Supplemental Data

Material & Methods

Murine 3T3L1 adipocytes

High-glucose DMEM (4.5 g/l D-glucose) containing 4 mM L-glutamine and FBS for 3T3-L1 adipocytes were from Invitrogen (Karlsruhe, Germany). For adipocyte differentiation, insulin and dexamethasone were obtained from Sigma-Aldrich (Steinheim, Germany), and 3-isobutyl-1-methylxanthine (IBMX) was purchased from Serva (Heidelberg, Germany).

Murine 3T3-L1 preadipocytes (American Cell Culture Collection, Manassas, VA, USA) were maintained in DMEM with 4.5 g/l glucose supplemented with 10% FBS and 1x penicillin/streptomycin (PS) in a 5% CO_2 humidified environment at 37°C prior to adipocyte differentiation. At 100% plus one day confluency, differentiation of preadipocyte cultures was induced using high-glucose DMEM containing 10% FBS, 1x PS, 5 µg/ml insulin, 0.5 mM IBMX, and 0.25 µM dexamethasone (day 0). On day 3, medium was replaced by high-glucose DMEM containing 10% FBS, 1x PS and 5 µg/ml insulin and changed every 2 days thereafter. Adipocytes were differentiated over a 7-day period and used for metabolite investigations on day 7.

Figure Legend

Supplementary Figure S1. Pharmacological treatment with the carnitine palmitoyltransferase 1 inhibitor etomoxir followed by palmitate loading in 3T3-L1 adipocytes.

Representative acylcarnitine products of short- (C4:0), medium- (C8:0), and long-chain (C16:0) species following inhibition of fatty acid β-oxidation by etomoxir pre-treatment and subsequent palmitate loading (100 µM, 120 min) are depicted. Etomoxir was dissolved in H_2O. Prior to permeabilization, 3T3-L1 adipocytes were pre-treated with different etomoxir concentrations (10, 20, 50, 100 µM) for 30 min (42) in DMEM (1 g/l glucose) and 0.25% fatty acid-free BSA.

Three independent experiments were performed in triplicates each. Results are presented as mean ± SEM and expressed as fold decrease in metabolite abundance compared to the untreated control (H_2O; indicated by the dotted line). Statistically significant differences of pharmacological treatment relative to the untreated control are indicated by an asterisk ($p < 0.05$). For each metabolite, values not sharing a common letter indicate statistically significant differences in metabolite abundance between concentrations ($p < 0.05$).

Supplementary Fig. S1