Supplementary File

Effect of postruminal supply of linseed oil in dairy cows: 1. Production performance and fate of postruminally available α-linolenic acid

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Material and methods

Analyses of total mixed ration

After being pooled by period, samples were analysed for residual moisture (method 934.01; AOAC International, 2005), neutral detergent fibre (Ankom Technology method 6: NDF in feeds-filter bag technique for A200, with solutions as in Van Soest et al., 1991 and the inclusion of heat-stable α-amylase), acid detergent fibre (Ankom Technology, Fairport, NY; method 5: ADF in feeds filter bag technique for A200 with solutions as in method 973.18; AOAC International, 2005), crude protein (N × 6.25; method 990.03; AOAC International, 2005), and ash (method 942.05, AOAC International, 2005). The concentration of organic matter was calculated as the difference between 100 and the percentage of ash.

Plasma fatty acid analysis

Plasma samples were thawed at room temperature and equal volumes of pre- and postprandial samples were pooled by cow and by period before lipid determination. Plasma lipids were extracted three times with chloroform/methanol (2:1). The extract was filtered through a Whatman paper filter (#1; GE Healthcare, Chicago, IL, USA), evaporated to dryness, dissolved again in chloroform, and stored at -20°C until lipid-class separation. Total lipids were separated using the solid-phase extraction column method and then methylated according to Valckx *et al*. (2014). Fatty acid composition of each plasma lipid class was determined by gas chromatography (Agilent 7820; Agilent Technologies) using a HP-Innowax capillary column (30 m × 0.32 mm i.d. × 0.25 µm film; Agilent Technologies Canada Inc.) and a flame ionization detector. The split ratio was 150:1. The initial oven temperature was set to 185°C for 30 sec, then raised at a rate of 3°C/min to 220°C and maintained isotherm for 18 min. The inlet and detector temperatures were set to 240 and 250°C, respectively. Individual FA concentrations were expressed as a

percentage of total FA and quantified using, as internal standards, heneicosanoic acid (21:0) for free FA (FFA) and PL, and tritridecanoin (13:0/13:0/13:0) for TAG and CE. Fatty acid peaks were identified and quantified using either a quantitative mixture or pure methyl ester standards (Larodan Fine Chemicals, Malmö, Sweden; Sigma-Aldrich, Oakville, ON, Canada; Matreya LLC, Pleasant Gap, PA, USA; and Nu Chek Prep, Elysian, MN, USA). Total FA concentration was calculated as the sum of those quantified in TAG, PL, CE, and FFA.

References

Association of Official Analytical Chemists (2005) *Official Methods of Analysis*, 18th Edn. AOAC International, Arlington, VA, USA: AOAC International.

Van Soest PJ, Robertson FB and Lewis BA (1991) Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *Journal of Dairy Science* **74,** 3583-3597.

Valckx SDM, Arias-Alvarez M, De Pauw I, Fievez V, Vlaeminck B, Fransen E, Bols PEJ and Leroy JLMR (2014) Fatty acid composition of the follicular fluid of normal weight, overweight and obese women undergoing assisted reproductive treatment: a descriptive cross-sectional study. *Reproductive Biology and Endocrinology* **12,** 13.

Table S1. Ingredients and chemical composition of basal diet[1]

Item	g/kg of dry matter
Ingredient	
Corn silage	492
Grass silage	233
Ground corn	142
Soybean meal	55
Corn gluten meal	55
Mineral and vitamin premix[2]	23
Chemical composition	
Organic matter	935
Crude protein	153
Neutral detergent fibres	305
Acid detergent fibres	200
Fatty acids	
14:0	0.1
16:0	3.9
cis-9 16:1	0.1
18:0	0.6
cis-9 18:1 + *cis*-11 18:1	4.1
cis-9, *cis*-12 18:2	10.8
cis-9, *cis*-12, *cis*-15 18:3	2.2
Others[3]	0.4
Total	22.2

[1]Dry matter content = 42.8 g/100 g as fed.

