Canadian Science Publishing | Canadian Journal of Physiology and Pharmacology

Neuromuscular electrical stimulation but not photobiomodulation therapy improves cardiovascular parameters of rats with heart failure

Journal:	*Canadian Journal of Physiology and Pharmacology*
Manuscript ID	cjpp-2020-0339
Manuscript Type:	Article
Date Submitted by the Author:	22-Jun-2020
Complete List of Authors:	Capalonga, Lucas; Universidade Federal de Ciências da Saúde de Porto Alegre, Programa da Pós-Graduação em Ciências da Saúde Araújo, Cintia; UFCSPA Hentschke, Vítor; UFCSPA, Rossato, Douglas; Centro Universitario Franciscano, Physiotherapy Quagliotto, Edson; Federal University of Rio Grande do Sul, Graduate Course in Neurosciences; Becker, Tiago; Universidade Federal do Rio Grande do Sul, Engenharia Rigatto, Katya; UFCSPA, Ferraresi, Cleber; Universidade de São Paulo, Engenharia biomédica Parizotto, Nivaldo; Universidade Federal de São Carlos, Fisioterapia Dal Lago, Pedro; Federal University of Health Sciences of Porto Alegre, Department of Physical Therapy
Is the invited manuscript for consideration in a Special Issue:	Not applicable (regular submission)
Keyword:	Heart failure, Electric stimulation, Photobiomodulation, Myocardial infarction, Autonomic nervous system

Neuromuscular electrical stimulation but not photobiomodulation therapy improves cardiovascular parameters of rats with heart failure

CAPALONGA, LUCAS[1,2]; DE ARAUJO, CINTIA LAURA PEREIRA[1,3]; HENTSCHKE, VÍTOR SCOTTA[4]; ROSSATO, DOUGLAS DALCIN[5]; EDSON QUAGLIOTTO[6]; BECKER, TIAGO[7]; RIGATTO, KATYA[8]; CLEBER FERRARESI[9]; NIVALDO ANTONIO PARIZOTTO[9,10,11]; DAL LAGO, PEDRO[1,2,3]

[1] Laboratory of Experimental Physiology, Universidade Federal de Ciências da Saúde de Porto Alegre (UFCSPA), Porto Alegre, RS, Brazil

[2] Programa de Pós-Graduação em Ciências da Saúde, Universidade Federal de Ciências da Saúde de Porto Alegre (UFCSPA), Porto Alegre, RS, Brazil

[3] Programa de Pós-Graduação em Ciências da Reabilitação, Universidade Federal de Ciências da Saúde de Porto Alegre (UFCSPA), Porto Alegre, RS, Brazil

[4] Universidade Luterana do Brasil (ULBRA), Cachoeira do Sul, RS, Brazil.

[5] Centro Universitário Franciscano, Santa Maria, RS, Brazil.

[6] Universidade de Santa Cruz do Sul (UNISC), Santa Cruz do Sul, RS, Brazil.

[7] Departamento de Engenharia Mecânica, Universidade Federal do Rio Grande do Sul (UFRGS), Porto Alegre, RS, Brazil.

[8] Laboratório de Fisiologia Translacional, Universidade Federal de Ciências da Saúde de Porto Alegre (UFCSPA), Porto Alegre, RS, Brazil.

[9] Biomedical Engineering, Universidade do Brasil, São Paulo, Brazil.

[10] Biotechnology in Regenerative Medicine and Medical Chemistry, Universidade de Araraquara, Araraquara, Brazil.

[11] Physical Therapy Department, Universidade Federal de São Carlos, São Carlos, Brazil.

