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EFFECTS OF β_1- AND $\beta_1+\beta_2$-ANTAGONISTS ON TRAINING-INDUCED MYOCARDIAL HYPERTROPHY AND ENZYME ADAPTATION

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Abstract—The effects of β_1- and $\beta_1+\beta_2$-antagonists on the myocardial adaptation to exercise training were investigated in male Sprague–Dawley rats randomly divided into: trained (treadmill, 1 hr/day, 5 days/week for 10 weeks at 27 m/min, 15% grade) without drug (TC), sedentary without drug (SC), trained treated with atenolol (TA) (10 mg/kg body wt, i.p.), trained treated with propranolol (TP, 30 mg/kg body wt, i.p.), and sedentary propranolol. Doses of both β-antagonists were titrated to decrease the exercise heart rate by 25% compared to the controls. The heart weight and heart/body weight ratio were significantly greater in TC (1.28 ± 0.07 g ($P < 0.01$); 296 ± 12 mg/100 g body wt ($P < 0.05$) respectively) than in SC (1.09 ± 0.04 g and 268 ± 11 mg/100 g body wt), or in TP and TA. Myocardial mitochondrial protein was unchanged by training or β-blockade. Citrate synthase and β-hydroxyacyl CoA dehydrogenase activities were not altered. Carnitine palmitoyltransferase activity was increased in SP compared to SC. Training increased hexokinase activity only in TC (5.22 ± 0.12 vs 4.26 ± 0.23 μmol/min/g wet wt, $P < 0.01$). Lactate dehydrogenase activity increased significantly ($P < 0.01$) in both TC (383 ± 14 μmol/min/g wet wt) and TA (372 ± 14 μmol/min/g wet wt) compared to SC (276 ± 14 μmol/min/g wet wt), but not in TP versus SP. These data indicate that (1) β-adrenergic blockade prevents training-induced cardiac hypertrophy; (2) β-antagonists have little effect on the myocardial oxidative capacity; and (3) while the training induction of myocardial hexokinase is inhibited by both β_1- and $\beta_1+\beta_2$-antagonists, myocardium may increase its ability to utilize lactate during exercise with training despite β_1-blockade.

Beta-adrenergic receptor blockade is known to alter the hemodynamic responses to both acute exercise and chronic training [1–5]. There is also evidence that β-blockade, at least the non-cardiac-selective β-blocking agent, propranolol, affects the biochemical adaptation to training in skeletal muscle [6–8]. However, relatively little is known about the effects of β-antagonists on myocardial morphological and metabolic functions adapting to an exercise training program, which is of great importance in cardiac patients.

Compensatory hypertrophy has been documented by numerous investigators as the main cardiac morphological change following exercise training [9–11]. Some investigators found that, when β-blocking drugs were administered in association with exercise training in the rat, cardiac hypertrophy did not occur [6, 12]. It has been suggested that cardiac sympathetic innervation plays a critical role in eliciting cardiac hypertrophy [6, 13], but controversy still exists [14] because it is experimentally difficult to separate the stimulation to myocardium by increased volume/pressure work from sympathetic activity associated with exercise.

The effect of physical training on heart mitochondria has been studied extensively, and all investigators agree that there is little change in myocardial oxidative capacity after training [10, 11, 15, 16]. It appears that the myocardium possesses sufficient oxidative enzyme activity to meet the energy demands during exercise. However, an increased turnover of fatty acid through the triglyceride pool has been observed in trained rat heart [17], although a key enzyme in fatty acid metabolism, β-hydroxyacyl CoA dehydrogenase (HADH), was reported unchanged after training [6]. Furthermore, marked increases in resting glycogen stores and glycogen synthase activity in trained guinea pig heart [18] and elevated pyruvate kinase and lactate dehydrogenase activities in rat heart [19] have been reported. Because adipose tissue lipolysis and myocardial glycolysis are largely controlled by sympathoadrenergic systems [20, 21], it is of great interest to know whether β-blocking agents affect the training-induced myocardial adaptation of these enzymes. Furthermore, energy substrates for heart are supplied mainly from exogenous sources, i.e. blood-borne fatty acids, glucose, lactate and probably also ketone bodies [22]. β-Adrenergic blocking drugs prevent elevation of free fatty acids (FFA) in blood and cause hypoglycemia due to an increase in the glucose uptake by the exercising skeletal muscle without compensating increased gluconeogenesis [23]. This altered blood substrate profile may also affect myocardial metabolic functions.

