Autonomic Neuroscience: Basic and Clinical 203 (2017) 33–40

Contents lists available at ScienceDirect

Autonomic Neuroscience: Basic and Clinical

journal homepage: www.elsevier.com/locate/autneu

Physical training associated with Enalapril but not to Losartan, results in better cardiovascular autonomic effects

Karina Delgado Maida, Ada Clarice Gastaldi, Tabata de Paula Facioli, João Eduardo de Araújo, Hugo Celso Dutra de Souza *

Exercise Physiology Laboratory, Ribeirão Preto Medical School, University of São Paulo, Ribeirão Preto, SP, Brazil

ARTICLE INFO

Article history:
Received 7 June 2016
Received in revised form 4 December 2016
Accepted 6 December 2016

Keywords:
Hypertension
Autonomic nervous system
Enalapril
Losartan
Exercise

ABSTRACT

Background: We investigated the cardiovascular autonomic effects of physical training associated with Enalapril or Losartan pharmacological treatments in spontaneously hypertensive rats (SHR).
Methods: SHRs, 18 weeks of age ($N = 48$) was assigned to either sedentary ($N = 24$) and trained ($N = 24$; aerobic training by swimming for 10 wk). Each group was subdivided in 3 subgroups ($N = 8$) vehicle (control); Enalapril (10 $mg \cdot kg^{-1} \cdot d^{-1}$); and Losartan (5 $mg \cdot kg^{-1} \cdot d^{-1}$). All animals received a 10-week treatment in drinking water. In the last week of the treatments, the animals had their femoral artery and vein cannulated for blood pressure recording and drug injection, respectively. The autonomic assessment was performed by means of different approaches: double cardiac autonomic block with atropine and propranolol, spectral analysis of heart rate variability (HRV) and systolic arterial pressure (SAPV) and assessment of baroreflex sensitivity (BRS).
Results: The groups treated with Enalapril, sedentary and trained, showed more significant decrease in blood pressure when compared to the other groups. Autonomic evaluation showed that the sedentary group treated with Enalapril or Losartan had similar results, characterized by decreased effect of sympathetic tone and/or increased effect of cardiac vagal tone associated with improved BRS. Isolated physical training attenuated only the effect of sympathetic tone. The association of physical training with Enalapril showed the best results, characterized by the predominance of vagal tone in cardiac autonomic balance, increased HRV, reduced SAPV and increased BRS.
Conclusions: Enalapril and Losartan promoted similar beneficial cardiovascular autonomic effects in sedentary animals, while only the association of physical training with Enalapril potentiated these effects.

1. Introduction

Angiotensin II is a polypeptide that plays an important role in regulating cardiovascular homeostasis through multiple actions, including those in the central nervous system (Stein et al., 1984; Reid, 1992; Fyhrquist et al., 1995; Wollert and Drexler, 1999; Ylitalo et al., 1999; Mehta and Griendling, 2007; Oparil and Schmieder, 2015; Leenen, 2014). Thus, angiotensin II plays a key role in the pathophysiology of cardiovascular disease as in hypertension, interfering with the cardiac autonomic balance (Stein et al., 1984; Reid, 1992; Fyhrquist et al., 1995; Wollert and Drexler, 1999; Ylitalo et al., 1999; Mehta and Griendling, 2007; Leenen, 2014; Oparil and Schmieder, 2015). This action is characterized by increased sympathetic influence and reduced cardiac vagal influence, resulting in important autonomic changes, such as reduction in baroreflex sensitivity (Reid, 1992; Wollert and Drexler, 1999; Ylitalo et al., 1999; Mehta and Griendling, 2007; Oparil and Schmieder, 2015).

In this context, clinical and experimental studies have shown that drugs that act by reducing the action of angiotensin II may lead to favorable adaptations in cardiovascular autonomic control when high blood pressure is present (Laflamme et al., 1997; Krum, 2001; Azevedo et al., 2003). Angiotensin converting enzyme inhibitors (ACEi) such as Enalapril and AT_1 receptor blockers such as Losartan, are widely used in the clinical practice due to these effects (Gottlieb et al., 1993; Dimitrova et al., 1998; Bonner et al., 2009, Inaba et al., 2011).