Corresponding author: Prof. Pedro Dal Lago

Programa de Pós-Graduação em Ciências da Saúde (PPG-CS), Universidade Federal de Ciências da Saúde de Porto Alegre (UFCSPA), Rua Sarmento Leite, 245, 90050-170, Porto Alegre, RS, Brazil, e-mail: pdallago@ufcspa.edu.br

Abstract:

The aim was to analyze the effect of neuromuscular electrical stimulation (NMES) and photobiomodulation (PBMT) on cardiovascular parameters, hemodynamic function, arterial baroreflex sensitivity (BRS), and autonomic balance (ANS) of rats with heart failure (HF). Male Wistar rats (220-290g) were organized into five groups: Sham (n=6), Control-HF (n=5), NMES-HF (n=6), PBMT-HF (n=6), and NMES+PBMT-HF (n=6). Myocardial infarction (MI) was induced by left coronary artery ligation. Animals were subjected to an eight-week NMES and PBMT protocol. Statistical analysis with the General Linear Model (GLM) followed by the Bonferroni post hoc test. Rats of NMES-HF group showed a higher MI area than Control-HF group (P=0.003), PBMT-HF (P=0.002), and NMES+PBMT-HF (P=0.012). NMES-HF and NMES+PBMT-HF showed higher pulmonary congestion (P=0.004 and P=0.02), and lower systolic pressure (P=0.019 and P=0.002) than Sham group. NMES+PBMT-HF showed lower mean arterial pressure (P=0.02) than Sham group. Control-HF showed a higher heart rate than NMES-HF and NMES+PBMT-HF (P=0.017 and P=0.013). There was no difference in the BRS and ANS variables between groups. In conclusion, eight-week NMES isolated or associated with PBMT protocol reduced basal heart rate, systolic and mean arterial pressure, without influence on baroreflex sensibility and autonomic control, and no effect of PBMT was seen in rats with HF.

Keywords: Heart failure, Electric Stimulation, Photobiomodulation, Myocardial infarction, Autonomic nervous system.

1. Introduction

Heart failure (HF) is a clinical syndrome characterized by symptoms such as dyspnea and fatigue, resulting from a structural or functional cardiac disorder that impairs the hemodynamic function of the heart (Ponikowski et al., 2017; Poole, Hirai, Copp, & Musch, 2012). During the development of HF, some changes arise early, even before the symptoms appear. The main one is the sympathetic nervous system hyperactivity associated with the reduction of parasympathetic activity, which characterizes the state of neurohumoral excitation, directly related to the baroreceptors dysfunction and responsible for modulating heart rate and arterial vasomotor tone (Besnier et al., 2017; Lazzarotto Rucatti et al., 2015). The arterial baroreflex system is one of the most important mechanisms that act on cardiovascular homeostasis, can regulate heart rate (HR) and blood pressure (BP), and therefore, directly influence the autonomic nervous system (Bristow, Honour, Pickering, Sleight, & Smyth, 1969; Eckberg, Drabinsky, & Braunwald, 1971; Quagliotto et al., 2008).

In the early stages of the disease, the sympathetic excitation plays a compensatory role to keep hemodynamic parameters within the normal range. However, it becomes detrimental as the disease progresses, contributing to the aggravation of cardiovascular function and the consequent death of the patient with HF (Gronda et al., 2014).

Currently, is known that NMES, at short-term, has been reducing pulmonary congestion, preventing muscular atrophy, increasing muscle blood vessel density, recovering GLUT-4 protein levels (de Leon et al., 2011), reducing sympathetic-vagal ratio over heart rate, activating arterial baroreceptors, and providing better cardiovascular autonomic control in rats with HF (Lazzarotto Rucatti et al., 2015).

Moreover, PBMT was tested a few years ago by our laboratory group in an innovative way, to elucidate its effects on rats with HF. So far, it has been possible to notice that the short-term application of PBMT can improve inflammatory profile at both levels systemic and skeletal muscle (Vítor S. Hentschke et al., 2013); reduce oxidative stress and DNA damage when applied at low doses (3J/cm^2) but may cause DNA damage at higher doses (21J/cm^2) (Biasibetti et al., 2014). However, the long-term effects are related to the improvement of functional capacity, by increasing the distance, time and speed in the maximal exercise test (Capalonga et al., 2016), as well as by the increase in maximal oxygen uptake ($\dot{V}O_2$max), oxygen uptake of the reserve ($\dot{V}O_2$reserve) and in basal oxygen uptake ($\dot{V}O_2$basal), when associated to resistance training (Vítor Scotta Hentschke, Capalonga, Rossato, Perini, Alves, Stefani, et al., 2017). In healthy rats, it was possible to notice that, when applied in higher doses (61.2J/cm^2), but not in lower does (8.7J/cm^2), PBMT promoted improvement in ($\dot{V}O_2$basal), ($\dot{V}O_2$max), maximum carbon dioxide output ($\dot{V}CO_2$max) and distance covered in a dose-dependent pattern (Perini, Scotta Hentschke, Sonza, & Dal Lago, 2016).

