Arachidonic Acid Stimulates the Cytosolic Phospholipase A_2-dependent Synthesis of Lipid Droplets via JNK and p38 Phosphorylation

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In this work we present data to suggest that these results suggest that concomitant activation of p38 and JNK by arachidonic acid (AA) cooperate to activate group IVA cytosolic phospholipase A_2 (cPLA$_2\alpha$), which is in turn required for lipid droplet formation. Lipid droplet formation by AA can be completely inhibited by selective inhibition of cPLA$_2\alpha$ by pyrrophenone, pointing out this enzyme as a key regulator of AA-induced signaling. Lipid droplet formation in AA-treated monocytes can also be blocked by the combined inhibition of the mitogen-activated protein kinase family members p38 and JNK, which correlates with inhibition of cPLA$_2\alpha$ activation by phosphorylation.

SLIDE 1 – AA-induced TAG and CE formation in human monocytes. The cells were treated with 10 μM AA for 2 h. Afterward, fatty acids in TAG and CE were analyzed by GC-MS after converting the fatty acid glyceryl and cholesteryl esters into fatty acid methyl esters.

SLIDE 2 – AA-induced LD formation in human monocytes. Monocytes, treated without or with 3 μM triacsin C for 30 min, were exposed to AA, palmitic acid (16:0), oleic acid (18:1), linoleic acid (18:2), or γ-linolenic acid (γ 18:3) for 2 h. After fixation and permeabilization, cells were stained with BODIPY493/503to visualize the lipid droplets.

SLIDE 3 – Effect of AA on the expression of genes involved in de novo fatty acid synthesis in human monocytes. The relative expression of genes in control or cells treated with AA was determined by qPCR.

SLIDE 4 – AA-mediated signaling leading to lipid droplet formation requires cPLA$_2\alpha$. Monocytes were untreated or treated with AA for 2 h in the presence of the indicated inhibitors.

SLIDE 5 – Stimulation of mitogen-activated protein kinases and cPLA$_2\alpha$ by AA in human monocytes. Analysis of the effect of various MAPK inhibitors on phosphorylation of the enzyme by immunoblot and activity by in vitro assay.

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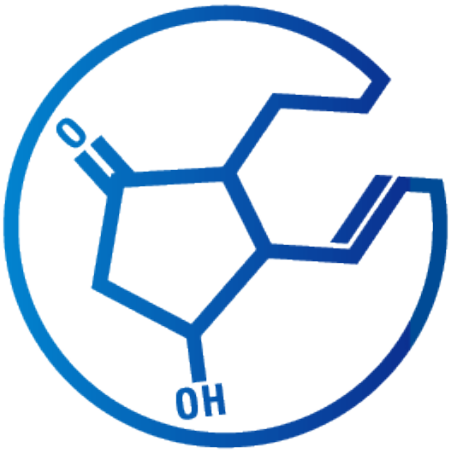

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