SUPPLEMENTAL MATERIAL AND METHODS

Energy expenditure

Energy expenditure in mice acutely exposed to chow or ketogenic diets was monitored using a 32-cage combined indirect calorimetry system. Chronic diet exposure was monitored using a similar 24-cage system (PhenoMaster, TSE Systems GmbH, Bad Homburg, Germany). Automated measurements of energy expenditure and respiratory quotient were further combined with food and water intake measurements by integrating scales into the sealed cage environment. Locomotor activity was monitored by using infrared light beam break systems, with higher infrared light beam density at x-y-direction in the 32-cage-system compared to the low-density beam break monitoring in x-direction for the 24-cage setup.

Hepatic and renal PEPCK activity assay

About 50 mg of snap frozen liver and kidney samples were homogenized in 4 ml/g (liver) or 2 ml/g (kidney) buffer containing 10 mM Tris-HCL (pH 8), 0.3 M Sucrose, 0.5% BSA, 1 mM EDTA and 1 mM dithiothreitol. Cytosolic fraction were obtained by 1 h of centrifugation at 100.000 g and at 4°C. 20 µl of the cytosolic fraction were assayed at 37°C in 96-well plates in a solution containing 110 mM imidazole-Cl, pH 6.8, 13 mM NaF, 3 mM $MgSO_4$, 3 mM $MnCl_2$, 10 mM phenylalanine, 1 µM rotenone, 45 mM $NaHCO_3$, 0.15 mM NADH, 6 units malate dehydrogenase, 2 mM phosphoenolpyruvate. Reactions were started by adding 0.5 mM dGTP and monitored by measuring changes in absorbance at 340nm. Control samples were measured simultaneously in absence of phoesphoenolpyruvate and $NaHCO_3$. Enzyme activity was normalized to the amount of total protein present in the supernatant. Protein

amounts were determined by the Bicinchoninic acid assay (BCA) (Pierce, Rockford, IL) method according to the manufacturer's instructions.