However, there is a gap in the literature that describes the effects of these therapies on autonomic balance, baroreflex activity, and heart rate variability in the long-term. Therefore, this study aimed to test the hypothesis that an eight-week program of NMES and PBMT could improve the cardiovascular parameters, hemodynamic function, baroreflex sensitivity, heart rate variability, and autonomic balance of rats with HF.

2. Material and methods

2.1 Animals

This experimental controlled study was performed on 43 male Wistar rats (220 to 290 g), from the Animal Breeding Unit of the Universidade Federal de Ciências da Saúde de Porto Alegre (UFCSPA). The rats were housed three per cage and received food and water ad libitum in an animal room under a 12:12-h light-dark cycle, at 22 ºC. The study's protocol followed the ethical rules established by the Guide for Care and Use of Experimental Animals published by the National Institute of Health (NIH publication 85-23, revised in 1996). The UFCSPA's Ethics Committee on Animal Use approved all procedures (protocol 189/16). For the sample size calculation, we used as the main outcome the maximum gain variable. To detect a minimum difference of 66% (Lazzarotto Rucatti et al., 2015), with an alpha error of 0.05, an effect size of 0.80, and a power of analysis of 95%, we estimated a total of 35 animals. The software used for the calculation was GPower version 3.1.9.2 for Windows (University of Düsseldorf, Düsseldorf, Germany).

2.2 Surgery to induce myocardial infarction (MI)

The rats were weighed and then anesthetized with inhaled isoflurane 2% (Isoforine, 100 ml - Cristália, Brasil) in a bell glass. The surgical procedure started with the trichotomy of the left hemithorax. A small incision was made in the skin at the fourth and fifth ribs, and the pectoral muscles were removed. Briefly, the heart was exposed through a left thoracotomy. A mono-nylon suture 6-0 was threaded around the main left descending coronary artery between 1 and 2 mm distal to the edge of the left atrium, and the left coronary was ligated. Sham-operated animals underwent the same procedure, without tying the suture, and served as controls. The chest wall was closed; the skin was sutured with mono-nylon suture 3-0, and the pneumothorax was drained using continuous aspiration. After the surgery, saline was applied, 1 ml intraperitoneally at room temperature for fluid replacement, as well as a single dose of gentamicin, 0.1 ml intramuscularly for prevention of infection. For the management of post-operative pain, Turbogesic® analgesic (butorphanol) was used, 0.5 mg/kg intraperitoneally 12/12h during the first 24 hours. For recovery, the rats were placed in a heated environment (Capalonga et al., 2016).

2.3 Experimental design

To compare the effects of the interventions, of the 43 animals initially started, only 35 survived the MI. After the eight-week recovery period, they were divided into five experimental groups: Sham control group (Sham, n = 6), HF control group (Control-HF, n = 6), neuromuscular electrical stimulation group (NMES-HF, n = 8), photobiomodulation therapy (PBMT-HF, n = 8) and neuromuscular electrical stimulation plus photobiomodulation therapy (NMES + PBMT-HF, n = 7). After this period, the animals underwent eight weeks of NMES, PBMT, or both protocols. At the end of the protocols, a hemodynamic analysis was performed, followed by euthanasia, in which six rats showed no signs of left ventricular dysfunction, characterized by Left Ventricular End Diastolic Pressure (LVEDP) less than 15mmHg or myocardial infarction area less than 20%, totaling 32.5% loss. Thus, 29 rats were selected for study: Sham control group (Sham, n = 6), HF control group (Control-HF, n = 5), neuromuscular electrical stimulation group (NMES-HF, n = 6), photobiomodulation therapy (PBMT-HF, n = 6), and neuromuscular electrical stimulation plus photobiomodulation therapy (NMES + PBMT-HF, n = 6), as shown in Figure 1.

