

Effects of an antioxidant supplementation on oxidative stress and muscle dysfunction in FSH-MD patients

The purpose of this study was to gain further insights into the specific targets of endogenously generated mitochondrial oxidative stress in the FSHD skeletal muscle and to test whether or not an antioxidant supplementation has a therapeutic interest for FSHD patients. Our results have important implications for the successful implementation of rational antioxidant therapy in FSHD in which cell loss could be linked to mitochondrial dysfunction and oxidative stress.

To analyze relevant and specific changes in protein abundance and /or modification associated with FSHD, we conducted a non biases proteomic analysis of the skeletal biopsies of FSHD and control patients. The data we have obtained regarding oxidative stress and mitochondrial function were in favour of an impaired mitochondrial energy metabolism and oxidative free radical damage that could in turn lead to a fiber type-related differential oxidative injury. Electron microscopy revealed gross morphological abnormalities in mitochondria of FSHD. Accompanying these deficits, FSHD also exhibited decreased levels of proteins involved in protection from oxidative stress.

Consistent with these findings, FSHD patients showed a decreased serum antioxidant capacity and increased proteins and lipid peroxydation. The combination of proteomic, genetic and physiological analyses have provided the first direct evidence of mitochondrial dysfunction and oxidative stress in FSHD (paper in preparation).

Accordingly, we propose to evaluate the therapeutic interest of an antioxidant supplementation on functional deficits and molecular skeletal muscle abnormalities in patients FSHD, according to a randomised and double-blind crossover protocol. Two teams, including scientists and physicians, will participate to this study. The antioxidant supplementation efficiency will be assessed by:

- 1/** Clinical evaluation (evaluation of diet history, evaluation of the levels of physical activity, quality of life, body composition, pulmonary function and electrocardiogram),
- 2/** Functional evaluation (respiratory, quadriceps and deltoid muscles and whole body exercise evaluations),
- 3/** Biochemistry and molecular analysis: for the assessment of mitochondrial properties we propose to evaluate the ultra-structure of mitochondria by electronic microscopy and mitochondria function on permeabilized skinned fibers by oxygraphic method. Further more we will complete this study by protein analysis approaches.

For evaluation of oxidative stress we propose to measure reactive oxygen species (ROS) by electron spin resonance and spin trapping. The mitochondrial and cytoplasmic potential redox will also be evaluated. Lipid peroxidation and oxidized proteins levels will be evaluated in plasma by the quantification of malondialdehydes (MDA) and within the skeletal muscle by the quantification of lipofuscin inclusions. Measurement of antioxidants status will be evaluated in plasma by total antioxidant capacity (TAC) and within skeletal muscle by the measurement of the GSSG level (oxidized glutathione), and determination of the GSH/GSSG ratio. Samples of *Vastus lateralis* muscle will be obtained by needle biopsy after local lidocaine anaesthesia at mid thigh level using the Bergström technique. Thirty FSHD male and female between 20 and 60 years old} that met the criteria for definite FSHD (i.e. presenting between 4 and 9 repetitions) will be recruited.

Manuel Cabral
Président bénévole

Amis FSH
Luttons contre la dystrophie Facio-Scapulo-Humérale
www.FSHD-Group.EU