In the present investigation, we administered a cardiac-selective β_1-blocking agent, atenolol, and the $\beta_1+\beta_2$-blocker, propanolol, to antagonize the β-

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adrenergic receptors in rat heart. The dose levels of the drugs were selected by titration to decrease exercise heart rate (EHR) by 25% at treadmill speed of 27 m/min, 15% slope as compared with the untreated control rats. The absolute and relative heart weights, and the myocardial mitochondrial protein content were used as an index of the morphological changes following exercise training with or without β-blockade. Enzymes involved in the myocardial tricarboxylic acid cycle, fatty acid oxidation and glycolysis were measured to evaluate the metabolic adaptations of the heart to training with differing sympathoadrenergic influences, and possibly an altered blood substrate profile.

METHODS

Animal care and training program. Male Sprague–Dawley rats (Sasco, Oregon, WI), age 2 months, body wt 250–275 g, were used in this experiment. Animals were housed 2–3 per cage in a temperature-controlled room (22°) with a dark–light cycle of 12–12 hr, and were maintained with Purina rat chow and tap water *ad lib.* On the day of arrival, the animals were assigned randomly into five experimental groups: chronically trained and treated with a $\beta_1+\beta_2$-adrenergic blocking agent, propranolol (TP); trained and treated with a β_1-selective agent, atenolol (TA); trained without β-blocker (TC); sedentary without drug treatment as controls (SC); and sedentary treated with the same dose of propranolol as the TP group (SP) to evaluate the chronic effects of β-blocker itself. Food intake of the SC and SP groups was restricted to match the body weight gain of their respective trained counterparts.

The exercise training was performed on two motor-driven rodent treadmills at the beginning of the dark period, 5 days per week for 10 weeks, at a speed of 27 m/min (1.0 mph). The details of the training program have been reported previously [7]. Propranolol-treated rats had some difficulty performing treadmill running during the first 3 weeks but were subjected to the same training protocol as the non-drug controls. Under constant and careful monitoring and progressive training, these rats gradually adjusted to and accomplished the training sessions effectively without necessarily receiving more electrical shocking. To control the effect of routine handling, the control groups (SC and SP) also ran on the treadmills at 15 m/min, 0% slope for 5 min, scheduled at the same time of day as the training groups.

Administration of β-adrenergic blocking agents. The dose levels of β-blocking drugs used in previously reported experiments varied substantially. We therefore did a titration experiment to determine the doses of the non-selective and the cardiac-selective drugs required to attenuate the EHR during the training period. The titration experiment was conducted 2 weeks after the onset of the training program during which low doses of β-blocking drugs (propranolol, 5 mg/kg body wt; atenolol, 2.5 mg/kg body wt, in 0.9% saline) were injected intraperitoneally (i.p.) into TP and TA respectively. Heart rate (HR) was measured by inserting three sterilized brass electrodes subcutaneously on both sides of the chest and the back of the neck of the rat, which conducted the cardiac electrical impulses of the animal by wire leads to an electrocardiograph and a multichannel recorder. After the resting HR was observed stable, the animals performed a progressive exercise test during which EHR was recorded. The details of the titration experiment have been described elsewhere [7]. The results of the dose–response curves are illustrated in Fig. 1. The EHR responses to progressive treadmill running were approximately linear both in the non-drug rats and either propranolol or atenolol blocked rats. It was found that 10 mg atenolol/kg body wt and 30 mg propranolol/kg body wt reduced the EHR to a similar extent, i.e. 25% less than the pre-drug rate at 27 m/min, 15% grade. Rats treated with 30 and 50 mg propranolol/kg body wt and 30 mg atenolol/kg body wt could not complete the test when treadmill speed exceeded 27 m/min and their EHR was reduced by only an additional 5–10 bpm. In addition, it was observed that animals given these higher doses could not run effectively on the treadmill during the 16-min titration experiment and obviously would not be able to finish the 1-hr training session.