2.4 Neuromuscular Electrical Stimulation (NMES) protocol

Electrically stimulated groups were subjected to a protocol of neuromuscular electrical stimulation (FES, VIF 995, Quark, Piracicaba, Brazil) for eight weeks, five days per week for 30 minutes per day. The electrical stimulation was applied to the gastrocnemius muscle of the right leg through the surface electrode fixed by a system with a velcro fastener. Animals were placed in a rigid corset, along with the trunk, allowing the movement of fore and hind paws to prevent the removal of the electrodes. This followed previous studies and was considered appropriate after simulation of therapeutic intervention (Durigan et al., 2014). The dose used was enough to generate a visible muscle contraction, starting from four to six mA, ten mA until reaching the end of the protocol, respecting parameters described in Table 1 (Lazzarotto Rucatti et al., 2015). Training progression was ensured by increasing the dose during each session as well as during the entire period of the protocol. This procedure was applied immediately after the PBMT protocol for the group NMES + LEDT-HF.

2.5 Photobiomodulation therapy (PBMT) protocol

The rats were placed on a surgical table and immobilized manually by the researcher. The LEDT (equipment developed at the Institute of Physics at São Carlos of the Universidade de São Paulo in cooperation with researchers from the Laboratory of Electrophysical Agents of the Universidade Federal de São Carlos, SP, Brazil) was applied directly on the right gastrocnemius muscle, with a shaved paw as described above, for eight weeks, five days per week (Capalonga et al., 2016), according to the parameters of Table 1. This procedure was applied immediately before the NMES protocol for the group NMES + LEDT-HF.

2.6 Awake measurements of cardiovascular parameters

One week after the protocols, all rats were anesthetized with xylazine (12 mg/kg, i.p.) and ketamine (90 mg/kg, i.p.) for the femoral catheterization. Two catheters filled with saline (0.06 ml) and heparin (0.01 ml) were implanted into the abdominal aorta and inferior vena cava, which were used to measure mean arterial pressure (MAP) and drugs administration, respectively. Conscious rats were studied on a subsequent day after the catheter placement. The arterial catheter was connected to a 40 cm tube attached to a strain-gauge pressure transducer (Miniature Pulse Transducer RP-155, Houston, USA), coupled to a pressure amplifier (General Purpose Amplifier 4 - model 2, Stemtech Inc., Houston, USA), and blood-pressure signals were recorded over a 15 minutes at 1 kHz sampling frequency (Windaq - AT/CODAS, Dataq Instruments Inc., Akron, USA). On a beat-to-beat basis, the measured data were analyzed to quantify the variables of interest (Lazzarotto Rucatti et al., 2015; Quagliotto et al., 2008).

2.7 Baroreflex sensitivity (BRS) and autonomic modulation

On a subsequent day following the catheter placement, MAP and HR were registered for 15 minutes as baseline control. HR changes to test BRS were recorded during peak changes (augmentation or reduction) in MAP due to a single dose of venous injection of phenylephrine (8 µg/ml; Sigma Chemical, St. Louis, USA) or sodium nitroprusside (100 µg/ml; Sigma Chemical), respectively (Lazzarotto Rucatti et al., 2015; Quagliotto et al., 2008). The alterations in MAP were within the 10 to 30 mm Hg range, and the BRS determination was made by fitting the MAP and HR alterations to a sigmoidal logistic equation (Head & McCarty, 1987).

