Long-chain acyl-CoA synthetase 6 partitions fatty acids toward lipid synthesis in human and rodent skeletal muscle

Bruno G Teodoro1; Igor H Sampaio1; Lucas HM Bomfim4; André L. Queiroz1; Leonardo dos Reis Silveira4; Tai-Yu Huang2; Donghai Zheng2; P Darrell Neufer2; Ronald N Cortright2; Luciane C Alberici3.

1Universidade de São Paulo, Departamento de Bioquímica, Faculdade de Medicina de Ribeirão Preto, Ribeirão Preto, Brasil. 2East Carolina Diabetes and Obesity Institute, Greenville, NC, United States. 3Universidade de São Paulo, Departamento de Física e Química, Faculdade de Ciências Farmacêuticas de Ribeirão Preto, Ribeirão Preto, Brasil. 4 IB, Universidade Estadual de Campinas, Campinas, Brasil.

Introduction:

Fatty acids are activated intracellularly by long-chain Acyl-CoA synthetases (ACSL 1-6 isoforms) providing acyl-CoA substrate for mitochondrial β-oxidation or esterified lipids synthesis such as phospholipids and triacylglycerol. Whether ACSL6 influences the partitioning of acyl-CoAs, particularly in skeletal muscle, is unknown.

Objectives:

Using both gain- and loss-of-function strategies, we determined whether ACSL6 influences lipid partitioning toward mitochondrial oxidation or cytosolic synthesis in skeletal muscle cells from rats or humans.

Materials and Methods:

Skeletal muscle of rat lower limb and human vastus lateralis were cultured from satellite cells. Knockdown and overexpression of ACSL6 was induced by transfection of ACSL6-specific siRNA and an ACSL6 cDNA expression vector, respectively, for the following experiments: (1) mRNA expression (qRT-PCR) (2) lipid species profiling (MS-MS), (3) mitochondrial oxygen consumption, (4) \text{H}_2\text{O}_2 production, (5) fluorescence analysis of lipid content and (6) radioative palmitate oxidation.

Results And Discussion:

In rat cells, ACSL6 siRNA transfection reduced ACSL6 mRNA (70±8%; p<0.01). ACSL6 silencing increased C16:0 and C18:0 fatty acids (32±3% and 35±3%, p<0.05), basal cellular respiration (297.6±30.2 x 368.1±28.7 \text{O}_2 \text{pmol.s}^{-1}.10^6 \text{cells}^{-1}; p<0.05), mitochondrial proton leak (91.5±5.2 x 96.8±4.7 \text{O}_2 \text{pmol.s}^{-1}.10^6 \text{cells}^{-1}; p<0.05) and maximal uncoupled respiration (610.4±45.2 x 703.5±41.7 \text{O}_2 \text{pmol.s}^{-1}.10^6 \text{cells}^{-1}; p<0.01). The ACSL6 knockdown also increased
expression of oxidative genes PGC1α (50%), UCP2 (3 fold) and UCP3 (5 fold), palmitate oxidation (30%) and, decreased H$_2$O$_2$ production and several lipid species. In human primary myotubes, ACSL6 overexpression (50-fold mRNA increase) decreased radiolabeled palmitate oxidation (32%; p<0.05). Also in humans, ACSL6 mRNA increased in response to acute ingestion of a high fat meal (70% kcal from fat) in both lean and obese subjects (~2.5 times; p<0.05).

**Conclusion:** We report for the first time that ACSL6 is acutely induced by dietary lipid intake and that it appears to play an important role in lipid partitioning in skeletal muscle in both humans and rodents, driving acyl-CoA away from mitochondrial oxidation and toward lipid synthesis.

**Keywords:** Fatty Acid Oxidation, Lipid Synthesis, Mitochondria

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