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Research paper

Simvastatin and oxidative stress in humans: A randomized, double-blinded, placebo-controlled clinical trial

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ABSTRACT

Simvastatin reduces the blood concentration of cholesterol by inhibiting hydroxymethylglutaryl-coenzyme A reductase, the rate-limiting enzyme in cholesterol synthesis, and thereby reduces the risk of cardiovascular disease. In addition, simvastatin treatment leads to a reduction in fluxes in mitochondrial respiratory complexes I and II and might thereby reduce the formation of reactive oxygen species, which have been implicated in the pathogenesis of arteriosclerosis. Therefore, we hypothesized that simvastatin may reduce oxidative stress in humans in vivo.

We conducted a randomized, double-blinded, placebo-controlled study in which subjects were treated with either 40 mg of simvastatin or placebo for 14 days. The endpoints were six biomarkers for oxidative stress, which represent intracellular oxidative stress to nucleic acids, lipid peroxidation and plasma antioxidants, that were measured in urine and plasma samples.

A total of 40 participants were included, of which 39 completed the trial. The observed differences between simvastatin and placebo groups in the primary outcomes, DNA and RNA oxidation, were small and nonsignificant ($p=0.68$), specifically, 3% in the simvastatin group compared to 7.1% in the placebo group for DNA oxidation and 7.3% in the simvastatin group compared to 3.4% in the placebo group. The differences in biomarkers related to plasma were not statistically significant between the treatments groups, with the exception of total vitamin E levels, which, as expected, were reduced in parallel with the reduction in plasma cholesterol.

In healthy young male volunteers, short-term simvastatin treatment, which considerably reduces cholesterol, does not lead to a clinically relevant reduction in a panel of measures of oxidative stress. Whether simvastatin has effects on oxidative stress in diseased populations, such as diabetes or hemochromatosis, where oxidative stress is prominent, is unknown but seems unlikely.

1. Introduction

The statin group of drugs is widely used to treat hypercholesterolemia and to reduce the risk of cardiovascular events in risk groups [1]. One common side effect of statins is muscle pain, and the mechanism by which this pain occurs is not fully understood but could include oxidative stress. Statins have different effects in skeletal and cardiac muscle, which may be why muscle problems are only observed in skeletal muscle [2] and not in other types of muscles. The primary site of ROS production is in mitochondria [3,4]. However, there are many sources of ROS, both intracellular [5] and environmental [6]. Recently, a reduction in mitochondrial respiration in skeletal muscle was reported in patients treated with simvastatin. This reduction was mainly related to reduced respiration in mitochondrial respiratory complexes I and II mediated *via* a reduction in the content of the electron carrier Q10 [7]. Furthermore, atorvastatin and simvastatin reduced the oxidative stress induced by calcium [8].

Oxidative stress to RNA can be measured non-invasively based on the excretion of the ribonucleoside 8-oxoGuo (8-oxo-7,8-dihydroguanosine) in urine, and its clinical relevance has been

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demonstrated in type 2 diabetes, where high oxidative stress is predictive of death [9]. We have hypothesized that RNA oxidation is related to the production of hydrogen peroxide by mitochondria, which could contribute to mitochondrial respiration/dysfunction and excess ROS production in diabetes [10,11]. A reduction in oxidative stress by statins would therefore represent a novel and interesting therapeutic mechanism.

We conducted a randomized, double blinded, controlled trial of simvastatin versus placebo in healthy male volunteers with DNA and RNA oxidation as the primary endpoints. The secondary endpoints were the plasma concentrations of malondialdehyde, vitamin C, vitamin E, and biopterin, which are markers of

