intake regulation, as well in reproductive and immune function (30). Because leptin is secreted mainly by adipocytes, circulating levels correlate directly with BMI in humans (31), with a positive correlation between circulating leptin levels and adiposity also reported in other species (32). In sows, Estienne et al. (33) found that serum leptin levels at farrowing were higher in sows with more backfat. At weaning, however, serum leptin levels were similar for sows that had been classified at farrowing as fat or thin, even though backfat thickness at weaning was still significantly different.

Because CLA is reported to reduce body fat content (34), a reduction in plasma leptin levels would be expected in CLA-supplemented animals (35). However, published findings on the effects of CLA dietary supplementation on plasma leptin are contradictory. Bouthergourd et al. (36) reported that 6 wk of CLA dietary supplementation did not affect plasma leptin in hamsters; and Corino et al. (37) reported that 0.5% of CLA supplementation significantly increased circulating leptin levels in rabbits.

The effects of dietary CLA on plasma leptin have not been extensively investigated in lactating sows. In our experiment, dietary CLA significantly increased serum leptin level at weaning ($P < 0.05$), and did not influence feed intake and body condition in lactating sows. Serum concentrations of IGF-1, insulin, glucose, thyroxin, and NEFA did not differ in relation to CLA supplementation. Data for IGF-1 agree with the findings of Poulos et al. (38) in weanling rats. By contrast. Li et al. (39) found that CLA increases serum IGF-1 in rats, suggesting the hypothesis that CLA modulates body mass through mechanisms involving the IGF system. In addition, Sisk et al. (40) reported that 0.5% CLA supplementation for 8 wk did not affect plasma insulin or NEFA in rats. Ostrowska et al. (41) found that dietary CLA increases plasma NEFA in pigs, with no effect on plasma glucose and insulin.

As expected, NEFA levels were highest in serum on d 2 of lactation for both groups ($P < 0.01$) compared with later days, indicating the intensive lipolysis and the negative energy balance characteristic of sows at this time (42). The IGF-1 levels on d 2 of lactation were lower than on d 10 and 20 (P

TABLE 9

Immunologic variables in the serum of postweaning piglets born to CLA-treated and control sows and fed diets with (T-T, C-T) and without (C-C, T-C) CLA supplementation in the postweaning period[1]

Time post-weaning, *d*	Diet				SEM	*P*-values[2]		
	T-T	T-C	C-C	C-T		L	PW	L × PW
IgG, *g/L*								
15	8.65	8.37	7.92	7.71	0.65	0.329	0.914	0.716
25	9.01	8.97	7.60	8.25	0.59	0.277	0.047	0.952
35	10.28	8.36	8.38	9.81	0.53	0.788	0.003	0.455
Lysozyme, *mg/L*								
15	6.49	5.44	4.83	5.96	0.83	0.293	0.097	0.924
25	6.23	3.52	3.07	4.62	0.81	0.386	0.029	0.810
35	3.01	2.22	2.66	3.58	0.55	0.427	0.089	0.658
AGP, *mg/L*								
15	37	1122	980	1023	95.85	0.588	0.719	0.442
25	45	1013	1115	958	102.44	0.549	0.248	0.743
35	88	967	851	779	93.21	0.159	0.040	0.208

[1] Values are means ± pooled SEM, = 15.
[2] Effect of lactation (L) and PW or that of the interaction between lactation and the PW (L × PW).

< 0.01) probably because of the reduced feed intake in the immediate postfarrowing period (43).

Immunologic variables. Eicosapentaenoic acid [20:5(n-3)] competes with arachidonic acid [20:4(n-6)] for cyclooxygenase, reducing PGE_2 production. It has been suggested that CLA are elongated and desaturated to 20:4 isomers and also compete with linoleic acid in the biosynthesis of arachidonic acid. Because arachidonic acid is the precursor of PGE_2, it is possible that CLA supplementation may decrease PGE_2 production (44). Recent studies indicated that dietary CLA increases plasma IgG in rats (7) and piglets (5).