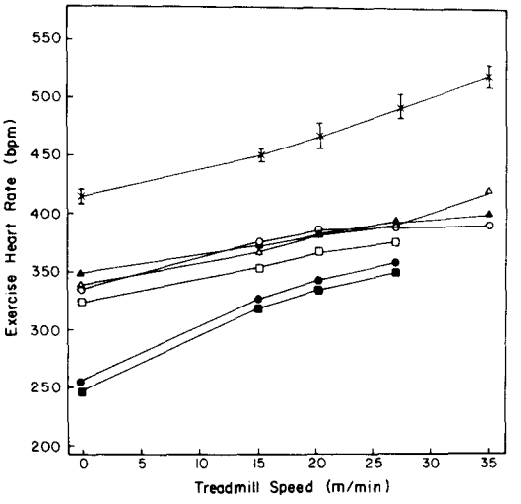

Fig. 1. Exercise heart rate response to a graded treadmill test at various speeds (meters/min at 15% grade) in rats on β_1- and $\beta_1+\beta_2$-antagonists. Key: control (mean ± SD) (×); propranolol; (▲) 10 mg/kg body wt, (●) 30 mg/kg body wt, and (■) 50 mg/kg body wt; atenolol; (△) 5 mg/kg body wt, (○) 10 mg/kg body wt, and (□) 30 mg/kg body wt. Heart rate was measured at min 4 of each workload.

Thus, in the following 8 weeks of the training, the TA group received atenolol, 10 mg/kg body wt, and the TP and SP groups received propranolol, 30 mg/kg body wt, 5 days per week. Drugs were injected i.p. approximately 30 min before each training session; the TC and SC groups received 1 ml of saline (placebo). To examine the effectiveness of the β-blockade in attenuating EHR, a week after drug administration (week 3 of the training), TC, TA and TP groups of rats were run on the treadmills for 1 hr according to the training protocol. EHR was recorded every 5 min during the entire exercise period. The results showed that, at the first 20 min,

TP rats had slightly lower EHR compared to the TA rats, but the difference diminished as exercise proceeded. There was no difference in EHR between the TP and TA after 30 min [7]. Both atenolol and propranolol reduced the EHR by 25% as compared to the controls ($P < 0.01$). This procedure was also periodically performed in the remaining 7 weeks to ensure the effectiveness of the β-blockade.

Tissue preparation and enzyme assays. All animals were killed 60 hr after their last training sessions in a resting, fed and non-medicated state; the sedentary control animals were killed at the same time of the day. To minimize day-to-day variation, one animal from each group was killed each day consecutively.

After decapitating and exsanguinating the rats, the abdominal cavity was opened and the heart was excised. The heart was rinsed, blotted, trimmed of the aorta as well as other vessels, and weighed. The ventricles and atria were cut along the vertical axis of the heart into two parts for mitochondrial preparation and enzyme assays respectively. One part was minced and homogenized at 0° in a medium containing 0.25 M mannitol, 70 mM sucrose and 1 mM EDTA (pH 7.4) with a motor-driven Potter–Elvehjem Teflon glass homogenizer at 350 rpm. The homogenate was centrifuged at 480 *g* for 5 min. The supernatant fraction was removed, and the pellet was resuspended in the original homogenization medium and recentrifuged at 480 *g* for 5 min. The two supernatant fractions were combined and centrifuged at 7700 *g* for 5 min. The resulting mitochondrial pellet was suspended in 0.25 M sucrose, 2 mM EDTA (pH 7.4) and frozen at −30° until mitochondrial enzyme assays were performed. The other half of the heart was frozen between plates pre-cooled with liquid nitrogen and stored at −160°. It was later homogenized in a medium containing 10 mM 4-(2-hydroxyethyl)-1-piperazine-ethanesulfonic acid (HEPES), 2 mM EDTA (pH 7.4) for glycolytic enzyme assays.