The autonomic modulation of HR was analyzed by spectral analysis. The signals were divided into windows of 300 beats for spectral analysis, with a 50% overlap with the next window. The signal spectral densities were estimated using an autoregressive algorithm by the Yule-Walker method. For the autoregressive algorithm, the filter order was determined for each sample based on the methodology shown in “Heart rate variability signal processing - a quantitative approach as an aid to diagnosis in cardiovascular pathologies.” Windows containing irregular beats were rejected in the analysis. The cut-off frequencies of LF= 0.25-0.75 Hz and HF=0.75-3.0 Hz were chosen according to (Dabiré, Mestivier, Jarnet, Safar, & Chau, 1998). When they presented non-stationary episodes, they were discarded, and a new random selection was performed (Quagliotto, Casali, Dal Lago, & Rasia-Filho, 2015; Rigatto, Casali, Shenoy, Katovich, & Raizada, 2013). The low-frequency spectral components (LF, 0.25 to 0.75 Hz) represents predominantly sympathetic modulation, and the high frequency (HF, 0.75 to 3.00 Hz) represents vagal modulation. The values were expressed in absolute (LFa and HFa; ms2) and normalized (LFnu and HFnu; %) units. This method estimates the frequency and power of each component and indicates the involvement of the heart modulation of sympathetic and parasympathetic systems. The ratio between the LF and HF (LF/HF) components was considered a synthesis of sympathovagal balance.

2.8 Hemodynamic evaluation

After the baroreflex evaluation, animals were anesthetized with xylazine (12 mg/kg, i.p.) and ketamine (90 mg/kg, i.p.) and a small incision was made in the anterior cervical region to access the right carotid artery and introduce a polyethylene catheter (PE-50) for cannulation of the left ventricle (LV) (Jaenisch et al., 2011). The AP was recorded first during for five minutes. Then, the catheter was positioned inside the LV, and the pulse wave was monitored using the typical graphic registration of ventricular pressure and recorded for 5 min. These data were used to determine LV systolic pressure (LVSP), LV maximum change in pressure over time (+dP/dtmax), LV minimum change in pressure over time (-dP/dtmax), and LV end-diastolic pressure (LVEDP).

2.9 Heart hypertrophy, lung, and hepatic congestion

The heart, lungs, and liver were removed and weighed. The right ventricle (RV) and left ventricle (LV) were dissected, separated, and weighed. The heart-to-body mass (H/BM), LV-to-body mass (LV/BM), and RV-to-body mass (RV/BM) were determined and used as an indication of heart hypertrophy. The lungs and liver of each animal were dehydrated (80°C) for 48h and then reweighed to determine the water content. The lung and liver wet-to-dry weight ratios were used to determine the percentage of water in those tissues, as an indication of congestion (Vítor Scotta Hentschke, Capalonga, Rossato, Perini, Alves, Quagliotto, et al., 2017).

2.10 Infarct size

The myocardial infarct scar and the total area of the left ventricle were traced manually on the scanned images and measured automatically by computer (Image Pro-Plus 6.1, Media Cybernetics, Silver Spring, USA) program. The infarct size, expressed as a percentage, was calculated by dividing the sum of infarct areas of all sections by the amount of the areas of the left ventricle (including those without infarct) and multiplied by 100. The analysis was performed in triplicate by three blinded observers (Alves, Nunes, Stefani, & Dal Lago, 2014). All data collection procedures are summarized in Figure 2.

2.11 Statistical analysis

The Shapiro-Wilk test was conducted to evaluate normality for all variables. Statistical differences among groups were identified using the General Linear Model (GLM), followed by the Bonferroni post hoc test. The infarct size was considered a covariate for the analysis of BRS, autonomic modulation and hemodynamic variables, and the infarct size of the Sham group was considered zero. A P value <0.05 was considered statistically significant. To further examine the statistical difference obtained, we performed Cohen's d test to measure the effect size test, followed by a power analysis of the comparison. We considered a reliable power analysis with at least 80.0%. All statistical analyses were performed with IBM SPSS Statistic Data Editor version 22. The images and graphs were created with GraphPad Prism version 7.0

for Windows (San Diego, California, USA). Data are presented as mean and standard deviation (SD).

