

Translational Control of Anabolic Resistance in Aging Muscle

NCT03839628

July 24, 2018

Study Protocol

Objectives

This is an interventional study enrolling older individuals aged 60-85y to observe changes during physical inactivity. This study is investigating the relation between short-term physical inactivity and the impact of muscle health and function.

Experimental Design and Methodology

Subject Characteristics. Older adults were recruited from the Salt Lake City area via local advertisements and Center on Aging registry. To determine eligible older adult volunteers, an initial screening visit occurred which included a medical history, a physical exam, blood tests to determine liver and kidney function (to exclude cardiovascular, liver and respiratory disease, uncontrolled hypo/hyperthyroidism and hypertension, history of deep vein thrombosis) and an oral glucose tolerance test (OGTT) and HbA1c to exclude those with pre-diabetes. Subjects had normal glucose regulation as determined from HbA1c. Participants read and signed the informed consent (IRB_00084354) and the study was reviewed and approved by the University of Utah Institutional Review Board and conformed to the Declaration of Helsinki and Title 45, US code of Federal Regulations, Part 46, "Protection of Human Subjects".

Lean Mass and Muscle Function Tests. Lean mass assessment via dual-energy X-ray absorptiometry (DXA), and knee extensor isometric strength was determined. These tests will occur within one week before the reduced activity experiment (below) and will be repeated similarly after completing the reduced activity period.

Leucine Anabolic Resistance Experiment and Muscle Biopsies. On a separate day subjects were admitted to the University of Utah clinical research center (CCTS) the evening before the anabolic resistance experiment and were provided a standardized meal (65% CHO, 20% fat, 15% protein) at dinner prepared by the University of Utah CCTS Metabolic Kitchen (dietician). The next morning, after an overnight fast (10h) and placement of an intravenous antecubital catheter, subjects underwent a muscle biopsy procedure. After the biopsy, subjects consumed a 500mL of low-caloric flavored water mixed with 2.5g of crystalline L-Leucine (Leu). We and others have shown that this dose of leucine in older adults is effective to elicit an acute anabolic response (mTORC1, protein synthesis) to ingestion, yet demonstrate anabolic resistance when repeated after bed rest or 14d of reduced activity. Additional biopsies occurred at 1 and 3h after consumption of the drink, representing key time points to capture peak and late leucine protein anabolic responses. Biopsies occurred on the same leg and out of separate incisions. Blood sampling for amino acid and insulin concentrations occurred at baseline and every 20min following leucine ingestion. Muscle tissue was immediately washed with saline and dissected of visible non-muscle tissue, flash-frozen in liquid nitrogen for later analysis. A remaining portion of non-frozen tissue was used for histochemical analysis for fiber CSA determination. After the reduced activity period (below), participants repeated the anabolic resistance experiment as detailed above except the biopsies occurred on the opposite leg.

Reduced Physical Experiment. After completion of the first leucine anabolic resistance experiment, subjects adhered to 14-days of reduced physical activity. The goal was to have the participant drop their physical activity levels by 75% of their normal activity levels (e.g., 8,000 to 2,000 steps) as determined by a hip-placed pedometer that the participant will be able to see and record. Reduced physical activity is effective to elicit protein-induced anabolic resistance by 25-50% as well as reduce muscle size and strength and glucose tolerance. Participants were encouraged to maintain their normal habitual diet during the reduced activity. On Day 14, after an overnight stay and fast, a second leucine anabolic resistance experiment coupled with muscle biopsies was repeated as detailed above. Lean mass, strength, power and OGTT measurements were re-assessed 24h later.