Betaxolol, a Cardioselective Beta-Adrenoceptor Antagonist, Attenuates Ischemic Myocardial Acidosis in Dogs

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Abstract—The effects of betaxolol, a cardioselective beta-adrenoceptor antagonist, on ischemic myocardial acidosis were studied in dog hearts, in which the left anterior descending coronary artery was partially occluded for 90 min, and were compared with those of atenolol and propranolol. Myocardial ischemia produced a decrease in myocardial pH (measured by a micro glass pH electrode) and an elevation of the ST segment of epicardial ECG (assessed by a surface electrode). Betaxolol (0.01, 0.03 or 0.1 mg/kg), atenolol (0.03 or 0.1 mg/kg) or propranolol (0.03 or 0.1 mg/kg), when injected i.v. 30 min after ischemia, restored myocardial pH and the ST segment of ECG that had been altered by partial occlusion. However, the effect of betaxolol on myocardial acidosis was more potent than that of atenolol or propranolol. The decrease in (+)dp/dt by betaxolol (0.03 mg/kg) was less potent than that by atenolol (0.1 mg/kg) and equivalent to that by propranolol (0.1 mg/kg), although the restorations of myocardial acidosis by the drugs were almost equivalent. These results have confirmed that beta-adrenoceptor antagonists attenuate the ischemia-induced myocardial acidosis and have shown that among three beta-adrenoceptor antagonists, betaxolol is the most effective in improving myocardial acidosis with a relatively weak effect on myocardial contractile function.

Betaxolol, (±)-1-[4-[[2-(cyclopropylmethoxy)ethyl]phenoxy]-3-(isopropylamino)-2-propanol hydrochloride, is a cardioselective beta-adrenoceptor antagonist which has no partial agonistic activity and little or no membrane-stabilizing activity (1–3). With its long elimination half-life and its high bioavailability after oral administration (4), betaxolol produces the potent and long lasting reduction in myocardial oxygen demand (estimated from the double product) and improvement of exercise tolerance in patients with angina pectoris (5).

It is well-established that the tissue pH decreases in response to myocardial ischemia probably because of the production of lactate and the breakdown of high energy phosphates in the ischemic myocardium (6, 7). Many investigators have demonstrated that beta-adrenoceptor antagonists inhibit the ischemia-induced myocardial acidosis (7–14). These findings suggest that the myocardial pH could be a useful indicator in evaluating the severity of myocardial ischemic injury and the efficacy of the drugs on ischemic heart diseases.

In the present study, we have investigated the effects of betaxolol on the ischemic myocardial acidosis in dog hearts subjected to partial occlusion of the left anterior descending coronary artery (LAD) in comparison with those of atenolol, another cardioselective beta-adrenoceptor antagonist, and propranolol, a nonselective beta-adrenoceptor antagonist.

Materials and Methods

Experimental preparation: Healthy mongrel dogs of either sex weighing 8 to 22 kg were anesthetized with sodium pentobarbital (30 mg/kg, i.v.). Dogs were ventilated with a positive-pressure respirator. A left thoraco-
tomy was performed at the fourth intercostal space and the left ventricular wall was ex-
posed. After a portion of LAD was dissected free from the adjacent tissues at the level just proximal to the first diagonal branch, an oc-
cluder consisting of a thread, a polyethylene tubing and a fine clip was placed around the LAD. The thread of the occluder was looped around the artery and passed through the tubing to maintain the partial occlusion. Coronary blood flow was measured with an electromagnetic flow probe (Nihon Kohden, FR-020T) placed around the LAD. The left ventricular pressure was determined by means of a pressure transducer-tipped catheter (Mi-
lar Instruments, MPC-500) passed through the left carotid artery into the left ventricle. The first derivative of the left ventricular pres-
sure (dp/dt) was obtained by electronic differentiat-
ion of the left ventricular pressure pulse. Systemic blood pressure was measured with a pressure transduced (Nihon Kohden, MPU-0.5) in the right femoral artery. Heart rate was counted from the pressure trace of blood pressure. An epicardial ECG was taken with a wire electrode from the surface of the left ventricular wall perfused by the LAD. All these parameters were recorded on a poly-
graph (Nihon Kohden, RE-6000) throughout the whole course of the experiments.