The mitochondrial suspensions were frozen (dry-ice, alcohol) and thawed three times to release the total membrane-bound enzyme activity. Myocardial citrate synthase (CS, EC 4.1.3.7) activity was determined spectrophotometrically at 25° according to Shepherd and Garland [24]; carnitine palmitoyltransferase (CPT I+II, EC 2.3.1.21) activity was assayed at 30° by the isotope exchange method of Norum [35] as modified by McGarry and Foster [26]. HADH activity was measured at 25° according to Essen *et al.* [27]. The recorded counts per minute (cpm) of the formed tritiated palmitoylcarnitine were used to express the enzyme activity.

For assay of hexokinase (HK, EC 2.7.1.1.) [28], the heart homogenate was treated with Triton X-100 (1%, v/v) to release the cell membrane-bound enzyme activity. Lactate dehydrogenase (LDH, EC 1.1.1.27) activity was determined according to Bergmeyer and Bernt [29]; myocardial mitochondria and total protein content were measured using the method of Bradford [30].

All chemical reagents and enzymes were of the highest purity available. Atenolol was a gift from Stuart Pharmaceutical (Wilmington, DE).

Statistics. Data were analyzed using the Planned Comparison method. The α level was 0.05.

RESULTS

Chronic administration of propranolol, a non-selective β-blocking drug, significantly attenuated the gain of body wt in the trained rats, whereas the β_1-selective blocker, atenolol, did not affect the growth (Table 1). As also shown in Table 1, the chronically trained rats had significantly greater heart mass (+17%, $P < 0.01$) and heart/body wt ratio (+10%, $P < 0.05$) compared to the sedentary controls. This training-induced cardiac hypertrophy was not observed in the trained rats treated with either propranolol or atenolol. Although statistically non-significant, propranolol treatment also slightly decreased the heart weight in the sedentary rats as compared to sedentary controls.

Table 2 depicts the rat myocardial mitochondrial protein and the total myocardial protein content measured in crude homogenate. Exercise training or β-blockade did not affect the protein content to any appreciable extent. Propranolol treatment seemed to decrease the mitochondrial protein somewhat, but it was not significant.

The activities of heart mitochondrial CS and two enzymes involved in fatty acid oxidation, HADH and CPT, are shown in Table 3. There was no significant difference between the trained and untrained rats or between the β-blocked and control rats in CS and HADH activities. Chronic treatment with propranolol significantly induced the myocardial CPT activity in the sedentary rats, as indicated by a 26% ($P < 0.05$) increase in the SP group versus SC.

Whereas exercise training essentially had no influence on the myocardial oxidative enzymes, it dramatically affected the myocardial HK and LDH activities, as demonstrated in Table 4. HK activity in the trained hearts (TC) was approximately 23% ($P < 0.01$) higher than in sedentary controls. However, the training influence on HK was inhibited by both β-adrenergic blockers propranolol and atenolol. Interestingly, propranolol treatment alone seemed also to increase the HK slightly, but it was not significant.

Table 1. Body weight, heart weight and heart/body weight ratio in the rat

		Body wt (g)	Heart wt (g)	Heart/Body wt ratio (mg/100 g body wt)
Trained				
Control	(9)	432 ± 9	1.28 ± 0.07*	296 ± 12†
Propranolol	(8)	368 ± 14‡	1.03 ± 0.05	278 ± 7
Atenolol	(8)	401 ± 17	1.14 ± 0.08	271 ± 12
Sedentary				
Control	(9)	417 ± 13	1.09 ± 0.04	268 ± 11
Propranolol	(7)	371 ± 11	1.04 ± 0.05	281 ± 11

Values are means ± SEM. The number of animals is given in parentheses.

* $P < 0.01$, trained control vs sedentary control.

† $P < 0.05$, trained control vs sedentary control.

‡ $P < 0.05$, trained propranolol vs trained control.