[2]Contained 16.0% Ca, 3.4% P, 6.5% Na, 4.1% Mg, 30.9 mg/kg I, 2488 mg/kg Fe, 412 mg/kg Cu, 1375 mg/kg Mn, 2026 mg/kg Zn, 13.8 mg/kg Co, 330 mg/kg F, 17.2 mg/kg Se, 209.4 kIU vitamin A, 68.8 kIU vitamin D, 1187 IU vitamin E, on a dry matter basis.

[3]Unidentified chromatogram peaks.

Table S2. Dry matter intake, milk yield, and milk composition in dairy cows abomasally infused with increasing levels of linseed oil

	Linseed oil infusion, mL/d						
Item	0	75	150	300	600	SEM	Contrast[1]
Dry matter intake, kg/d							
Ration	26.1	25.6	25.1	24.7	23.5	1.1	L*
Ration + infused oil	26.1	25.6	25.2	24.9	24.0	1.1	L*
Energy intake, Mcal/d							
Ration	43.3	42.3	41.6	41.4	38.8	1.8	L*
Ration + infused oil	43.3	42.7	42.3	40.3	41.7	2.3	
Milk yield, kg/d							
Actual	31.9	34.3	32.3	33.9	29.8	2.4	
Fat-corrected[2]	32.8	34.2	31.7	32.9	29.3	2.5	L**
Energy-corrected[3]	32.2	33.7	31.2	32.5	28.7	2.3	L**Q*
Milk fat							
Concentration, g/100 g	4.19	3.99	3.87	3.81	3.87	0.13	L*Q**
Yield, g/d	1338	1365	1249	1291	1159	108	L**
Milk protein							
Concentration, g/100 g	3.41	3.39	3.35	3.35	3.29	0.12	
Yield, g/d	1085	1160	1079	1126	970	72	L*
Milk lactose							
Concentration, g/100 g	4.43	4.48	4.47	4.51	4.50	0.11	
Yield, g/d	1419	1542	1445	1532	1345	128	
Milk urea N, mg/dl	15.1	15.1	16.6	16.4	16.8	1.3	L*
Somatic cell count, ‘000/ml	96	78	94	119	124	33	

[1]L = linear and Q = quadratic effects of the level of linseed oil infusion. $^{*}P \leq 0.05$, $^{**}P \leq 0.01$.

[2][0.4 × (milk yield in kg/d)] + [15.0 × (fat yield in kg/d)] (Gaines, 1928).

[3]{[23.8 × (protein yield in kg/d)] + [38.9 × (fat yield in kg/d)] + [16.3 × (lactose yield in kg/d]}/3.14 (Madsen et al., 2008).

Table S3. Intake, milk secretion and apparent transfer efficiency from diet to milk fat of polyunsaturated fatty acids in dairy cows abomasally infused with increasing levels of linseed oil

	Linseed oil infusion, mL/d						
Fatty acid, g/d	0	75	150	300	600	SEM	Contrast[1]
Intake, g/d							
cis-9, *cis*-12 18:2	281.7	286.2	290.7	306.4	333.3	11.8	L**
cis-9, *cis*-12, *cis*-15 18:3	57.4	93.7	130.0	204.0	351.1	2.4	L**
Milk fat concentration, g/100 g fat							
cis-9, *cis*-12 18:2[5]	2.01	2.33	3.01	3.69	4.54	0.14	L**Q*
cis-9, *cis*-12, *cis*-15 18:3	0.58	1.45	3.54	5.61	9.50	0.33	L**
Sum of n-6 fatty acids	2.30	2.64	3.31	3.98	4.84	0.14	L**Q**
Sum of n-3 fatty acids	0.82	1.70	3.84	5.95	9.91	0.33	L**Q*
Secretion in milk, g/d							
cis-9, *cis*-12 18:2	26.9	31.7	37.7	47.2	52.5	3.7	L**Q*
cis-9, *cis*-12, *cis*-15 18:3	7.6	19.8	44.2	72.0	109.2	6.6	L**Q*
Total n-6 fatty acids	30.7	36.0	41.4	50.9	56.0	4.0	L**Q*
Total n-3 fatty acids	10.7	23.1	48.0	76.4	113.8	6.8	L**Q*
Apparent transfer, %							
cis-9, *cis*-12 18:2	9.6	11.1	13.1	15.6	15.8	1.3	L**Q**
cis-9, *cis*-12, *cis*-15 18:3	13.5	21.0	34.2	35.4	31.1	3.4	L**Q**
Total n-6 fatty acids	10.9	12.5	14.4	16.8	16.9	1.4	L**Q**
Total n-3 fatty acids	18.9	24.6	37.1	37.5	32.4	3.5	L**Q**