The ACEi act primarily by preventing the conversion of angiotensin I to angiotensin II (Oigman and Fritsch, 1998), while AT_1 receptor blockers prevent the action of angiotensin II, especially in blood vessels, contributing to the reduction of arterial pressure (AP) (Gottlieb et al., 1993; Dimitrova et al., 1998; Oigman and Fritsch, 1998; Bonner et al., 2009, Inaba et al., 2011). While, the effectiveness of these two drugs in reducing AP is well known (Rizzoni et al., 1998), their effects on the cardiovascular autonomic control have not been widely investigated (Yee

* Corresponding author at: Ribeirão Preto Medical School, University of São Paulo, Exercise Physiology Laboratory, Av. Bandeirantes, 3900, Vila Monte Alegre, Ribeirão Preto, SP 14049-900, Brazil.
E-mail address: hugocds@fmrp.usp.br (H.C.D. de Souza).

http://dx.doi.org/10.1016/j.autneu.2016.12.002
1566-0702/

and Struthers, 1998; Kaya et al., 2003). Some studies in the literature suggest that ACEi promotes broader cardiac autonomic benefits when compared to the AT_1 receptor blockers (Kaya et al., 2003).

Nonetheless, other therapeutic approaches are also used in order to reduce blood pressure, improve autonomic parameters and reduce the potential cardiovascular risk associated with hypertension. Aerobic exercise training is often used as treatment for hypertension, either prior to or during pharmacological treatments. Aside from the benefits already mentioned, studies in the literature show that when performed regularly, aerobic exercises can decrease the levels of angiotensin converting enzyme (ACE) and consequently, the concentrations of angiotensin II. According to these studies, this effect of physical exercise would result in autonomic and morphofunctional adaptations that contribute safely to the improvement of cardiovascular homeostasis (Gu et al., 2014; O'Donnell et al., 2014; Wu et al., 2014).

It has also been shown that physical training, regardless of drug treatment, improves cardiac autonomic modulation of hypertensive patients (Cozza et al., 2012). Moreover, the association of physical training with AT_1 receptor blockers such as Losartan, has also been studied. However, the studies show controversial results. Experimental studies have shown that the association of physical training with Losartan treatment reduces sympathetic autonomic modulation of spontaneously hypertensive rats (SHR) (Guo et al., 2008), while others have shown that the association does not promote any additional effects when compared to Losartan treatment alone (Azevedo et al., 2003).

Therefore, this study aimed to investigate whether or not treatment with Enalapril and Losartan would have different effects on the cardiovascular autonomic control in SHR. We hypothesized that the two drugs would lead to different autonomic adaptations, due to their different forms of action. In addition, we investigated whether the association of physical training with Enalapril or Losartan treatment would increase the cardiovascular autonomic effects seen with the isolated pharmacological treatments.

2. Methods

2.1. Subjects

Forty eight male SHR 18 weeks of age and weighing about 230 g, at the start of the study, were used. The rats were transferred from the Animal Facility at the Ribeirão Preto School of Medicine, University of São Paulo, to the housing colony where they were kept in a room with a strictly controlled temperature (21 ± 1 °C) and a 12:12 light:dark cycle. The rats had unrestricted access to tap water and standard rat chow (Nuvilab CR-1, Nuvital, Brazil).

All experimental protocols performed in the current study were approved by the Committee on Animal Research and Ethics of the School of Medicine of Ribeirão Preto, University of São Paulo (Protocol N.092/2012).