Determination of myocardial pH: As de-
scribed previously by Abiko et al. (12), myocardial pH was measured continuously with a micro glass pH electrode (Microelec-
trodes, MI-410). The pH electrode was inserted at the depth of 6 to 8 mm in the left ventricular wall (i.e., in the subendocardial layers) perfused with LAD. Changes in myocardial pH were recorded on a pen recorder via a pH meter (Horiba, M-8s). The mean value of myocardial pH in each of the groups was determined in such a way that each of the pH values measured by the micro glass pH electrode was first converted to the corres-
ponding hydrogen ion concentration ([H⁺]) to calculate the mean value of [H⁺], which was converted again to the mean pH value.

Experimental protocol: After all the param-
eters were stabilized, myocardial ischemia was induced by partial occlusion of LAD. By using the occluder, the LAD flow was re-
duced to about one-third of the original flow and maintained constant at this level for 90 min. These procedures also have been de-
scribed previously by Abiko et al. (12). Thirty minutes after partial occlusion, saline, be-
taxolol, atenolol or propranolol solution was injected i.v. in the right femoral vein over a period of 30 sec. The partially occluded LAD was released 60 min after the drug injection. In some experiments with betaxolol, the heart was driven at a constant rate, being about 20 beats/min higher than the original rate, with an electronic stimulator. Rectangular pulses having 1 msec durations with the voltage slightly higher than threshold were applied to the right atrium.

Drugs: Betaxolol hydrochloride (Synthe-
labo), atenolol hydrochloride (Sigma) or pro-
pranolol hydrochloride (Sigma) was dis-
solved in saline solution immediately before use. The volume of injection was 0.5 ml/kg. The dose of the drugs was expressed in terms of the respective salts.

Statistical analysis: Results were evaluated by the analysis of variance followed by Bonferroni’s multiple comparison test, and a P value of 0.05 or less was considered signifi-
cant.

Results

Control experiments: Changes in LAD flow, myocardial pH, ST segment of epicar-
dial ECG, (+)dp/dt, heart rate and blood pres-
sure in dogs subjected to 90 min of partial oc-
closure of LAD are shown in Fig. 1. Myocar-
dial pH (measured by a micro glass pH elec-
trode) was 7.51 (mean) in the nonischemic myocardium. The partial occlusion of LAD (LAD flow was reduced from 17.4±2.2 to 7.3±1.7 ml/min) resulted in marked decrease in myocardial pH; the pH decreased rapidly in response to partial occlusion, reached a steady level (6.88) 30 min after occlusion, and maintained the decreased level during 90 min of partial occlusion. The partial occlusion of LAD also produced an elevation of the ST segment of epicardial ECG and a decrease in (+)dp/dt. Blood pressure and heart rate did not change during ischemia. When the par-
tially occluded LAD was released 90 min after occlusion, LAD flow increased quickly and overshoot the preocclusion level. Myocar-
dial pH and the ST segment of epicardial
ECG returned to preocclusion levels after reperfusion, whereas (+)dp/dt recovered incompletely after reperfusion.

**Betaxolol experiments:** The effects of betaxolol (0.1 mg/kg) on LAD flow, myocardial pH, ST segment of epicardial ECG,

![Graph showing changes in coronary flow, myocardial pH, ST segment of epicardial ECG, (+)dp/dt, heart rate, and blood pressure after partial occlusion of the left anterior descending coronary artery in dogs.](image)