[1]L = linear and Q = quadratic effects of the level of linseed oil infusion. $^{*}P \leq 0.05$ and $^{**}P \leq 0.01$.

Table S4. Fatty acid yields in dairy cows abomasally infused with increasing levels of linseed oil

	Linseed oil infusion, mL/d						
Item, g/d	0	75	150	300	600	SEM	Contrast[1]
4:0	31.631	32.946	28.787	29.001	25.043	3.150	L**
6:0	22.671	24.679	22.166	22.356	19.447	2.176	L*
8:0	14.118	15.843	15.023	14.966	13.194	1.340	
10:0	35.423	40.522	40.155	39.303	34.828	3.417	
10:1	3.346	3.464	3.351	3.004	2.667	0.390	L*
11:0	1.063	1.154	1.121	0.906	0.740	0.164	L*
12:0	45.916	50.343	50.496	48.569	42.572	4.300	
cis-9 12:1	1.305	1.303	1.375	1.211	1.035	0.157	
iso 13:0	0.305	0.306	0.320	0.373	0.289	0.035	
anteiso 13:0	0.282	0.303	0.262	0.212	0.195	0.041	L**
13:0	1.701	1.806	1.681	1.372	1.080	0.173	L**
iso 14:0	2.052	1.866	1.627	1.622	1.297	0.227	L**
14:0	141.180	146.890	135.420	131.210	108.100	9.039	L**
cis-9 14:1	12.416	11.528	10.261	8.737	7.009	1.448	L**
cis-11 14:1	0.769	0.836	0.926	0.862	0.706	0.120	
iso 15:0	2.293	2.367	2.075	1.852	1.404	0.135	L**
anteiso 15:0	5.369	5.646	4.741	4.620	3.255	0.433	L**
15:0	15.885	16.284	13.760	12.293	9.275	1.247	L**
iso 16:0	3.636	4.029	3.691	3.793	2.482	0.304	L**Q*
16:0	420.420	398.420	327.030	318.130	256.980	38.812	L**Q*
trans-9 16:1	0.582	0.536	0.475	0.493	0.343	0.055	L**
iso 17:0[2]	2.860	2.833	2.534	2.398	1.797	0.246	L**
cis-9 16:1	17.398	14.709	11.485	9.796	7.070	1.472	L**Q**
anteiso 17:0[3]	4.917	5.211	4.334	4.421	3.504	0.445	L**
cis-11 16:1	0.455	0.353	0.324	0.362	0.416	0.090	
cis-13 16:1	2.748	2.787	2.649	2.471	1.881	0.278	L**
17:0	6.596	6.592	5.585	5.405	4.191	0.559	L**