2.2. Experimental groups

The forty eight SHR were assigned to one of two groups: Sedentary group ($N = 24$) and Trained group ($N = 24$). The animals in the Trained group participated in daily swimming sessions for 10 weeks. Each group was subdivided into three smaller groups of 8 each; vehicle group (control); Enalapril group: animals treated for 10 weeks with Enalapril (10 $mg \cdot kg^{-1} \cdot d^{-1}$) (Enalapril) and Losartan group: animals treated for 10 weeks with Losartan (05 $mg \cdot kg^{-1} \cdot d^{-1}$) (Losartan).

2.3. Drug administration

All groups underwent a daily 10-week pharmacological treatment either with Enalapril or Losartan diluted in drinking water. The amount of drug given was calculated based on body weight and water consumption, which was recorded daily by graduated bottle. The control of water intake allowed for similar drug intake for all the animals, promoting similar reductions in AP during the pharmacological treatment. The dose of Enalapril was 10 $mg \cdot kg^{-1} \cdot d^{-1}$ and the dose of Losartan was 5 $mg \cdot kg^{-1} \cdot d^{-1}$ (Pahor et al., 1991).

2.4. Physical training

The rats in the trained group underwent a protocol of aerobic physical training that consisted of swimming sessions in a glass aquarium (100 cm long × 80 cm wide × 80 cm high), which allowed the simultaneous training of 6 animals. The tank was filled with 50 cm of warm water (30 ± 2 °C) and was changed after every group training session. The training program was conducted in 2 different stages over a total of 10 weeks: the first stage consisted of a 2-week adaptation period when session length was gradually increased from 5 to 45 min (incrementing 5 min per day), 5 times per week (Sant'Ana et al., 2011).

The second stage consisted of 8 weeks of 45-min physical training sessions, 5 times per week. To evaluate physical training intensity, blood was collected from the tail vein of the animals at the 3rd, 5th and 10th weeks immediately before and after 15 min of exercise, and the lactate concentration was measured (Accutrend® Plus, Roche Diagnostics, Mannheim, Germany). The expected lactate level was 5.5 to 6 mmol/L according to the procedure described by Gobatto et al. (2001). If animals did not reach the expected lactate concentration, we increased training exertion by fastening an impermeable, lead containing Velcro strap to their chest to increase body weight by 2 to 6% (Gobatto et al., 2001).

2.5. Tail plethysmography

All groups had the evolution of systolic (SAP), diastolic (DAP) and mean arterial pressure (MAP) recorded by tail plethysmography using CODA® system, Kent Scientific Corporation. This is an indirect AP recording system in which the rats are housed in a cylindrical acrylic tube, which are heated and vented properly for AP measurements. Thus, the rats had coupled in the proximal region of tail one cuff having the function inflate and deflate automatically at fixed intervals of approximately 15 s. Next cuff was attached to a pulse transducer (sensor) that captured the signals that were sent to and recorded in the computer system. All the animals underwent a period of adjustment to the experiment through three measures that were not used. When 12 new measurements were recorded, the AP was taken as the average of at least ten measurements.

These measurements were recorded at three different times: once prior to starting of the treatment (18 weeks of age), 5 weeks after starting of the treatment (23 weeks of age) and at the end of the 10-week-treatment (28 weeks of age). This procedure was performed in order to confirm the hypertension in all rats, and ensure that groups had similar mean AP before starting the protocols. The tail plethysmography was also used to monitor the development of hypertension in the vehicle group, and to monitor the development of the pharmacological and/or physical training treatments.

2.6. Experimental protocol

At the end of the experimental period, the rats were anesthetized with ketamine 80 mg/kg and xylazine 10 mg/kg, Polyethylene catheters made in our laboratory (PE-50 soldered to PE-10; Intramedic, Clay Adams, Parsippany, NJ, USA) were implanted into the left femoral artery and vein. Catheters were tunneled subcutaneously and exteriorized in the nape. To prevent blood from clotting, catheters were filled with heparinized saline solution (500 IU/mL). The rats were then allowed to recover for 24 h prior to the cardiac sympathovagal assessment protocol, which was carried out without anesthesia.