

DIFFERENTIAL PROTECTIVE EFFECTS OF POLYUNSATURATED FATTY ACIDS ON MUSCLE LIPOTOXOCITY

Aurélien BRUN*, Philippe DENIS*, Frédéric CAPEL*

*Unité de nutrition humaine – Equipe ASMS – 63000 Clermont-FD

Context: Preserving the mobility and autonomy of individuals by maintaining muscle mass and quality has become one of the significant public health issues in Western societies due to the increased proportion of people aged 65 and above. The discovery of myosteatorosis as a chronic process of ectopic lipid infiltration in skeletal muscle has been repeatedly demonstrated in cases of sarcopenia and cancer-cachexia. If mechanistic studies have demonstrated the links between myosteatorosis and muscle dysfunctions, some nutritional preventive strategies were proposed.

Methods: A physiopathological intracellular accumulation of saturated lipids was performed in C2C12 differentiated muscle cells exposed to 500 μ M of Palmitic acid (PAL) to mimic clinically observed myosteatorosis. Lipid-induced cytotoxicity was studied up to 72h. Nuclei permeabilization revealing cell death was detected after Hoechst 33258 coloration with a convolutional neural network that we trained on images over 30 000 nuclei. Cell viability was assessed with Cell Titer Glo 2.0 (#G9242) quantification. We then measured the effects of coincubation, preincubation with n-3 (Docosahexaenoic acid = DHA, Eicosapentaenoic acid = EPA, α -linolenic acid = ALA) and n-6 (Arachidonic acid = ARA) polyunsaturated fatty acids (PUFA) at different doses and timing to get a clear overview of differential effects of these nutrients.

Results: Over-representation of nuclei permeabilization of cells started at 12 hours of treatment with PAL meanwhile decreased viability was detectable after 16 hours ($p < 0.05$). Myotubes disappeared subsequently and viability was reduced, reaching only 40% and 25% of the control value after 24 and 48 hours of treatment respectively ($p < 0.05$). Coincubation with the different PUFA at 12.5, 25, 50, 100 μ M showed preventive effects of DHA and ARA at 25, 50 and 100 μ M.

Discussion: These results showed interesting preventive effects of ARA and DHA on lipid-induced muscle toxicity. A viability and cell death overview was necessary to understand what happens over time and which polyunsaturated fatty acids were good candidates for our subsequent tests. ARA and DHA were thereafter chosen for mitochondria function assessment and anabolic resistance assays.