Item, g/d	Linseed oil infusion, mL/d					SEM	Contrast[1]
	0	75	150	300	600		
cis-7 17:1	0.326	0.439	0.425	0.504	0.609	0.065	L**
cis-8 17:1	0.265	0.252	0.186	0.161	0.104	0.044	L**
cis-9 17:1	2.212	2.010	1.617	1.380	1.125	0.176	L**Q**
iso 18:0	0.769	0.678	0.642	0.732	0.498	0.090	L**
18:0	96.939	103.120	91.168	99.248	82.792	10.481	L*
trans-4 18:1	0.326	0.392	0.333	0.347	0.331	0.055	
trans-5 18:1	0.226	0.277	0.241	0.224	0.229	0.039	
trans-6-8 18:1	2.376	2.438	2.034	2.178	1.676	0.226	L**
trans-9 18:1	1.813	1.797	1.564	1.654	1.299	0.145	L**
trans-10 18:1	3.059	3.206	2.882	3.075	2.644	0.276	L*
trans-11 18:1	7.354	7.689	6.712	7.473	5.670	0.596	L**
trans-12 18:1	2.740	2.653	2.372	2.411	1.959	0.248	L**
trans-13/14 18:1	4.503	4.228	3.544	3.620	2.757	0.446	L**
trans-15 18:1	3.446	3.593	3.175	3.458	3.036	0.322	
trans-16 18:1	2.756	2.804	2.449	2.492	2.098	0.227	L**
cis-6-8 18:1	1.323	1.230	0.836	0.982	1.112	0.174	Q**
cis-9 18:1[4]	169.350	171.120	161.590	171.720	157.800	14.110	
cis-11 18:1	5.542	5.340	5.175	5.195	5.170	0.630	
cis-12 18:1	3.907	4.210	3.454	3.803	3.101	0.356	L*
cis-13 18:1	0.704	0.530	0.410	0.406	0.439	0.097	
cis-14 18:1	0.571	0.551	0.464	0.484	0.396	0.059	L**
cis-15 18:1	0.700	0.638	0.491	0.526	0.532	0.097	
cis-9, *cis*-12 18:2[5]	26.859	31.714	37.696	47.204	52.484	3.696	L**Q*
cis-9, *trans*-11 18:2[6]	3.884	3.800	3.265	3.221	2.388	0.223	L**
cis-9, *trans*-12 18:2	0.664	0.664	0.563	0.570	0.451	0.069	L**
cis-9, *trans*-13 18:2	2.786	2.822	2.258	2.182	1.666	0.235	L**
trans-8, *cis*-13 18:2	1.232	1.145	1.006	0.971	0.741	0.113	L**
trans-9, *trans*-12 18:2	0.200	0.219	0.227	0.255	0.194	0.031	
trans-9, *cis*-12 18:2	0.301	0.295	0.255	0.325	0.230	0.045	