3. Results

3.1 Mortality, morphological characteristics, pulmonary and hepatic congestion

The rats with HF showed a mortality level of ~20% during and after MI-induced surgery. Table 2 summarizes the data on body mass, heart hypertrophy, infarct size, and pulmonary and hepatic congestion. Sham group animals showed lower wet to dry lung ratio compared with NMES-HF ($P = 0.004$; effect size = 2.60; power analysis = 99%), and NMES + PBMT-HF groups ($P = 0.02$; effect size = 2.36; power analysis = 98%). Importantly, the animals in the NMES-HF group developed larger infarct areas than the Control-HF animals ($P = 0.003$, effect size = 1.92, power analysis= 89%), the PBMT-HF animals ($P = 0.002$; effect size = 2.21; power analysis = 97%), and the NMES + PBMT-HF animals ($P = 0.012$; effect size = 1.81; power analysis = 89%). There was no difference in other morphological variables and hepatic congestion.

3.2 NMES and PBMT did not influence autonomic control

Although the animals in the NMES-HF and NMES + PBMT-HF groups had higher participation of the high-frequency component HF and lower LF/HF ratio values when compared to the Control-HF group, this difference was not significant. The infarct size did not influence ($p > 0.05$) any of the outcomes. Data is presented in Table 3.

3.3 NMES and PBMT on cardiovascular parameters and hemodynamic function

The values of hemodynamic parameters are presented in Table 4. The animals of the NMES-HF and NMES + PBMT-HF groups showed lower systolic arterial pressure (SAP) than Sham group ($P = 0.019$, effect size = 2.21, power analysis = 97%; and $P = 0.002$, effect size = 2.79, power analysis = 99%) respectively. The NMES + PBMT-HF group showed lower mean arterial pressure (MAP) than Sham group ($P = 0.02$, effect size = 2.22, power analysis = 97%). The animals of the Control-HF group demonstrated higher heart rate (HR) than NMES-HF group ($P = 0.017$, effect size = 1.75, power analysis = 84%), NMES + PBMT-HF ($P = 0.013$, effect size = 1.86, power analysis = 88%), and Sham ($P = 0.058$, effect size = 1.73, power analysis = 83%) respectively.

Heart failure groups showed higher LVEDP than Sham group, Control-HF ($P = 0.002$, effect size = 3.65, power analysis = 99%), NMES-HF ($P < 0.001$, effect size = 7.89, power analysis = 100%), PBMT-HF ($P < 0.001$, effect size = 4.10, power analysis = 99%), and NMES + PBMT-HF ($P < 0.001$, effect size = 3.07, power analysis = 99%) respectively. The infarct size did not influence ($p > 0.05$) any of the outcomes.

3.4 NMES and heart rate response

In the evaluation of the baroreflex by the logistic analysis of the sigmoidal baroreceptor curve, we verified that the animals of the NMES-HF group showed lower MAP_{50} than PBMT-HF group (90.8 ± 12.5 vs 104 ± 5.1, P = 0.075; effect size = 1.38; power analysis = 72%). Furthermore, the NMES-HF group showed lower HR range, and higher Maximum gain than other groups, but not significantly. The infarct size did not influence ($p > 0.05$) any of the outcomes. Data are shown in Table 5 and summarized in Figure 3. Blood pressure variability and heart rate during baroreflex sensitivity analysis are presented in Figure 4.

4. Discussion

The main novelties of this investigation are associated with the long-term changes in hemodynamic function that induces an autonomic imbalance and BRS adaptation to a new set point due to heart failure. Through the application of the different interventions in the peripheral muscles of rats with HF, we showed that NMES and PBMT induce: 1) a decrease in systolic blood pressure; 2) a reduction in mean arterial pressure; 3) and a reduction of the heart rate. Interestingly, although the animals submitted to the therapies have shown improvement in the parameters of autonomic control, by increasing the HF component and reducing the LF/HF ratio, suggesting greater participation of the parasympathetic nervous system, as well as improving baroreflex sensitivity, by the reduction of the HR range and MAP_{50}, and increased gain, these results were not significant. We believe in the hypothesis that, as it is a long-term protocol, there was an adaptation of peripheral receptors.

