Intrinsic Rate and Cholinergic Sensitivity of Isolated Atria from Trained and Sedentary Rats[1] (37592)

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One of the characteristic changes in the mammalian cardiovascular system associated with endurance training is a decrease in the resting heart rate (1–3). Although several mechanisms have been proposed to account for this bradycardia, its origin is still not known. It has been suggested that an increase in tonic cardiac vagal activity in the trained mammal may be a primary factor (4–6).

Experiments in man and other mammals using the cholinergic antagonist atropine have yielded inconclusive results. Herxheimer (7), studying human subjects, failed to bring about a complete parasympathetic block. The results of Kauf (8) and Slapak (9) are difficult to assess since neither presented adequate control data. Raab *et al.* (5) noted only a small difference between the cardio-acceleration in trained and untrained men after intravenous injection of atropine sulfate. Tipton and Taylor (10) reported that an intraperitoneal injection of atropine sulfate elicited a smaller cardio-acceleration in trained than in untrained rats. This result was attributed to a greater availability of atrial nonneural acetylcholine to compete with the atropine at the cholinergic receptor sites in the trained rats. Unfortunately they did not examine the effect of larger doses of atropine which may have been capable of establishing a complete cholinergic block.

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Several possible intracardiac mechanisms have been proposed for the bradycardia of training. It has been suggested that there might be a greater release of nonneural acetylcholine in the trained animal (10). Hall (11) considered that changes in atrial stretch resulting from atrial distension and hypertrophy, might modify the normal sinoatrial rhythm.

If there is, in the trained mammal, an increase in the tonic cardiac parasympathetic activity, it is likely to be accompanied by a change in the sensitivity of the sinoatrial node to acetylcholine. In this report we present evidence of a decrease in the intrinsic atrial rhythm and a reduction of the sensitivity of the sinoatrial node to acetylcholine in physically exercised rats.

Methods. Male rats of the Wistar strain (120–150 g) were randomly assigned to a sedentary and an exercised group. All rats were kept in environmentally controlled quarters and allowed rat chow and water *ad libitum*. Exercised animals were swum for 1 hr/day, 5 days/wk, for 15 wk. Plastic buckets (13 in. in diameter, 24 in. deep) were utilized as swimming tanks (4 rats/tank) with water temperature kept between 32 and 34°. Animals in the sedentary group were periodically handled prior to the final experiments.

During week 14 of training the body weights and resting heart rates (12) of both sedentary and exercised animals were determined. Animals were sacrificed by a blow to the head and decapitation. The heart was rapidly removed to a dish containing Krebs–Henseleit solution and the atria were carefully dissected free, ensuring that the sinoatrial node was intact. Following transfer to

an organ bath containing Krebs–Henseleit solution at 30°, aerated with a mixture of 95% oxygen and 5% carbon dioxide, the atria were attached to a strain gauge (Grass force-displacement transducer FT10C), and diastolic tension was adjusted so that the preparation was contracting at the peak of its length–tension curve (13). Contractions were recorded using a Grass Model 7 polygraph.

The preparations were allowed to stabilize for at least 1 hr. When a steady rhythm was established, the atrial rate was determined over a 30 sec period. This will be referred to as the "intrinsic atrial rate." A known amount of acetylcholine was then added to the bath. (Acetylcholine was prepared from the iodide in deionized water.) Acetylcholine was added in amounts which gave final bath concentrations of 10^{-7} to 10^{-4} *M*. Since it was found that the atria attained a new and steady rate between 2 and 2.5 min after addition of acetylcholine, the rate over this 30 sec period was used in determining the response. The response was calculated in terms of the percentage change in atrial rate upon adding the drug (*i.e.*, percentage of the maximal response). Immediately following a test, the preparations were washed twice and allowed to equilibrate to the control rate. Dose–response curves were then constructed.

The differences between the means of the body weights, resting heart rates, intrinsic atrial rates, and atrial chronotropic responses to acetylcholine, of the exercised and sedentary groups were tested using Student's *t* test.

Results. Body weights of sedentary and exercised animals (week 14) were 475 ± 10 g and 411 ± 7 g, respectively ($p < 0.01$). Exercised rats had significantly lower resting heart rates than sedentary rats (Table I). Also the atria from exercised rats exhibited lower intrinsic rates than atria from sedentary rats (Table I).

TABLE I. Resting Heart Rates and Intrinsic Atrial Rates of Exercised and Sedentary Rats.

	Sedentary ($n = 12$)	Exercised ($n = 10$)
Resting heart rate (beats/min)	318 ± 7[a]	288 ± 6[b]
Intrinsic atrial rate (beats/min)	146 ± 5	128 ± 3[b]

[a] Mean ± SEM.

[b] Significantly different from the sedentary value $p < 0.01$.

