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Polyunsaturated fatty acid metabolism in monocyte differentiation

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The differentiation of peripheral blood monocytes is a key step in the development of functional macrophages. Polyunsaturated fatty acids (PUFA) exert an important influence on cell function by determining membrane fluidity, and hence the biophysical environment of integral proteins, and by providing substrates secondary mediator synthesis[(1)]. However, knowledge about the effect of monocyte differentiation on membrane PUFA content is limited and conflicting[(2,3)]. It has been shown that macrophages are able to synthesise PUFA[(2)], although the functional significance is unclear. Here we investigated the effect of monocyte to macrophage differentiation using the THP-1 monocytic cell line as an in vitro model on the PUFA content of total cell lipids and on the mRNA expression of FADS1 and FADS2 which encode two key enzymes in PUFA biosynthesis, delta-5 and delta-6 desaturases, respectively.

Human THP-1 monocytes were differentiated to macrophages using 100 nM phorbol 12-mystiate 13-acetate for 72 hours[(4)]. Differentiation was confirmed by an increase in CD11c and CD36 expression by flow cytometry. Membrane fatty acid composition was measured by gas chromatography[(5)]. FADS1 and FADS2 mRNA expression was measured by real-time RT-PCR using cyclophilin as the reference gene[(6)].

THP-1 monocyte differentiation induced an increase in the proportion of 18:2*n*-6 and a decrease in 20:4*n*-6 decrease in all *n*-3 PUFA, in particular 20:5*n*-3, 22:5*n*-3 and 22:6*n*-3 (Table). The expression of both FADS1 and FADS2 was lower in THP-1 macrophages than monocytes (Table).

	Proportion of total fatty acids (g/100g)					
	Monocyte		Macrophage			
Fatty Acid	Mean	SD	Mean	SD	Difference (%)	*P*
18:2*n*-6	10.8	0.4	12.1	0.5	12.0	0.022
18:3*n*-6	0.2	0.01	0.14	0.01	8.7	0.078
20:3*n*-6	1.6	0.1	1.4	0.1	11.3	0.24
20:4*n*-6	5.5	0.3	4.7	0.2	15.7	0.015
18:3*n*-3	0.1	0.01	0.1	0.01	5.0	0.004
20:5*n*-3	1.1	0.04	1.0	0.04	10.8	0.023
22:5*n*-3	2.1	0.1	1.7	0.1	20.1	0.008
22:6*n*-3	3.2	0.1	2.4	0.1	24.1	0.002
	mRNA expression (ng target relative to ng cyclophilin)					
FADS1	1.08	0.27	0.51	0.20	47.7	0.022
FADS2	0.60	0.16	0.12	0.04	80.0	0.0001

Statistical comparisons were by unpaired Student's *t*-test. A significant difference was assumed at $P>0.05$

These findings show that differentiation of THP-1 monocytes was accompanied by changes in the proportions of specific PUFA. The increased in 18:2*n*-6 and decrease in 20:4*n*-6, 20:5*n*-3, 22:5*n*-3 and 22:6*n*-3, and the reduction in FADS1 and 2 mRNA expression suggests that decreased PUFA synthesis may be involved. Overall, these findings suggest that THP-1 monocytes and macrophages, cultured under identical conditions, maintain membrane PUFA content independent of the fatty acids present in the medium. In addition, the changes in membrane PUFA content during monocyte differentiation suggests PUFA synthesis *de novo* may represent a novel locus of control of monocyte differentiation and macrophage function.

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