Item, g/d	Linseed oil infusion, mL/d					SEM	Contrast[1]
	0	75	150	300	600		
trans-10, *cis*-12 18:2	0.278	0.303	0.296	0.368	0.230	0.041	Q*
trans-11, *cis*-15 18:2	0.596	0.921	0.656	1.193	1.317	0.328	
cis-6, *cis*-9, *cis*-12 18:3[5]	0.396	0.374	0.397	0.746	1.026	0.210	L**
cis-9, *cis*-12, *cis*-15 18:3[7]	7.586	19.756	44.244	71.981	109.150	6.584	L**Q*
cis-9, *trans*-11, *cis*-15 18:3	0.276	0.526	0.836	1.250	1.294	0.176	L**Q**
19:0	1.093	1.052	0.859	0.722	0.642	0.086	L**Q*
20:0	1.435	1.536	1.402	1.486	1.189	0.117	L**
cis-9 20:1	0.974	1.048	0.896	0.901	0.734	0.076	L**
cis-11, *cis*-14 20:2[5]	0.463	0.459	0.506	0.507	0.490	0.033	
cis-8, *cis*-11, *cis*-14 20:3[5]	1.049	1.163	0.901	0.675	0.566	0.105	L**Q*
cis-11, *cis*-14, *cis*-17 20:3[7]	0.218	0.320	0.627	1.026	1.319	0.106	L**Q*
cis-5, *cis*-8, *cis*-11, *cis*-14 20:4[5]	1.437	1.646	1.408	1.250	1.011	0.150	L**
cis-8, *cis*-11, *cis*-14, *cis*-17 20:4[7]	0.269	0.290	0.310	0.306	0.241	0.031	
cis-5, *cis*-8, *cis*-11, *cis*-14, *cis*-17 20:5[7]	0.884	0.935	1.051	1.177	1.524	0.190	L**
22:0	0.524	0.581	0.524	0.559	0.464	0.053	L*
cis-13 22:1	0.198	0.201	0.255	0.266	0.316	0.034	L**
cis-13, *cis*-16 22:2[5]	0.189	0.231	0.191	0.223	0.200	0.040	
cis-13, *cis*-16, *cis*-19 22:3[7]	0.196	0.242	0.259	0.219	0.187	0.037	
cis-7, *cis*-10, *cis*-13, *cis*-16 22:4[5]	0.302	0.399	0.327	0.299	0.194	0.044	L**
cis-7, *cis*-10, *cis*-13, *cis*-16, *cis*-19 22:5[7]	1.031	1.016	1.087	1.173	0.917	0.124	Q**
cis-4, *cis*-7, *cis*-10, *cis*-13, *cis*-16, *cis*-19 22:6[7]	0.220	0.227	0.245	0.259	0.216	0.035	
24:0	0.362	0.326	0.335	0.378	0.347	0.044	
cis-15 24:1	0.190	0.210	0.271	0.203	0.174	0.040	
Others	10.141	10.641	10.038	10.323	11.347	0.874	
Glycerol	162.990	167.070	152.550	156.310	139.260	13.175	L**
Sum of n-6 fatty acids	30.695	35.984	41.426	50.903	55.971	4.036	L**Q*
Sum of n-3 fatty acids	10.666	23.115	48.082	76.409	113.810	6.830	L**Q*

[1]L = linear and Q = quadratic effects of the level of linseed oil infusion. *$P \leq 0.05$ and **$P \leq 0.01$.

[2]Coelution with minor concentration of *trans*-10 16:1.

[3]Coelution with minor concentration of *cis*-10 16:1.
[4]Coelution with minor concentration of *cis*-10 18:1.
[5]n-6 family.
[6]Coelution with minor concentration of *trans*-7, *cis*-9 18:2.
[7]n-3 family.

Table S5. Fatty acid profile of plasma triacylglycerols in dairy cows abomasally infused with increasing levels of linseed oil

	Linseed oil infusion, mL/d						
Fatty acid	0	75	150	300	600	SEM	Contrast[1]
Concentration, mg/100 g plasma							
12:0	0.067	0.081	0.165	0.161	0.155	0.062	
14:0	0.21	0.27	0.61	0.58	0.54	0.25	
16:0	1.23	1.22	2.42	2.63	2.25	0.93	
16:1	0.065	0.069	0.084	0.123	0.110	0.034	
18:0	2.05	1.84	2.48	2.71	1.84	0.46	
18:1	0.92	0.97	1.61	2.00	1.98	0.55	
cis-9, *cis*-12 18:2	0.20	0.24	0.36	0.50	0.64	0.03	$L^{**}Q^{*}$
cis-9, *cis*-12, *cis*-15 18:3	0.05	0.22	0.47	0.97	1.46	0.05	$L^{**}Q^{**}$
Others	1.12	1.51	1.96	1.48	1.43	0.35	
Total fatty acids	5.90	6.42	10.16	11.15	10.41	2.45	
Relative proportion, g/100 g of fatty acids							
12:0	1.18	1.24	1.46	1.20	1.47	0.25	
14:0	3.95	4.08	4.88	4.08	5.05	0.96	
16:0	21.5	18.9	21.0	19.6	20.9	2.6	
16:1	1.07	1.06	0.81	0.98	1.03	0.13	
18:0	33.3	29.0	26.2	25.9	17.8	1.6	L^{**}
18:1	15.5	14.9	14.9	16.6	18.9	1.5	L^{*}
cis-9, *cis*-12 18:2	3.41	3.84	4.25	5.63	6.33	0.66	L^{**}
cis-9, *cis*-12, *cis*-15 18:3	1.39	3.38	5.70	10.99	14.70	1.48	L^{**}
Others	18.6	23.7	20.8	15.0	13.9	3.3	L^{*}

[1]L = linear and Q = quadratic effects of the level of linseed oil infusion. $^{*}P \leq 0.05$ and $^{**}P \leq 0.01$.