Table 2. Myocardial protein content in the rat

		Mitochondrial protein (mg/g wet heart)	Total myocardial protein (mg/g wet heart)
Trained			
Control	(9)	15.4 ± 2.2	220 ± 12
Propranolol	(8)	13.2 ± 1.4	196 ± 9
Atenolol	(8)	14.7 ± 1.6	222 ± 15
Sedentary			
Control	(9)	15.5 ± 1.5	239 ± 21
Propranolol	(7)	13.7 ± 1.4	204 ± 11

Values are mean ± SEM. The number of animals is given in parentheses.

Table 4. Myocardial glycolytic enzyme activities in the rat

		Hexokinase (μmol/min/g wet wt)	Lactate dehydrogenase (μmol/min/g wet wt)
Trained			
Control	(6)	5.22 ± 0.12*	383 ± 14*
Propranolol	(6)	4.47 ± 0.32	255 ± 16
Atenolol	(6)	4.56 ± 0.21	372 ± 14*
Sedentary			
Control	(6)	4.26 ± 0.23	276 ± 14
Propranolol	(6)	4.86 ± 0.23	315 ± 28

Values are mean ± SEM. The number of animals is given in parentheses.
* $P < 0.01$, trained control or trained atenolol vs sedentary control.

LDH activity was increased 40% by training ($P < 0.01$) in the TC group, and also in the TA group by a similar magnitude. However, this adaptation was completely absent in the trained rats treated with propranolol. In fact, the LDH activity in TP was even lower compared to SP, although only a trend ($0.05 < P < 0.10$) existed.

DISCUSSION

Effect of β-blocking drugs on EHR. Figure 1 illustrates that EHR of the β-blocked rats increased proportionally with increased workloads as was the case for normal rats. However, the increment was smaller as compared to the non-drug rats. At the assigned training intensity (27 m/min, 15%), the increase in EHR in the β-blocked groups represented only a fraction of that of the controls. Yet, with adjustment of the circulatory functions the rats were still capable of sustaining the 1-hr treadmill running. In the present investigation, the dose levels of the selective β_1-blocker atenolol and the non-selective β-blocker propranolol were titrated and verified by a later experiment such that the EHR of the β-blocked rats was decreased substantially to the same extent by propranolol and atenolol. This procedure has taken into consideration the different β-antagonizing potentials of various drugs [31–33]. Although we did not measure other cardiovascular indices, the EHR has provided useful information as to how the heart responds to submaximal exercise under normal conditions and under β-adrenergic blockade. The only study we are aware of that measured the rat's EHR as an index of β-blockade was conducted by Mullin *et al.* [12]. By injecting rats with atenolol, 100–150 mg/kg body wt i.p., they reduced the EHR of the trained rats by 25%. This dose was substantially higher than the doses we used in the present experiment (10 mg atenolol/kg, 30 mg propranolol/kg) to achieve a similar reduction of EHR. However, the training program they used was also more strenuous (27 m/min, 25%) compared to ours; therefore, a higher dose of β-blocking drug might be required.

Cardiac hypertrophy. Compensatory cardiac hypertrophy induced by chronic exercise training has been well documented and was discussed thoroughly by Schaible and Scheuer [11] in a recent review. Our finding that 10 weeks of treadmill running caused a significant increase in both heart mass and heart/body ratio in adult male rats is in general agreement with other investigators [9, 10, 34, 35]. However, it is noteworthy that in this experiment we restricted the food intake of the sedentary control animals to match the body wt gain in the trained rats, and this, according to some authors [36], could have affected the growth of the heart. Thus, the magnitude of cardiac enlargement induced by training may have been somewhat exaggerated. Nevertheless, the significantly increased heart/body weight ratio associated with training verified the prominent effect of chronic physical exercise on heart growth.

