



Regulation of T lymphocyte functions by novel engineered polyunsaturated fatty acids

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Summary

Natural polyunsaturated fatty acids (PUFA) were synthesized and assessed as potential anti-inflammatory agents. β -oxa, β -thia and γ -thia PUFA were found to inhibit T lymphocyte responses *in vitro*, assessed as lymphoproliferation as well as production of tumour necrosis factor- β (TNF β), interferon- γ (IFN γ) and interleukin-2 (IL-2). Three of these compounds were studied in detail and found to be more active than the inhibition seen with fish oil fatty acid 22:6 n -3. Interestingly these compounds unlike 22:6 n -3 were poor stimulators of the human neutrophil respiratory burst, an additional advantage in their use as anti-inflammatory agents.

Examination of one of these three, β -oxa 21:3 n -3, in *in vivo* models of chronic and acute inflammation showed that the fatty acids was highly inhibitory in both types of inflammatory responses. Thus the β -oxa 21:3 n -3 inhibited the footpad swelling induced by sheep red blood cell antigens in a delayed type hypersensitivity (DTH) reaction as well as in the carrageenan-induced paw inflammation. This finding with DTH which is dependent on T lymphocytes is consistent with the ability of β -oxa 21:3 n -3 to inhibit Th1 type cytokine production. While the reason for its effects on the carrageenan induced inflammation are less clear in this involves predominantly neutrophil infiltration, this reaction also has some T cell dependency. Studies on tissue distribution of the fatty acid, showed that β -oxa 21:3 n -3 was incorporated into membrane phospholipids of liver and blood cells. Preliminary toxicology studies also demonstrated that β -oxa 21:3 n -3 was well tolerated by the animals as several chemical and biochemical parameters of liver and kidney function remained within the normal range.

The mechanism of inhibition of T lymphocyte responses were in the main identified. The β -oxa 21:3 n -3 acted at a post receptor level affecting signals modules critical for lymphocyte

activation and cytokine production. The fatty acid caused inhibition of PHA-PMA induced translocation of PKC- β I and PKC- ϵ , but not PKC- α , PKC- β II or PKC- θ . This was further delineated to show inhibition of downstream ERK pathway activation but not the JNK or p38 pathways.

This work has identified a new class of anti-inflammatory compounds which are based on PUFA. The action of the fatty acids is through the selective inhibition of the PKC- β 1/ ϵ and ERK signalling module.