Effect of β-Hydroxy-β-Methylglutaryl Coenzyme A Reductase Inhibitor Atorvastatin on Contractility of the Isolated Rat Heart under Normal Conditions and during Oxidative Stress

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Long-term administration of β-hydroxy-β-methylglutaryl coenzyme A reductase inhibitor atorvastatin to rats was accompanied by an increase in the relative weight of the heart and decrease in the rate of pressure development in the isovolumic heart. During oxidative stress induced by addition of 100 μM H_2O_2 to the perfusate, the decrease in contractile function was more pronounced that in the control. Our results indicate that administration of atorvastatin is accompanied by a decrease in myocardial contractility, which becomes more pronounced under conditions of oxidative stress.

Key Words: *β-hydroxy-β-methylglutaryl coenzyme A reductase inhibitors; oxidative stress; myocardial contractility; isolated rat heart; atorvastatin*

Statins, cholesterol-lowering drugs from the class of inhibitors of β-hydroxy-β-methylglutaryl coenzyme A reductase (HMG-CoA reductase) block not only cholesterol synthesis, but also biosynthesis of the major natural antioxidants ubiquinone (ubiphenol) Q_{10} and glutathione peroxidase, a Se-containing enzyme utilizing lipid hydroperoxides [4,5,9, 10,12]. Hence, statins can provoke oxidative stress in the organism. Since coenzyme Q_{10} is involved in electron transfer in the mitochondrial electron transport chain, statin-induced decrease in coenzyme Q_{10} level can impair energy supply to tissues. This was demonstrated in our previous experiments on the myocardium [5,9]. These changes can modulate myocardial contractility and resistance to oxidative stress. The present work was designed to test this hypothesis.

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MATERIALS AND METHODS

Experiments were performed on Wistar rats weighing 370±30 g. Atorvastatin in starch gel was administered daily (10 mg/kg, 30 days) via a gastric tube. The hearts were removed under urethane anesthesia (1.7 g/kg) and perfused Krebs—Henseleit solution saturated with carbogen (5% CO_2 and 95% O_2, pH 7.4) and containing 11 mM glucose at 37°C through the aorta. The perfusion rate approached 10 ml/g/min. A latex balloon filled with physiological saline was introduced into the left ventricle (LV) through the left atrium. Balloon volume was set so that diastolic LV pressure remained constant (12-14 mm Hg). Pressure in the aorta and LV and dP/dt were recorded with Gould Statham P23 Db electromanometers using a Gould Brush 2400 polygraph. Under isovolumic conditions, the major parameters of myocardial contractility and energy consumption were developed pressure and contractile function intensity (CFI, product of developed pres-

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sure and heart rate). The maximum CFI was measured during a gradual 2-fold increase in the perfusion rate. Oxidative stress in the myocardium was induced by addition of 100 μM H_2O_2 to the perfusate. The use of a Sage rate-controlled infusion pump allowed us to maintain H_2O_2 concentration at a constant level. The results of statistical treatment are presented as $M \pm m$.

RESULTS

Despite similar body weight in control and atorvastatin-treated rats, the weight of heart ventricles in treated rats was higher than in controls and the ratio of heart ventricle weight to body weight in control and atorvastatin-treated rats was 0.317±0.009 and 0.339±0.005%, respectively ($p<0.05$). A strong correlation exists between body weight and weight of the heart. The increase in this ratio reflects the development of mild myocardial hypertrophy (7%). Our results contradict the notion that statins prevent the development of heart hypertrophy [13]. It should be emphasized that administration of simvastatin to untreated rat was not accompanied by changes in the heart weight/body weight ratio [7].

Under normal conditions, CFI of the heart in atorvastatin-treated rats was slightly lower than in control animals (by 13%, Table 1). The developed pressure differed insignificantly, but the rate of pressure development and drop decreased by 24% after atorvastatin administration ($p<0.01$). During gradual 2-fold increase in the perfusion rate CFI of the heart in atorvastatin-treated rats increased more significantly and the maximum value of this parameter did not differ from that in control animals (752±26 and 755±37 mm Hg/sec, respectively). At the same time, the maximum rate of pressure development in treated rats remained 17% below the control level (4260±190 *vs*. 5110±178 mm Hg/sec, respectively, $p<0.05$). Other parameters of myocardial function did not differ in treated and control rats. It should be emphasized that there are no data on variations in contractile activity of the myocardium during long-term administration of statins. *In vivo* studies showed that long-term treatment with simvastatin blocks the positive ionotropic effect of dopamine [8]. However, administration of simvastatin in a wide range of concentrations was followed by an increase in contractility of the isolated heart [14]. We showed that the maximum rate of contraction decreases during functional load. Taking into account the fact that other parameters of contractile function do not differ from the control, it can be suggested that myosin ATPase activity in the myocardium decreases in atorvastatin-treated rats. These changes are typical of myocardial hypertrophy induced by pressure overload [1]. Simvastatin produces a direct activating effect on myocardial function [14], which contributes to the development of mild myocardial hypertrophy in rats of this group. Impairment of myocardial contractility under the influence of HMG-CoA reductase inhibitor is probably associated with a decrease in the level of macroergic phosphates in the myocardium and inhibition of coenzyme Q_{10} biosynthesis [5,9].