*Fig. 1.* Changes in coronary flow, myocardial pH, ST segment of epicardial ECG (ΔST), (+)dp/dt, heart rate and blood pressure after partial occlusion of the left anterior descending coronary artery in dogs. ΔST represents the difference in ST segment of epicardial ECG between before and after partial occlusion. Saline solution (0.5 ml/kg) was injected i.v. 30 min after partial occlusion. Vertical bars and open area in myocardial pH represent S.E.s. Number of dogs (n)=7.*
(+\text{d}p/\text{d}t, \text{heart rate and blood pressure in the ischemic hearts are shown in Fig. 2. Betaxolol was injected i.v. 30 min after partial occlusion when the decreased level in the pH had become stable. The myocardial pH, that had been decreased from 7.48 to 6.91 by partial occlusion, increased gradually and reached to 7.33 60 min after the injection of betaxolol, without any changes in the LAD flow. The elevation of the ST segment of epicardial ECG was also attenuated by betaxolol. Betaxolol decreased the heart rate from 144±

\begin{figure}
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\includegraphics[width=\textwidth]{figure2.png}
\caption{Effect of betaxolol on ischemic myocardial pH in dogs. Betaxolol (0.1 mg/kg) was injected i.v. 30 min after partial occlusion. Symbols are the same as in Fig. 1 (n=6).}
\end{figure}
7 to 106±8 beats/min (-27%) and (+)dp/dt from 2333±192 to 1830±123 mmHg/sec (-21%).

Because betaxolol decreased the heart rate as described above, an attenuation of myocardial acidosis by betaxolol could result from a decrease in cardiac work. Therefore, we examined the effect of betaxolol (0.1 mg/kg) on myocardial acidosis in the paced hearts. As shown in Fig. 3, betaxolol attenuated the ischemia-induced myocardial acidosis even when the heart rate was fixed at 166±6 beats/

Fig. 3. Effect of betaxolol on ischemic myocardial pH in the paced hearts. In these experiments, the hearts were driven electrically at the constant rate of 166±6 beats/min. Betaxolol (0.1 mg/kg) was injected i.v. 30 min after partial occlusion. Symbols are the same as in Fig. 1 (n=6).
min by electric pacing; the pH was restored from 6.82 to 7.23 60 min after the injection of betaxolol.

**Atenolol experiments:** The effects of atenolol (0.1 mg/kg) on LAD flow, myocardial pH, ST segment of epicardial ECG, (+) dp/dt, heart rate and blood pressure in the ischemic hearts are shown in Fig. 4. Myocardial pH, that had been decreased by partial occlusion from 7.51 to 6.87, increased to 7.24 60 min after the injection of atenolol. Atenolol decreased heart rate from 163±11
Propranolol experiments: The effects of propranolol (0.1 mg/kg) on the ischemic hearts are shown in Fig. 5. Propranolol attenuated the myocardial pH during ischemia; the pH was restored from 6.91 to 7.23 60 min after the administration of the drug. Propranolol decreased heart rate from 152±13 to 127±4 beats/min (-21%) and (+)dp/dt from 2540±370 to 1732±109 mmHg/sec (-29%).

**Fig. 5.** Effect of propranolol on ischemic myocardial pH in dogs. Propranolol (0.1 mg/kg) was injected i.v. 30 min after partial occlusion. Symbols are the same as in Fig. 1 (n=5).
Comparison of the effects of betaxolol, atenolol and propranolol on myocardial acidosis and the left ventricular function in the ischemic hearts: For comparison of the potency of the drugs on myocardial acidosis, the effect of betaxolol (0.01, 0.03 or 0.1 mg/kg), atenolol (0.03 or 0.1 mg/kg) or propranolol (0.03 or 0.1 mg/kg) was evaluated. The data of heart rate, (+)dp/dt and myocardial acidosis are summarized in Fig. 6. Betaxolol at the dose of 0.01, 0.03 or 0.1 mg/kg significantly restored the myocardial [H+] that had been increased by partial occlusion by 46±3, 60±8 or 66±4%, respectively. The restoration caused by 0.03 or 0.1 mg/kg of atenolol was 29±11 or 55±5%, respectively; and that by 0.03 or 0.1 mg/kg of propranolol was 41±3 or 50±8%, respectively. Therefore, when compared in the dose restoring myocardial [H+] by 50%, betaxolol was about 3 times more potent than atenolol and propranolol on a weight-to-weight basis.