Table S6. Fatty acid profile of plasma phospholipids in dairy cows abomasally infused with increasing levels of linseed oil

	Linseed oil infusion, mL/d						
Fatty acid	0	75	150	300	600	SEM	Contrast[1]
Concentration, mg/100 g plasma							
12:0	0.134	0.123	0.144	0.185	0.146	0.017	Q^{*}
14:0	0.121	0.114	0.122	0.136	0.143	0.009	L^{**}
16:0	8.70	8.43	9.22	10.28	10.72	0.79	L^{**}
16:1	0.204	0.176	0.150	0.157	0.160	0.020	
18:0	15.7	15.7	17.8	21.6	21.5	1.7	$L^{**}Q^{**}$
18:1	6.14	5.47	5.29	6.30	7.68	0.48	$L^{**}Q^{*}$
cis-9, *cis*-12 18:2	21.8	21.8	24.9	31.4	31.0	2.5	$L^{**}Q^{**}$
cis-9, *cis*-12, *cis*-15 18:3	1.32	2.73	5.25	8.62	11.42	0.63	$L^{**}Q^{**}$
Others	10.3	10.0	11.1	13.1	13.9	1.1	L^{**}
Total fatty acids	64.4	64.6	73.9	91.7	96.7	6.9	$L^{**}Q^{*}$
Relative proportion, g/100 g of fatty acids							
12:0	0.212	0.192	0.193	0.217	0.152	0.026	L^{*}
14:0	0.190	0.181	0.170	0.153	0.147	0.012	L^{**}
16:0	13.6	13.2	12.5	11.2	11.1	0.3	$L^{**}Q^{**}$
16:1	0.329	0.279	0.206	0.170	0.165	0.035	$L^{**}Q^{*}$
18:0	24.3	24.2	24.0	23.5	22.3	0.3	L^{**}
18:1	9.69	8.58	7.21	6.92	7.96	0.53	$L^{*}Q^{**}$
cis-9, *cis*-12 18:2	33.7	33.6	33.5	34.1	32.1	0.5	L^{*}
cis-9, *cis*-12, *cis*-15 18:3	2.08	4.30	7.21	9.38	11.76	0.40	$L^{**}Q^{**}$
Others	15.9	15.5	15.0	14.3	14.4	0.4	$L^{**}Q^{**}$

[1]L = linear and Q = quadratic effects of the level of linseed oil infusion. $^{*}P \leq 0.05$ and $^{**}P \leq 0.01$.

Table S7. Fatty acid profile of plasma cholesterol esters in dairy cows abomasally infused with increasing levels of linseed oil