Oxidative stress in the isolated heart can be modeled by adding of H_2O_2 to the perfusate. Degradation of this compound results in generation of reactive oxygen species (ROS), primarily bioactive hydroxyl radical. Addition of H_2O_2 to the perfusate was followed by a progressive decrease in contractile function of the heart from control animals (Fig. 1). Cardiac function in atorvastatin-treated rats decreased more significantly under these conditions; the differences were significant at various periods after H_2O_2 administration ($p<0.001$). Similarly to control animals, these changes in treated rats were related to the decrease in developed pressure and maximum rate of pressure development. However, the frequency of contractions in rats of both groups increased from 4.0-4.1 to 4.5-4.6 Hz after H_2O_2 administration. The effect of H_2O_2 after long-term treatment with atorvastatin was also accompanied by a progressive increase in perfusion pressure and end-diastolic pressure. By the end of H_2O_2 administration, this parameter was 2-fold higher than in the control. Hence, the myocardium in atorvastatin-treated rats was characterized by low resistance to oxidative stress. Our results contradict the hypothesis that statins reduce the elevated concentration of ROS after norepinephrine treatment [11] and prevent the development of H_2O_2-induced apopto-

TABLE 1. Contractile Function of the Heart under Basal Conditions

Parameter	Control (n=10)	Atorvastatin (n=10)
Frequency of contractions, Hz	3.8±0.1	3.7±0.1
Developed pressure, mm Hg	146±5	137±5
Myocardial CFI, mm Hg/sec	573±26	496±23*
Maximum rate of pressure development, mm Hg/sec	3950±204	3000±154*
Maximum rate of pressure drop, mm Hg/sec	2720±128	2280±148*
Minimum diastolic pressure, mm Hg	3±1	4±1
Perfusion pressure, mm Hg	56±1	60±2

Note. *$p<0.05$ compared to the control.

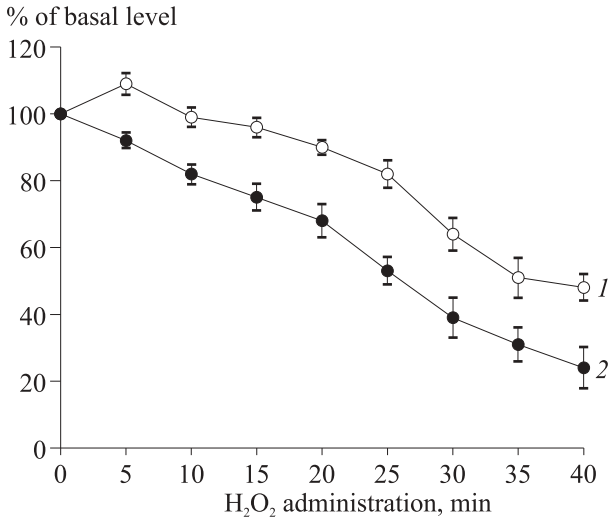

Fig. 1. Effect of H_2O_2 on CFI of the isolated hearts from control rats (*1*) and atorvastatin-treated animals (*2*).

sis [6]. This effect probably depends on the dose of the test preparation, since simvastatin in high concentration decreases the number of viable cardiomyocytes in the culture [14]. Our previous studies on the same model showed that coenzyme Q_{10} plays an important role in protection of the myocardium from oxidative stress [2,3]. Hence, it is not unexpected that inhibitor of coenzyme Q_{10} biosynthesis (atorvastatin) increases the severity of oxidative stress in the myocardium. The results of our study are consistent with the notion that HMG-CoA reductase inhibitors can induce side prooxidant effect [4,9,10]. These changes result in a decrease in contractile function of the myocardium, particularly under conditions of oxidative stress. We previously [2,3] showed that coenzyme Q_{10}-containing drugs effectively protect the myocardium from adverse effects of oxidative stress.

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