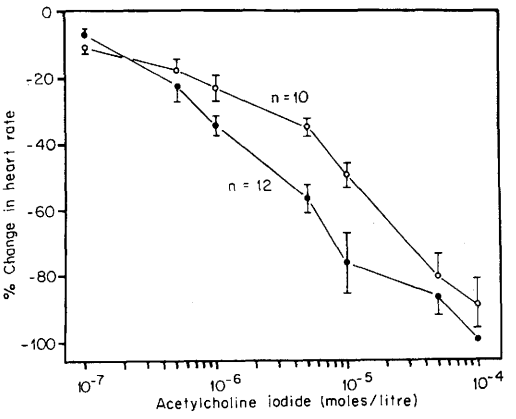

FIG. 1. The chronotropic influence of acetylcholine on rat atria; (○—) exercised animals; (●—) sedentary animals.

The dose–response curves of the preparations from exercised and sedentary groups are shown in Fig. 1. Preparations from exercised animals demonstrated a sensitivity approximately one-fifth that of the preparations from sedentary animals in the region from 25 to 80% of the maximal response. The difference between the responses of atrial preparations from exercised and sedentary animals was significant at acetylcholine concentrations of 10^{-6}, 5×10^{-6} and 10^{-5} *M* ($p < 0.05$, $p < 0.01$, $p < 0.01$).

Discussion. Rats engaged in an endurance training programme gain weight more slowly than sedentary rats (Table I). This is probably due to both a higher caloric output and a lower food intake in the exercising rat (14–16).

A complete autonomic blockade of the heart can be accomplished in the conscious mammal by the simultaneous intravenous injection of atropine and propranolol (17). The sinoatrial node subsequently assumes a rate which has been termed the intrinsic heart rate (IHR). Jose and Stitt (18) have suggested that the IHR is dependent upon the ionic balance at the myocardial membrane.

The intrinsic rates of atrial preparations from rats trained for 15 wk were significantly lower than those from sedentary animals. Sutton *et al.* (19) have found that in young men the greater the aerobic capacity the lower the IHR. Furthermore, they observed that regular exercise, resulting in an increase in aerobic capacity, could lower the IHR. Similar but less marked changes in the IHR were observed by Frick, Elovaino and Somer (20) in human subjects following training.

Chronic changes in myocardial electrolyte distribution may occur in training. Raab (21) considers that the cardio-protective value of exercise may be associated with an increase in myocardial potassium. Such chronic alterations could affect ionic mechanisms in the rate determining sinoatrial pacemaker cells. To date, however, there is no data on myocardial electrolyte distribution in the trained mammal.

Training brings about a considerable increase in the acetylcholine content of rat atria (4). Acetylcholine released from beating atrial preparations could have a slowing effect on the sinoatrial node. However, Grodner *et al.* (22) have found that the addition of atropine to the isolated rat atrial preparation has no effect on its intrinsic rate.

In the exercised rats there was a considerable subsensitivity to acetylcholine in comparison to the response of the sedentary group. Similar alterations in cholinergic sensitivity have been demonstrated in iris, another autonomically innervated structure (23). Bito, Dawson and Petrinovia (23) found that the cat iris developed super- or subsensitivity to acetylcholine if the animals were kept in either continuous darkness or bright light. They considered the target organ sensitivity to be a dynamic feature; any changes in tonic parasympathetic activity leading to an alteration in the cholinergic sensitivity of the iris.

A similar mechanism may be involved in the control of the cholinergic sensitivity of the atria. There are two ways in which the concentration of acetylcholine in the vicinity of the sinoatrial node may be chronically increased. A twofold elevation in the atrial acetylcholine content of trained rats has been observed by Herrlich, Raab and Gigee (4). However, as discussed above, addition of atropine to an isolated atrial preparation does not alter its rate. It seems probable that in the trained rat the sinoatrial node is not affected by the increased atrial stores of acetylcholine.

An increase in the concentration of acetylcholine at the sinoatrial node is more likely to be the result of an increase in the activity of the parasympathetic cardio-inhibitory fibers. That is, the sensitivity of the sinoatrial node to acetylcholine probably varies inversely with the tonic vagal cardio-inhibitory traffic.

From our results we suggest that the bradycardia of training is a result of both a decrease in the intrinsic sinoatrial rate, and an increase in tonic vagal cardio-inhibitory activity. While a decrease of the IHR in trained men has been observed previously, the current study demonstrates a lower intrinsic sinoatrial rate of the trained mammal in controlled conditions *in vitro*. An increase in tonic vagal activity has been proposed by several authors as the cause of training bradycardia. Our findings of a decrease in atrial cholinergic sensitivity in the trained rat are consistent with this hypothesis but reliable data concerning changes in cardiac parasympathetic activity and training are required to substantiate this concept.

Summary. The intrinsic rate of contraction of isolated atria from male rats, exercised for 15 wk, was considerably lower than that of sedentary controls. There was a marked subsensitivity to acetylcholine of atria from exercised rats. This subsensitivity may be related to an increase in the tonic release of acetylcholine by vagal cardio-inhibitory fibers in the exercised rat. It is concluded that the bradycardia of training probably involves at least two mechanisms; a decrease in the intrinsic rate of the sinoatrial node and an increase in cardiac parasympathetic activity.

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