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Effect on lymphoproliferation and *in vitro* Ig production of splenocytes from suckling rats when supplemented with conjugated linoleic acid

C. Ramírez-Santana, F. J. Pérez-Cano, M. Castell, A. Franch and C. Castellote

Department of Physiology, Faculty of Pharmacy, University of Barcelona, Barcelona, Spain

Conjugated linoleic acid (CLA) refers to a mixture of positional and geometric isomers of linoleic acid that contain two conjugated double bonds, of which cis-9, trans-11 and trans-10, cis-12 predominate. Besides the health effects described in human subjects, it has been reported that CLA modulates immune cell functions in animal models, although there are no reports on this effect during early life. Thus, the objective of the present work was to establish the effect of CLA supplementation during gestation and/or lactation on ability of spleen lymphocytes to produce Ig and proliferate under ex vivo conditions. Pups were divided into four groups (A, B, C and W), with each group comprising ten rats per lactating mother. Mothers of C and W group pups were fed standard pellet chow from day 7 of gestation and throughout the study period. Mothers of group A and B pups were fed pelleted chow with 10 g CLA (80% cis-9, trans-11, 20% trans-10, cis-12; Lipid Nutrition B.V. Wormerveer, The Netherlands)/kg during gestation and mothers of group A pups continued with this supplemented chow until weaning. Groups B and C received the CLA isomer mixture by daily oral supplementation while dams were fed standard pelleted chow during lactation. Animals were killed and their spleens were removed at day 21, the end of suckling period. Splenocytes were isolated and cultured. After incubation for 7 d, in vitro IgG and IgM production was quantified using the ELISA sandwich technique. Cells stimulated with phorbyl myristate acetate-ionomycin (250 ng/ml) were used to: (1) quantify IL-2 and interferon-y (IFN-y) levels in culture supernatant fractions after 24 h incubation using ELISA; (2) to measure cell proliferation after incubation for 72 h (see Table). Cell viability was also evaluated. Conventional ANOVA and post hoc comparisons (LSD test) were performed. Significant differences were accepted at P < 0.05. No differences among groups were seen in *in vitro* IL-2 and IFN- γ production or lymphocyte proliferation (see Table), irrespective of life period (gestation or suckling), duration and route of supplementation. However, spontaneous IgM production by splenocytes from group B animals was higher compared with that of groups C and W (P<0.05; see Table). These results suggest that supplementation with CLA during the gestation and suckling periods increases spontaneous Ig production by immunocompetent cells.

Group	IgM production (ng/ml)		Proliferation (%)	
	Mean	SE	Mean	SE
A	937.9	144.0	196.9	37.60
3	1225.6*†	193.5	257.2	29.33
2	539.5	139.7	234.5	57.89
N	534.9	157.3	191.5	33.23

n 10. Mean value was significantly different from that for group C: *P < 0.05. Mean value was significantly different from that for group W: $\dagger P < 0.05$.