EFFECTS OF QUANTITATIVE TRAIT LOCUS FOR LIPID PHENOTYPES IN THE RAT ARE INFLUENCED BY AGE
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The analysis of genetic factors involved in lipid regulation might shed light on genetic mechanisms underlying features of the coronary heart disease, hypertension, non-insulin-dependent diabetes and obesity which are major causes of ill health in industrial societies. Our previous study on the backcross hybrids derived from a cross of the spontaneously hypertensive rat (SHR/Ma) and the spontaneously diabetic BB/D-KR rat demonstrated the existence of quantitative trait loci (QTLs) affecting lipid phenotypes on chromosome 4 and suggestive linkage of lipid phenotypes with markers on chromosome 1. Since the previous study was performed with backcross hybrids at 12 weeks of age, and it is known that lipid phenotypes can show age-related differences, in this study the effect of QTLs (chromosome 1 and 4) on serum triglycerides and cholesterol was analysed at 20, 28 and 32 weeks of age of backcross hybrids. The results of this study showed that the effect of QTL on chromosome 4 (between Ldh1 and Danatrl) for serum triglycerides was maximal at 28 weeks of age, but disappeared at 32 weeks of age. On the other hand the effect of QTL on serum total cholesterol on chromosome 4 (Olp-Spl) was maximal at 32 weeks of age. In contrast to our previous (12 weeks), the longitudinal investigation showed significant linkage of D/Min14 marker with lipid phenotypes on chromosome 1. Our findings indicate the necessity of considering the important role of age in QTL analysis of phenotypes showing temporal changes.

PLASMA FATTY ACID UTILIZATION AND FATTY ACID BINDING PROTEIN CONTENT ARE DIMINISHED IN SKELETAL MUSCLE OF TYPE 2 DIABETIC SUBJECTS

Impairments in muscle fatty acid (NEFA) metabolism were studied in patients with type 2 diabetes. 8 obese patients with mild type 2 diabetes (50±59 g y, body fat: 38±8%, HbA1c: 6.4±0.3%) and 8 non-obese controls (CON; 51±62 g y, body fat: 16±8%, %) were studied using the forearm balance technique and indirect calorimetry during infusion of the stable isotope tracer [1-13C]palmitate after an overnight fast and during infusion of the non-selective β3-agonist isoprenaline (ISO, 20 ng/kg 1.8mL−1 min−1). Additionally, markers of skeletal muscle oxidative, glycolytic, and fatty acid transport capacity were determined in biopsies from the vastus lateralis muscle. Both during fasting as well as ISO-infusion, muscle NEFA uptake (CON vs type 2; fast: 44±28 vs 259±42, ISO: 715±126 vs 591±70 mmol·100 ml·isotopic τ−1, min−1, P<0.05), and release were lower in type 2 diabetes as compared to CON. Also, muscle plasma NEFA oxidation during ISO-infusion was blunted in type 2 diabetes (CON vs type 2; ISO: 44±274 vs 18±70 mmol·100 ml·isotopic τ−1, min−1, P=0.05). The diminished skeletal muscle fat utilization in type 2 diabetes was accompanied by a lowered cytosolic content of fatty acid binding protein (CON vs type 2; 1.1±0.2 vs 0.5±0.1 mg protein, P<0.5), a lowered activity of 3-hydroxyacyl-CoA dehydrogenase (CON vs type 2; 25±6 vs 29±7 U total protein, P=0.05), and a tendency towards a lowered citrate synthase activity (CON vs type 2; 102±19 vs 62±11 U total protein). In summary, there is a pronounced reduction in skeletal muscle NEFA uptake and oxidation of type 2 diabetic subjects both overnight fasted and ISO-stimulated conditions. The diminished fat utilization was accompanied by a lowered cytosolic FABP content and a lowered oxidative capacity, indicating that a lowered cytosolic transport capacity is involved in disturbances in fat utilization.

MEASUREMENT OF NON-ESTERIFIED FATTY ACIDS IN INTERSTITIAL FLUID OF SUBCUTANEOUS ADIPOSE AND MUSCLE TISSUE
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Non-esterified fatty acids (NEFA) play a potential role in the pathogenesis of muscle resistance and type 2 diabetes. Aim of this study was to develop a method for direct access of extracellular fluid of the subcutaneous adipose and muscle tissue. Method: The novel method of open flow microperfusion is based on a microcapsulized perfused double lumen catheter perfused by a sterile Krebs Ringer solution. The microcapsulized perfusion of the outer lumen of the catheter enables a free exchange of molecules independent of their size between the interstitial fluid of the surrounding tissue and the perfuse solution. The partially equilibrated fluid was transported to the collecting cannula by a high precision roller pump at a flow rate of 1.0 µl/min. In basic evaluation studies, zero flow rate protocols showed a flow dependent recovery of NEFA, albumin, urea, and glucose. We measured albumin and NEFA in 5 healthy male volunteers (age 43±23 years, BMI 22±5±1.2 m²/kg) after overnight fast. Interstitial fluid was sampled in intervals of 30 min, corresponding plasma samples were obtained. NEFA were measured using NEFA-C (Wako Chemicals, Germany). Results: In the fasting state NEFA concentration in the sampled fluid of muscle was (mean±SEM) 0.31±0.12 mmol/l and significantly higher to subcutaneous adipose tissue (0.10±0.01 mmol/l, P<0.008). NEFA levels in the corresponding plasma samples were 0.41±0.08 mmol/l. Discussion: Open flow microperfusion allows direct measurement of NEFA concentrations in subcutaneous adipose and muscle tissue. In the fasting state NEFA levels in muscle are considerably higher compared to adipose tissue. This novel technique will provide valuable insight into human lipid and carbohydrate metabolism.