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Statin-Induced Increases in Atrophy Gene Expression Occur Independently of Changes in PGC1α Protein and Mitochondrial Content

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Abstract

One serious side effect of statin drugs is skeletal muscle myopathy. Although the mechanism(s) responsible for statin myopathy remains to be fully determined, an increase in muscle atrophy gene expression and changes in mitochondrial content and/or function have been proposed to play a role. In this study, we examined the relationship between statin-induced expression of muscle atrophy genes, regulators of mitochondrial biogenesis, and markers of mitochondrial content in slow- (ST) and fast-twitch (FT) rat skeletal muscles. Male Sprague Dawley rats were treated with simvastatin (60 or 80 mg kg⁻¹ day⁻¹) or vehicle control via oral gavage for 14 days. In the absence of overt muscle damage, simvastatin treatment induced an increase in atrogin-1, MuRF1 and myostatin mRNA expression; however, these were not associated with changes in peroxisome proliferator gamma co-activator 1 alpha (PGC-1α) protein or markers of mitochondrial content. Simvastatin did, however, increase neuronal nitric oxide synthase (nNOS), endothelial NOS (eNOS) and AMPK α-subunit protein expression, and tended to increase total NOS activity, in FT but not ST muscles. Furthermore, simvastatin induced a decrease in β-hydroxyacyl CoA dehydrogenase (β-HAD) activity only in FT muscles. These findings suggest that the statin-induced activation of muscle atrophy genes occurs independent of changes in PGC-1α protein and mitochondrial content. Moreover, muscle-specific increases in NOS expression and possibly NO production, and decreases in fatty acid oxidation, could contribute to the previously reported development of overt statin-induced muscle damage in FT muscles.
Introduction

Statin drugs lower blood cholesterol, and thus reduce the risk of coronary heart disease and stroke, by inhibiting the rate limiting enzyme of the mevalonate pathway, 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase (for review see [1]). With reductions in low-density lipoprotein (LDL) cholesterol of up to 55%, statins have become the most commonly prescribed drug in the world today, with more and more populations being indicated for their use [2,3]. Although generally well tolerated, one of the main side effects of statin medications is skeletal muscle myopathy, with clinical symptoms that include muscle pain (myalgia), inflammation (myositis), weakness, fatigue and cramping [4,5]. With an incidence of >10% of statin users in the general population [6,7], hundreds of thousands of people worldwide are likely to experience some form of statin-induced myopathy. Statin related muscle symptoms also appear to be exacerbated by exercise [8]. Thus, statin-induced myopathy has the potential to markedly affect levels of physical activity and quality of life [9], and could prompt the discontinuation of the statin therapy altogether. Therefore, a thorough understanding of the molecular mechanism(s) underlying statin myopathy is essential for the future identification of specific biomarkers to detect adverse statin-induced events prior to the potential onset of debilitating clinical symptoms and/or for the development of safer alternative cholesterol lowering agents.

Although the exact mechanism(s) responsible for statin-induced myopathy remains to be definitively determined, recent studies have suggested that mitochondria and/or the activation of muscle atrophy-related genes may play a role (for reviews see [10–12]). For example, statins have been shown to up regulate the expression of the muscle-specific ubiquitin proteasome system (UPS) E3-ligases, atrogin-1 and MuRF1, in a range of model systems including statin-myopathy patients [13–18]. Importantly, statin-induced muscle atrophy/damage was markedly reduced in myotubes from atrogin-1 knockout mice and in Zebra fish transfected with atrogin-1 siRNA [14]. Together, these findings suggest that an increase in the expression of muscle specific E3-ligases (i.e. atrogin-1 and MuRF1) play a crucial role in statin-induced muscle fiber atrophy/damage and may help to explain muscle pain and weakness associated with statin-myopathy.

Numerous clinical, animal and cell culture studies have provided evidence that statin-myopathy is also associated with impaired mitochondrial function and morphology (e.g. [17–29]). In addition, recent studies also suggest that statins induce a reduction in mitochondrial content/volume [14,23,30–32]; an effect that could, in part, be due to reduced mitochondrial biogenesis. Mitochondrial biogenesis is positively regulated by a variety of signaling molecules and transcriptional co-activators, including the peroxisome proliferator gamma co-activator 1 alpha (PGC1α) [33]. For example, PGC1α binds to and co-activates nuclear respiratory factor 1 (NRF1) which, in turn, regulates the transcription of mitochondrial transcription Factor A (Tfam) [33]. Recent studies have reported a statin-induced reduction in PGC1α mRNA expression in humans, rodents and cultured cells [23,32]. Therefore, statins could down regulate mitochondrial biogenesis via a reduction in PGC1α expression. Importantly, PGC1α has also been proposed to play an inhibitory role against the activation of atrophy gene expression and muscle atrophy [14,34,35]. Thus, a statin-induced decrease in PGC1α protein and/or co-transcriptional activity could reduce mitochondrial biogenesis and also play a role in the induction of atrophy gene expression. To date, however, no studies have examined the relationship, if any, between changes in PGC1α protein, markers of mitochondrial content and the expression of atrophy genes in skeletal muscle with in vivo statin treatment.

In contrast to their effect on PGC1α expression, non-muscle studies have shown that statins also positively regulate two other important activators of mitochondrial biogenesis