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ORIGINAL ARTICLE

Weight increase and overweight are associated with DNA oxidative damage in skeletal muscle ☆

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KEYWORDS
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Summary
Background and aims: Weight maintenance within normal standards is recommended for prevention of conditions associated with oxidative injury. To compare oxidative damage in a post mitotic tissue, between adults differing in long-term energy balance.
Methods: During hernia surgery, a sample of skeletal muscle was obtained in 17 non-obese adults. Subjects were divided into two groups according to their self-reported weight change: weight maintainers (WM) reported <4 kg increase, and weight gainers (WG) reported >5 kg increment. Muscle immunohistochemistry for 8-hydroxy-deoxyguanosine (8OHdG), 4-Hydroxy-2-nonenal (4HNE), and TNF-α, as markers of oxidative injury and inflammation, were performed. As known positive controls for oxidative injury, we included 10 elderly subjects (66–101 yr). Anthropometric measures and blood samples for clinical laboratory and serum cytokines (TNF-α and IL-6) were obtained.
Results: 8OHdG was higher in WG compared with WM (149.1 ± 16.2 versus 117.8 ± 29.5, $P=0.03$), and was associated with anthropometric indicators of fat accumulation. 4HNE was similar in WG compared with WM (10.9 ± 7.6 versus 9.8 ± 6.3) but noticeably higher in elderly subjects (21.5 ± 15.3, $P=0.059$). TNF-α protein in WG was higher compared with WM (114.0 ± 41.7 versus 70.1 ± 23.3, $P=0.025$), and was associated with weight increase.

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Conclusions: Moderate self-reported weight increase, and body fat accumulation, suggesting long-term positive energy balance is associated with muscle DNA oxidative injury and inflammation.

Introduction

The most accepted hypothesis of aging proposes that physiological age-related changes are a consequence of oxidative damage to cellular macromolecules due to free radical attack originated in mitochondria.[1] Intracellular concentrations of oxidized macromolecules such as lipids, proteins and DNA, increase as a function of age in different animal species, including human beings.[2] This would signal the occurrence of an imbalance between free radical production during cell metabolism and the cellular antioxidant defense and repair systems.[3] Most evidences supporting this hypothesis of aging derive from animal studies, which confirm an inverse relationship between longevity and oxidative stress.[4] One of the most constant scientific findings is the prolongation of life span in animal models, through caloric restriction (CR).[5,6] The underlying mechanisms are still not completely understood, however the prevention of oxidative injury is the most widely accepted hypothesis.[7]

The age-related decline in muscle mass and performance has been attributed to oxidative damage of mitochondrial genome and enzymes (e.g. aconitase), which cause a deficiency in the activity of respiratory chain complexes.[8] In human beings, higher concentrations of protein carbonyls have been detected in post mitotic tissues such as brain, muscle and eye lens.[9] An age-dependent increase of human muscle oxidative damage to DNA, lipids and proteins was demonstrated by measuring 8-hydroxy-2-deoxyguanosine (8OHdG), malondialdehyde (MDA) and protein carbonyl groups, respectively, in skeletal muscle samples obtained during orthopedic surgery.[10] Nevertheless, whether these products of oxidative stress are the best biomarkers of ageing, is still a matter of debate.[11]

Epidemiological data support the anti-ageing effects of leanness,[12,13] however long-term follow-up studies are lacking.[14] On the other hand, some evidences indicate that obesity, the phenotypic expression of a positive energy balance, induces oxidative stress,[15–17] and a pro-inflammatory condition associated with elevation of adipose-derived cytokines and peptides.[18,19] In fact obese subjects die young, mostly from cardiovascular and metabolic complications associated with fat accumulation. An acceleration of ageing or of age-related cellular changes has not been directly demonstrated in overweight or obese subjects.

Furthermore, age-related muscle wasting (sarcopenia) has been attributed to increased expression of TNF-α,[20] a cytokine also related to obesity and centripetal fat distribution.[21] Actually in elders, the accretion of fat has been ascribed to the loss of muscle mass which decreases metabolic rate.[22,23] However, under certain circumstances, overnutrition can cause steatosis, lipotoxicity, and lipoapoptosis. The most affected tissues are β cells, myocardium and skeletal muscle.[24] Both aging and the metabolic syndrome could represent these lipotoxic conditions.[25,26] Nevertheless the relationship between adipose tissue growth and the expression of this cytokine in skeletal muscle has been studied mostly in obese or old patients, but has not been evaluated in young healthy subjects, as a measure of ''physiologic age''.

Therefore, the present study aimed to verify if healthy middle-aged non-obese weight gainers (WG) show signs of higher oxidative damage compared with weight maintainers (WM), and thus leaner subjects. With this purpose we compared oxidative injury and TNF-α protein expression in skeletal muscle, between subjects reporting maintenance of body weight during most of their adult life, and those reporting progressive weight increase and body fat accumulation, with a maximal body mass index (BMI) of 30 kg/m^2.

Subjects and methods

Among adult male subjects with abdominal or inguinal hernias, scheduled for surgery during the next few days, we selected those who accepted to participate in this study by signing a written informed consent. Exclusion criteria were age below 30 yr, cigarette smoking ($\geqslant$1 cigarette/day), vegetarianism or repeated dieting with weight fluctuations, diabetes mellitus, immune diseases, acute infectious conditions, BMI $>$ 30, ingestion of vitamin supplements, treatment with