	Linseed oil infusion, mL/d						
Fatty acid	0	75	150	300	600	SEM	Contrast[1]
Concentration, mg/100 g plasma							
12:0	0.051	0.041	0.046	0.053	0.051	0.006	
14:0	0.71	0.63	0.61	0.62	0.55	0.06	L^{*}
16:0	3.22	2.78	2.87	3.23	3.22	0.29	
16:1	0.44	0.37	0.27	0.22	0.21	0.08	$L^{**}Q^{*}$
18:0	0.61	0.49	0.44	0.47	0.39	0.07	
18:1	2.71	2.33	2.05	1.96	1.88	0.26	$L^{**}Q^{*}$
cis-9, *cis*-12 18:2	55.3	50.5	53.2	59.9	58.9	5.0	
cis-9, *cis*-12, *cis*-15 18:3	7.97	10.95	18.52	29.26	38.62	1.44	$L^{**}Q^{**}$
Others	4.75	3.93	3.35	3.13	2.77	0.51	$L^{**}Q^{*}$
Total fatty acids	76.2	72.4	81.7	99.1	106.9	7.2	L^{**}
Relative proportion, g/100 g of fatty acids							
12:0	0.068	0.060	0.059	0.055	0.049	0.009	L^{**}
14:0	0.94	0.89	0.74	0.63	0.52	0.05	$L^{**}Q^{**}$
16:0	4.23	3.82	3.49	3.24	3.01	0.11	$L^{**}Q^{**}$
16:1	0.60	0.53	0.32	0.23	0.20	0.11	L^{**}
18:0	0.80	0.65	0.53	0.47	0.38	0.07	L^{**}
18:1	3.61	3.22	2.47	1.96	1.75	0.21	$L^{**}Q^{**}$
cis-9, *cis*-12 18:2	72.5	69.5	64.6	60.4	55.1	0.8	$L^{**}Q^{**}$
cis-9, *cis*-12, *cis*-15 18:3	10.5	15.3	23.3	29.6	36.2	1.1	$L^{**}Q^{**}$
Others	6.15	5.39	4.00	3.15	2.58	0.32	$L^{**}Q^{**}$

[1]L = linear and Q = quadratic effects of the level of linseed oil infusion. $^{*}P \leq 0.05$ and $^{**}P \leq 0.01$.

Table S8. Plasma free fatty acid profile in dairy cows abomasally infused with increasing levels of linseed oil

	Linseed oil infusion, mL/d						
Fatty acid	0	75	150	300	600	SEM	Contrast[1]
Concentration, mg/100 g plasma							
12:0	0.030	0.025	0.033	0.028	0.024	0.005	
14:0	0.142	0.135	0.139	0.112	0.123	0.027	
16:0	2.02	1.98	1.88	1.65	1.66	0.29	
16:1	0.041	0.025	0.019	0.017	0.022	0.009	
18:0	2.04	2.09	1.87	1.83	1.83	0.39	
18:1	0.90	0.64	0.83	0.58	0.73	0.10	
cis-9, *cis*-12 18:2	0.25	0.24	0.24	0.33	0.36	0.03	L**
cis-9, *cis*-12, *cis*-15 18:3	0.05	0.08	0.17	0.34	0.52	0.04	L**
Others	0.70	0.65	0.66	0.47	0.58	0.13	
Total fatty acids	6.17	5.88	5.85	5.37	5.85	0.88	
Relative proportion, g/100 g of fatty acids							
12:0	0.49	0.43	0.56	0.53	0.45	0.08	
14:0	2.33	2.23	2.27	2.04	2.21	0.28	
16:0	32.7	33.5	31.6	30.8	28.6	1.3	L**
16:1	0.67	0.45	0.33	0.31	0.32	0.12	
18:0	32.9	34.9	31.6	33.2	29.7	2.0	
18:1	14.8	11.6	15.3	10.8	12.9	1.7	
cis-9, *cis*-12 18:2	4.01	4.33	4.30	6.48	6.38	0.44	L**
cis-9, *cis*-12, *cis*-15 18:3	0.88	1.48	2.88	7.20	9.29	0.76	L**
Others	11.22	11.17	11.21	8.65	10.10	1.53	

[1]L = linear effect of the level of linseed oil infusion. **$P \leq 0.01$.