The model of rats with HF, induced by left descending coronary artery ligation surgery, presents as central characteristic hemodynamic dysfunction, which is directly related to the area of MI, especially when it is higher than 30% (Vítor Scotta Hentschke, Capalonga, Rossato, Perini, Alves, Quagliotto, et al., 2017; Jaenisch et al., 2011; Pfeffer et al., 1979). Previous studies have shown that rats submitted to MI induction surgery showed more significant impairment of cardiac function (lower ejection fraction and shortening fraction), increased chemoreflex activity, greater sympathetic activation, and reduced parasympathetic activity with a consequent reduction in HRV and increased LF/HF ratio, besides more significant impairment of baroreflex sensitivity, with areas of infarction of 38% of the LV (Del Rio, Marcus, & Schultz, 2013). In the present study, the NMES group had a larger MI area (36.2%) than the other experimental groups, associated with hemodynamic dysfunction. These changes are directly related to the surgical procedure itself, as they were all performed by the same researcher.

Muscle contraction generated by NMES improves the morphological and metabolic characteristics of the skeletal muscle, providing an increase in blood vessel density, GLUT-4 protein content, and cross-sectional muscle area of HF rats (de Leon et al., 2011).

Sympathetic nervous system hyperactivation is a strong predictor of morbidity and mortality in patients with HF. Changes in autonomic control are directly related to several cardiovascular reflexes and central adjustments that contribute to sympathetic hyperactivity in HF (Triposkiadis

et al., 2009). The attenuation of the baroreflex system is a determinant factor in the neural control of the cardiovascular system. It is used as a significant prognostic value and a predictor of sudden cardiac death in patients with HF (La Rovere, Bigger, Marcus, Mortara, & Schwartz, 1998; La Rovere, Pinna, & Raczak, 2008; Zhang & Anderson, 2014). As seen, rats of the NMES-HF and NMES + PBMT-HF groups increased the gain and reduced the HR range and MAP_{50} when compared to the other groups, but not significantly. These can be represented by a more significant variation of the heart rate to maintain blood pressure within normal parameters. A study from our laboratory has also found similar results; animals with HF undergoing the four-week NMES protocol had increased sensitivity of arterial baroreceptors, reduction in sympathetic modulation, increase in parasympathetic modulation, and a decrease in sympathovagal balance (Lazzarotto Rucatti et al., 2015). Another interesting study from our laboratory showed that an eight-week protocol of electroacupuncture (EA) was able to improve the baroreflex sensitivity of rats with HF; animals of the EA-HF group showed higher gain than the Sham group animals. However, it did not evaluate the sympathovagal balance, which could help to explain this result (Lima et al., 2015).

Importantly, the animals of the Control-HF group showed smaller MI areas than the NMES-HF group, which may influence the results. Smaller MI areas are related to lower structural cardiac impairment. Besides, BRS may lose function over time, being replaced by long-term changes that are triggered by other neuropeptides such as angiotensin II, vasopressin, and aldosterone (Gallinat, Busche, Raizada, & Sumners, 2000; Macova, Pavel, & Saavedra, 2009). It is well documented in the literature that the central overactivation of angiotensin AT1 receptors are responsible for baroreceptor desensitization, sympathetic hyperactivity, and decreased sympathetic inhibition after myocardial infarction in rats and rabbits (Szczepanska-Sadowska, Czarzasta, & Cudnoch-Jedrzejewska, 2018). Nevertheless, aldosterone has pro-fibrotic effects that are capable of reducing arterial compliance and compromising baroreflex activity, which is directly related to impaired autonomic control of cardiac and vascular functions that are generated by hypertension and heart failure (Struthers & MacDonald, 2004).