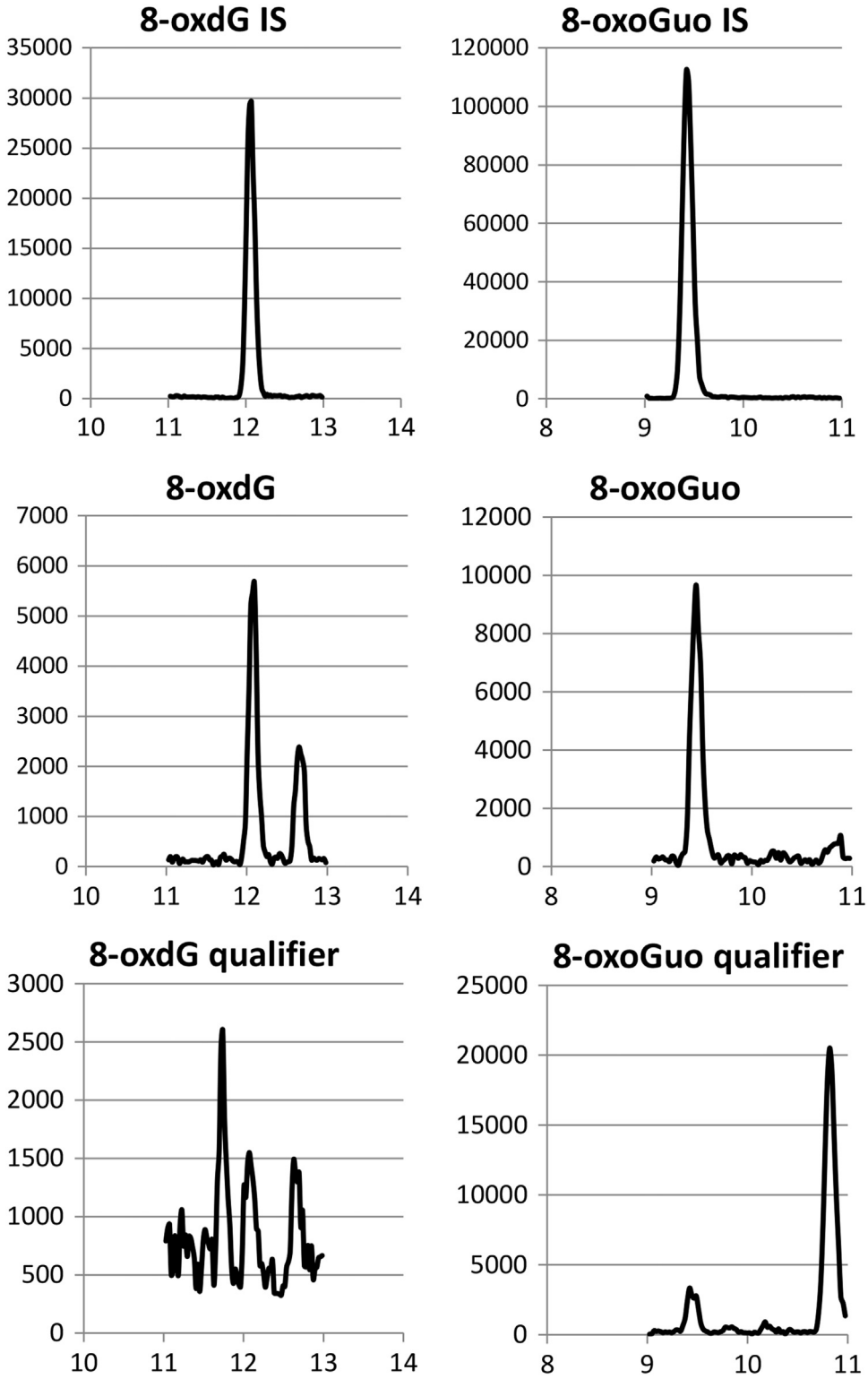

Fig. 1. Chromatogram tracings from analysis of a urine sample showing the applied ion-traces for each analyte, the $^{15}N_5$-labelled internal standard (IS), the quantifier ion, and the qualifier ion. The sample concentrations of 8-oxoGuo and 8-oxodG was 14.0 nM and 14.2 nM, respectively. The transitions are: *m/z* 303.186 > 212.99 (8-oxoGuo IS). *m/z* 298.186 > 207.99 (8-oxoGuo). *m/z* 298.186 > 165 (8-oxoGuo qualifier). *m/z* 287.186 > 196.939 (8-oxodG IS). *m/z* 282.186 > 191.939 (8-oxodG). *m/z* 282.186 > 150 (8-oxodG qualifier).