Table 3. Heart mitochondrial enzyme activities in the rat

		Citrate synthase (μmol/min/mg protein)	β-Hydroxyacyl CoA dehydrogenase (μmol/min/mg protein)	Carnitine palmitoyltransferase (cpm/mg protein)
Trained				
Control	(6)	1.75 ± 0.3	0.49 ± 0.03	1024 ± 42
Propranolol	(6)	1.56 ± 0.4	0.57 ± 0.03	972 ± 100
Atenolol	(6)	1.69 ± 0.2	0.49 ± 0.06	968 ± 100
Sedentary				
Control	(6)	1.68 ± 0.3	0.51 ± 0.05	908 ± 52
Propranolol	(6)	1.52 ± 0.3	0.56 ± 0.05	1142 ± 86*

Values are mean ± SEM. The number of animals is given in parentheses.
* $P < 0.05$, sedentary propranolol vs sedentary control.

One of the major findings of the present investigation was that chronic administration of the β-antagonists, propranolol and atenolol, can abolish the training-induced myocardial hypertrophy. This should not be surprising if one considers that both atenolol and propranolol substantially decreased the EHR of the β-blocked rats. In fact, the EHR in the β-blocked rats was even lower than the resting heart rate of the sedentary controls. Therefore, the physical stimulus on hearts of the β-blocked rats was reduced markedly during the 10-week training period. It is possible that coronary blood flow and myocardial oxygen consumption ($M\dot{v}O_2$) need to reach a critical level to produce hypertrophy [11]. Since EHR was decreased 25% by β-blocking drugs, coronary flow and $M\dot{v}O_2$ were also decreased and this could explain the failure of a training-induced cardiac hypertrophy. There is considerable controversy as to whether an increase in volume/pressure work of the heart or the functional sympathoadrenergic stimulation is more essential for a training-induced cardiac hypertrophy to occur. Some investigators [6, 13] suggested that the increased sympathetic stimulation associated with exercise, rather than the increased volume and/or pressure work *per se*, was the main requirement that evoked cardiac hypertrophy. This hypothesis was supported by the evidence that chronic treatment of rat heart with a β-agonist (isoprenaline) without exercise could cause hypertrophy [37, 38], whereas chemical sympathectomy with guanethidine or long-term treatment with propranolol prevented the exercise-induced cardiac hypertrophy in the rat [6, 39]. However, these authors did not measure the EHR or other cardiac performance indices of the rats during training (swimming), and therefore no conclusion could be drawn from these experiments. In the present investigation, although EHR was attenuated substantially by β-blocking drugs, stroke volume could increase to compensate for the impaired cardiac rate in order to deliver oxygen and energy substrate to the working muscle during exercise [2]. Cardiac output and total myocardial work during 1-hr treadmill running invariably increased above the resting levels in the β-blocked rats. In the present study, we did not measure systemic blood pressure and stroke volume of the rat; therefore, the quantitative work performed by the β-blocked heart was unknown. Nevertheless, the fact that the heart/body weight ratio did not change at all in a period of 10 weeks of training provides strong evidence that sympathoadrenergic stimulation is required to produce cardiac hypertrophy.

Mitochondrial oxidative capacity. It is generally agreed that physical training has little effect on cardiac muscle mitochondria, either morphologically or metabolically [15, 40], although some controversy still exists. For example, Penpargkul *et al.* [41] reported an increase in mitochondrial protein without apparent cardiac hypertrophy in male rats trained by swimming, whereas Oscai *et al.* [40] reported no differences in mitochondrial protein in hearts of trained and untrained rats. Hickson *et al.* [9] found that cytochrome *c*, a mitochondrial marker, did not change in trained, hypertrophied or non-hypertrophied rat hearts. Data in this experiment have provided no evidence that mitochondrial protein or total myocardial protein content alters with intensive treadmill training in the male rat. Neither did our data show any appreciable differences in regard to the effect of β-adrenergic blockade on protein content in the heart, which is in agreement with the results of Harri [6]. It also verified that the prominent differences in heart size among various treatment groups of rats were not of abnormal composition.