In the present study, dietary CLA increased colostrum IgG. Bourne and Curtis (45) showed that essentially all IgG present in the colostrum of sows is derived from serum and that uptake into the mammary gland is mediated by specific epithelial cell receptors (46); in fact, we found that IgG was also significantly increased in the serum of treated sows. Furthermore, serum IgG levels in nursing piglets from CLA-supplemented sows were significantly higher than controls throughout the nursery period. Passive immunity, in the form of IgG absorbed from colostrum and milk, is vital during early life because it provides virtually the only means of resisting infections (47). However, colostrum IgG levels are highly variable among sows, even in the same unit, for a variety of reasons. The all-important initial dose of colostrum IgG received by a piglet depends on birth order and length of farrowing; those born late in the birth order may receive insufficient colostrum IgG (8). Furthermore, recent evidence suggests that IgG synthesis by piglets is directly related to the amount of maternal IgG received (8).

Serum IgG levels in postweaning piglets were higher ($P < 0.05$) at d 25 and 35 in CLA-fed piglets (C-T; T-T) compared with control piglets (C-C; T-C). Sugano et al. (7) found that IgG, IgA, and IgM levels were higher and IgE lower in the serum of rats supplemented with 0.5% and 1% CLA for 3 wk compared with controls. Corino et al. (5) also reported greater serum IgG in piglets fed 0.5 and 1% CLA for 28 d after weaning compared with controls. Weber et al. (48) reported that CLA supplementation increased serum antibodies against *Mycoplasma hyopneumoniae* in weanling piglets. Active immunologic responses are weak in piglets, and several weeks are required before they reach adulthood levels. Our study indicates that CLA supplementation to postweaning piglets might hasten the development of active immunity.

Lysozyme, present in external secretions, polymorphonuclear leukocytes, and macrophages, is highly active against gram-positive bacteria. The enzyme preferentially hydrolyzes the glycosidic β-1,4-link between *N*-acetylmuramic acid and *N*-acetylglucosamine present in the mucopeptide cell wall of various microorganisms. Lysozyme levels were higher in CLA-fed sows and CLA-fed piglets than controls during lactation; in the postweaning phase, piglets fed CLA had higher lysozyme levels ($P < 0.05$) only at d 25. These data agree with the findings of Corino et al. (5) in weaned piglets and is a further indication that CLA supplementation can enhance the antibacterial defenses of the body.

α-1 Acylglycoprotein is synthesized by the liver and released into the bloodstream in response to local inflammation, bacterial infection, or trauma as part of the acute phase of the inflammatory reaction (49). There is little specific information about this protein in the acute phase response of pigs. AGP gene expression is controlled by interleukin-1 (IL-1), IL-6, and tumor necrosis factor (TNF)-α (50). We found that dietary CLA did not affect serum AGP in sows or suckling piglets in agreement with the our previous observations (5). In postweaning piglets, AGP is lower ($P < 0.05$) in those fed CLA at 35 d. This lack of effect in the presence of enhanced IgG and lysozyme levels is probably due to reduced PGE_2 production as a result of interference in the synthesis of arachidonic acid from linoleic acid (27,51). As reported by Yu et al. (52), CLA is an activator of peroxisome proliferator-activated receptor (PPAR)-γ. A study by Jiang et al. (53) indicated that activation of PPAR-γ inhibits production of the pro-inflammatory cytokines TNF-α, IL-6 and interferon-γ. A CLA-induced decrease in PGE_2 synthesis would be expected to downregulate cytokine release (54) and consequentially AGP release.

In conclusion, dietary supplementation of 0.5% CLA to sows during late gestation and lactation increased the IgG concentration of colostrum and also affected its fatty acid composition. CLA supplementation to sows increased serum IgG and lysozyme levels and may therefore have a positive influence on immune response in sows and suckling piglets. We also found that serum IgG was increased by CLA supplementation to postweaning piglets. CLA seems promising, therefore, as an immunologic activator in young pigs that have not yet developed a more specific immune response repertoire. However, further investigations are required to confirm this action and to determine the most appropriate doses and times for supplementation.

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