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N-3 AND N-6 POLYUNSATURATED FATTY ACIDS DIFFERENTLY AFFECT CITRATE CARRIER PROMOTER ACTIVITY

Fabrizio Damiano, Simone Alemanno, Eleonora Stanca, Luisa Siculella and Gabriele Vincenzo Gnoni

Laboratory of Biochemistry and Molecular Biology, DiSTeBA, University of Salento, Via Monteroni, Lecce 73100, Italy

Citrate carrier (CiC), a mitochondrial membrane protein, plays an important metabolic role by transporting, in the form of citrate, acetyl-CoA from mitochondria into the cytosol for fatty acid and cholesterol synthesis. A PUFA (polyunsaturated fatty acids) response region, composed of a NF-Y site, an E-box like site, a SRE1 like site and four Sp1 sites, has been identified within the CiC promoter. Transcription factor SRE-Binding Protein-1 (SREBP-1c) is target for PUFA down-regulation of CiC transcription. Transfection and gel mobility shift assays indicated that a functional E-box like confers responsiveness to SREBP-1c. In H4IIE cells overexpression of SREBP-1c overrides arachidonic acid suppression but does not prevent the repression by docosahexaenoic acid. ChIP assay showed that docosahexaenoic acid affects the binding of NF-Y, Sp1 and SREBP-1 to PUFA response region whereas arachidonic acid alters only the binding of SREBP-1. PUFA inhibition of CiC gene transcription is mediated not only by the SREBP-1c but might also involve a reduction in Sp1 and NF-Y DNA binding, suggesting differential mechanisms in the CiC gene regulation by different PUFA.

KINETIC CHARACTERIZATION OF A NOVEL COPPER AMINE OXIDASE ACTIVITY FROM RAT LIVER MITOCHONDRIA MATRIX

**Sara Cardillo, A. De Iuliis, V. Battaglia, G. Sinigaglia, M. A. Grillo, M. Magro, A. Toninello
R. Stevanato and F. Vianello**

Department of Biological Chemistry, University of Padua, Padua, Italy

The present study reports preliminary results on the presence of a novel Copper containing amine oxidases (Cu-AO, EC 1.4.3.6.) in rat liver mitochondria lysates. Such enzymatic activity was found in the soluble mitochondrial fraction, obtained by simple osmotic shock. The enzyme was isolated by a new procedure based on the binding on iron oxide nanoparticles chemically modified with an enzyme substrate. SDS-PAGE showed a single band at about 60 kDa. The crude enzyme activity was tested by spectrophotometric measurements, determining hydrogen peroxide production following oxidative deamination of different substrates, such as polyamines (spermine, spermidine, putrescine and cadaverine) and monoamines (dopamine and benzylamine). The highest activity was found with 1,2-diamino-ethane and the highest affinity with 1,5-diamino-pentane. The enzyme was preliminary kinetically characterized, using spermine, spermidine and putrescine as substrates, as a function of ionic strength.