The results in the present investigation indicating that heart mitchondrial enzyme marker CS, and the rate-limiting enzyme in fatty acid oxidation, HADH, did not change after training with or without β-blockade support earlier conclusions by other authors [6, 9–11] that myocardium has sufficient pre-existing oxidative capacity to supply the energy requirement during exercise. In a previous investigation [7], we reported that the training induction of skeletal muscle mitochondrial CS and HADH activities could be prevented by propranolol but not atenolol, providing some evidence that the β_2-adrenergic system is involved in local enzymatic adaptation to chronic exercise. This was certainly not the case in the heart. Despite the severely suppressed EHR during the entire training period, the β-blocked myocardium showed no alteration in these two key enzymes in the aerobic metabolism of heart.

One surprising finding in this study was that the activity of CPT, which controls the transport of long-chain fatty acid across the mitchondrial membrane, increased significantly in the propranolol-treated sedentary rats. We are not aware of any direct regulatory mechanism of CPT by the sympathoadrenergic pathway in the heart. It is known that perfusion of rat heart with lactate can lead to an activation of acetyl CoA carboxylase and thus raise the concentration of malonyl CoA [42], which is a strong inhibitor of CPT [43]. The moderate change in blood lactate under β-blockade in sedentary subjects is too small to explain the significant increase in CPT activity in the propranolol-treated rats. However, β-blocking drugs are known to inhibit myocardial glycolysis and to decrease blood fatty acid levels [21] which tend to reduce the acetyl CoA level in the heart. Whether or not the intracellular malonyl CoA level was also reduced by β-blocking drugs is not known at present.

Glycolytic enzymes. While the myocardial oxidative capacity has been investigated extensively, relatively little literature deals with adaptation of the glycolytic enzymes to physical conditioning. Lamb *et al.* [18] showed that glycogen synthetase activity was increased in the hearts from trained guinea pigs. Higher resting glycogen stores in the trained rat heart have also been reported [44]. York *et al.* [19] studied several glycolytic enzymes in rat heart and found that pyruvate kinase and lactate dehydrogenase activities were increased substantially after physical conditioning. Our data revealing a dramatic training adaptation in myocardial HK and LDH suggested that heart muscle adapts to chronic overload by increasing its glycolytic capacity, which is in direct contrast to skeletal muscle.

The training-induced myocardial LDH activity is of particular metabolic importance. In the heart, high mitochondrial enzyme activities and a greater

tricarboxylic acid cycle turnover rate make it possible for a faster removal of pyruvate formed from either phosphoenolpyruvate or lactate. Since the lactate concentration is increased during severe exercise, an elevated lactate dehydrogenase activity may facilitate lactate extraction from circulating blood as a source of energy via oxidative pathways [45, 46]. York *et al.* [19] reported that this increased LDH activity in the heart is due to an increase in the M-subunits of the enzyme. We have shown previously that training not only increases the maximal activity of myocardial LDH, but also decreases its Michaelis constant (K_m) for lactate, thereby facilitating the uptake of lactate by myocardium [47]. These changes may represent a metabolic adaptation of the heart to chronic exercise.

During prolonged exercise under either selective or non-selective β-adrenergic blockade, fatty acid turnover is severely impaired due to an anti-lipolytic effect of the drugs [21]; skeletal muscle has to shift the energy substrate from fat to blood-borne glucose. Therefore, a hypoglycemic tendency occurs sooner or later [23]. Some authors showed that, in humans, propranolol inhibits muscle glucogenolysis and hence lactate release during dynamic exercise [48, 49]. In contrast, β_1-selective blockers atenolol and metoprolol have little effect on the exercise-raised blood lactate levels [50, 51]. If the blood lactate concentration was also lower in the TP group of rats than in TA and TC during training sessions in the present experiment, the discriminative effects of he two β-blocking drugs on the myocardial LDH adaptation could be explained by the inability of the TP rats to utilize lactate during exercise.

In conclusion, the results of the present experiment indicated that β-adrenergic blocking agents abolish cardiac hypertrophy caused by chronic exercise training but have little effect on myocardial oxidative enzymes. While both β_1– and $\beta_1+\beta_2$-antagonists inhibited the increase of myocardial HK activity with training, only the non-selective drug prevented the training adaptation of LDH which may be important for the heart to utilize lactate during exercise.

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