Table S9. Distribution of fatty acids among plasma lipid classes in dairy cows abomasally infused with increasing levels of linseed oil

	Linseed oil infusion, mL/d						Contrast[1]
Distribution, %	0	75	150	300	600	SEM	
12:0							
Triacylglycerols	25.3	29.8	36.8	30.6	40.4	7.0	
Phospholipids	47.1	45.3	41.2	49.0	39.5	5.6	
Cholesterol esters	16.6	15.7	13.5	13.4	13.7	2.1	
Free fatty acids	11.0	9.2	8.5	7.0	6.3	1.2	L**
14:0							
Triacylglycerols	19.5	23.0	33.3	31.3	37.6	7.4	L*
Phospholipids	10.0	10.0	9.8	11.2	10.8	1.3	
Cholesterol esters	58.5	55.5	47.5	49.8	42.6	6.3	L*
Free fatty acids	12.3	11.5	9.4	7.8	9.0	1.4	L*Q*
16:0							
Triacylglycerols	8.7	8.4	13.8	13.9	12.2	4.2	
Phospholipids	56.9	58.5	57.5	58.6	60.5	4.1	
Cholesterol esters	20.7	19.2	17.4	18.3	18.1	1.2	
Free fatty acids	14.0	13.9	11.3	9.2	9.2	1.4	L**Q*
16:1							
Triacylglycerols	10.3	12.0	16.5	20.7	23.1	4.9	L*
Phospholipids	31.7	28.4	32.3	34.0	33.5	5.8	
Cholesterol esters	53.1	55.6	47.6	42.1	39.5	7.5	L**
Free fatty acids	5.9	4.1	3.6	3.3	3.9	1.1	
18:0							
Triacylglycerols	10.2	9.5	11.0	10.6	7.1	1.9	
Phospholipids	76.3	77.6	78.7	80.7	84.2	3.1	L**
Cholesterol esters	3.1	2.4	1.9	1.7	1.6	0.3	L**Q*
Free fatty acids	10.4	10.6	8.4	7.0	7.1	1.7	L**

	Linseed oil infusion, mL/d						Contrast[1]
Distribution, %	0	75	150	300	600	SEM	
Σ18:1							
Triacylglycerols	9.3	10.8	15.7	17.6	15.9	4.2	
Phospholipids	57.7	57.6	54.7	59.1	62.9	3.5	
Cholesterol esters	25.4	24.7	20.7	18.1	15.3	1.7	L^{**}
Free fatty acids	8.2	6.9	9.0	5.2	6.0	1.1	
cis-9, *cis*-12 18:2							
Triacylglycerols	0.26	0.35	0.48	0.56	0.70	0.05	$L^{**}Q^{*}$
Phospholipids	28.8	29.8	31.9	33.8	34.2	1.4	$L^{**}Q^{*}$
Cholesterol esters	70.5	69.5	67.3	65.3	64.7	1.3	$L^{**}Q^{**}$
Free fatty acids	0.34	0.34	0.33	0.37	0.40	0.05	L^{*}
cis-9, *cis*-12, *cis*-15 18:3							
Triacylglycerols	0.8	1.6	2.0	2.5	2.8	0.3	$L^{**}Q^{**}$
Phospholipids	14.9	19.2	21.4	21.8	21.9	1.0	$L^{**}Q^{**}$
Cholesterol esters	83.5	78.5	76.0	74.8	74.2	1.0	$L^{**}Q^{**}$
Free fatty acids	0.64	0.62	0.69	0.88	1.01	0.12	L^{**}
Others							
Triacylglycerols	7.2	9.9	11.1	8.6	7.6	1.9	
Phospholipids	60.5	61.8	65.5	71.4	74.4	2.6	$L^{**}Q^{*}$
Cholesterol esters	28.0	24.2	19.3	17.2	14.8	1.7	$L^{**}Q^{**}$
Free fatty acids	4.4	4.2	4.1	2.8	3.2	1.0	
Total fatty acids							
Triacylglycerols	4.1	4.4	6.0	5.6	4.7	1.4	
Phospholipids	42.7	43.0	43.2	43.9	44.1	1.7	
Cholesterol esters	49.1	48.5	47.4	47.8	48.6	1.0	
Free fatty acids	4.3	4.0	3.5	2.6	2.7	0.5	L^{**}

[1]L = linear and Q = quadratic effects of the level of linseed oil infusion. $^{*}P \leq 0.05$ and $^{**}P \leq 0.01$.