All the drugs decreased heart rate and (+)-dp/dt. However, the effect of betaxolol or propranolol on myocardial contractile force was

![Graphs showing data for heart rate, (+)dp/dt, and myocardial acidosis comparison between saline, betaxolol, atenolol, and propranolol.](image-url)
less potent than that of atenolol at the dose producing the equivalent restoration of myocardial acidosis; the decrease in \((+\)dp/dt\) caused by betaxolol (0.03 mg/kg), atenolol (0.1 mg/kg) or propranolol (0.1 mg/kg) was 17±3, 29±6 and 18±4%, respectively, whereas the decrease in heart rate by the drugs was 19±1, 21±3 or 20±4%, respectively.

**Discussion**

The present study has shown that betaxolol attenuates the ischemic myocardial acidosis induced by partial occlusion of LAD in anesthetized dogs. Since the accumulation of \([H^+]\) has been demonstrated to correlate well with an increase in lactate and with decreases in ATP and creatine phosphate in the ischemic myocardium (9), the tissue pH may reflect the metabolic state (aerobic or anaerobic) and the depletion of high energy phosphates in the myocardium. Therefore, the results of the present study suggest that betaxolol improves myocardial metabolism in the ischemic myocardium. In fact, this may be supported by our biochemical evidence that betaxolol inhibited the accumulation of lactate and the depletion of high energy phosphates due to coronary ligation in dogs (Y. Abe et al., unpublished data). Taken together, these findings indicate that betaxolol reduces the severity of myocardial ischemic injury. The reduction of heart rate and \((+\)dp/dt\) lowers cardiac work, leading to a decrease in oxygen consumption of the hearts. These effects of betaxolol would be responsible for the attenuation of myocardial acidosis during ischemia. However, the findings that betaxolol attenuated the ischemia-induced myocardial acidosis, even when heart rate was fixed by electric pacing, suggest that the beneficial effect of betaxolol on the ischemic myocardium is not primarily due to the decrease in heart rate.

In the present study, betaxolol, atenolol and propranolol have been demonstrated to inhibit the ischemia-induced myocardial acidosis in dogs. The results of the present study are in agreement with those by Abiko and his co-workers (12), who demonstrated that beta-adrenoceptor antagonists attenuate myocardial acidosis most effectively. However, betaxolol was about 3 times more potent than atenolol or propranolol on a weight-to-weight basis when compared in the dose producing 50% restoration of myocardial acidosis. This is probably results from the potency and duration of beta\(_1\)-adrenoceptor antagonistic activity, since betaxolol has the most potent, selective and long lasting beta\(_1\)-adrenoceptor antagonistic activity among the drugs (3). Furthermore, the contribution of beta\(_1\)-adrenoceptors, but not beta\(_2\)-adrenoceptors, to the ischemic myocardial acidosis has been proposed by Sakai and Abiko (13). These findings suggest that the main mechanism responsible for the beneficial effects of the drugs on ischemic myocardium is the beta\(_1\)-adrenoceptor antagonistic property, although the possible role of the membrane stabilizing action of some beta-adrenoceptor antagonists cannot be excluded.

Although the decrease in myocardial oxygen demand through the decrease in cardiac work would be responsible for the anti-ischemic effects of the drugs, the negative inotropic effects of beta-adrenoceptor antagonists may limit their usefulness for patients with left ventricular dysfunction. However, the present experiments have shown that the depression of contractile force by betaxolol was less potent than that by atenolol and equivalent with propranolol, when compared in the equivalent dose improving myocardial acidosis (approximately 50%). Recently, Satoh et al. also have found that the negative inotropic effect of betaxolol is less potent than that of atenolol or propranolol in anesthetized dogs (unpublished data). This property of betaxolol may be beneficial in the treatment of ischemic heart diseases. In fact, clinical study demonstrated that betaxolol reduced angina frequency and improved exercise capacity without any adverse effects on the left ventricular ejection fraction even in patients with mild to moderate left ventricular dysfunction (15).

In conclusion, these results have confirmed that beta-adrenoceptor antagonists attenuate ischemia-induced myocardial acidosis and have shown that among three beta-adrenoceptor antagonists, betaxolol is the most effective in improving myocardial acidosis with a relatively weak effect on myocardial
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