We are pioneers in the use of PBMT in rats with HF. To date, it has been possible to observe that PBMT improves the systemic and muscular inflammatory profile, increasing the levels of anti-inflammatory cytokines (Vítor S. Hentschke et al., 2013) and reducing oxidative stress when applied in low doses ($3J/cm^2$). Still, it can cause DNA damage in higher doses ($21J/cm^2$) (Biasibetti et al., 2014). Besides, PBMT improves functional capacity by increasing $\dot{V}O_2$basal, $\dot{V}O_2$max, $\dot{V}O_2$reserve, run distance, time to exhaustion, and maximum velocity reached in the cardiopulmonary exercise test (Capalonga et al., 2016; Vítor Scotta Hentschke, Capalonga, Rossato, Perini, Alves, Stefani, et al., 2017). It is also important to highlight that, so far, the main effects of PBMT (LEDT or LLLT) are related to the peripheral muscle tissue without significant influences at the central hemodynamic level, which was also confirmed in the present study.

Our findings confirm the effects of NMES, but it has not been possible to establish the impact of photobiomodulation therapy on the hemodynamic function of HF rats. However, it should be noted that NMES alone or combined with PBMT was not able to maintain its effects on baroreflex sensitivity and autonomic control for a longer-term. Thus, reinforcing information that these therapies, alone, do not provide the necessary benefits to improve hemodynamic function and autonomic control. At the same time, it has not yet been possible to describe the main mechanisms responsible for the improvement in hemodynamic function and arterial pressure, reinforcing the need for further studies to investigate these results.

CONCLUSIONS

In conclusion, the main finding of the present study was to show the effects of long-term NMES isolated and associated with PBMT in the rat model of heart failure. Here we showed that, despite the greater area of infarction, the animals of the NMES-HF group had improved their hemodynamic function and arterial pressure control, without influence on baroreflex sensibility and autonomic control. Besides that, the association with the PBMT did not bring additional effects on these variables.

ACKNOWLEDGMENTS

We are thankful to Giuseppe Potrick Stefani for their assistance during the development of this study.

GRANTS

This research was supported by the Fundação de Amparo à Pesquisa do Rio Grande do Sul (FAPERGS, Programa Pesquisador Gaúcho - PqG, 2017) and the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES).

DISCLOSURES

The authors Cleber Ferraresi and Nivaldo Parizotto declare that they applied for a patent for the LEDT equipment used in the present study. There are no conflicts of interest to be declared by the other authors.

Reference

Alves, J. P., Nunes, R. B., Stefani, G. P., & Dal Lago, P. (2014). Resistance training improves hemodynamic function, collagen deposition and inflammatory profiles: Experimental model of heart failure. *PLoS ONE*. https://doi.org/10.1371/journal.pone.0110317

Besnier, F., Labrunée, M., Pathak, A., Pavy-Le Traon, A., Galès, C., Sénard, J.-M., & Guiraud, T. (2017). Exercise training-induced modification in autonomic nervous system: An update for cardiac patients. *Annals of Physical and Rehabilitation Medicine*, *60*(1), 27–35. https://doi.org/10.1016/j.rehab.2016.07.002

Biasibetti, M., Rojas, D. B., Hentschke, V. S., Moura, D. J., Karsten, M., Wannmacher, C. M. D., … Dal Lago, P. (2014). The influence of low-level laser therapy on parameters of oxidative stress and DNA damage on muscle and plasma in rats with heart failure. *Lasers in Medical Science*, *29*(6), 1895–1906. https://doi.org/10.1007/s10103-014-1597-1

Bristow, J. D., Honour, A. J., Pickering, G. W., Sleight, P., & Smyth, H. S. (1969). Diminished baroreflex sensitivity in high blood pressure. *Circulation*. https://doi.org/10.1161/01.CIR.39.1.48

Capalonga, L., Karsten, M., Hentschke, V. S., Rossato, D. D., Dornelles, M. P., Sonza, A., … Dal Lago, P. (2016). Light-emitting diode therapy (LEDT) improves functional capacity in rats with heart failure. *Lasers in Medical Science*, *31*(5). https://doi.org/10.1007/s10103-016-1922-y

Dabiré, H., Mestivier, D., Jarnet, J., Safar, M. E., & Chau, N. P. (1998). Quantification of sympathetic and parasympathetic tones by nonlinear indexes in normotensive rats. *American Journal of Physiology - Heart and Circulatory Physiology*.

de Leon, E. B., Bortoluzzi, A., Rucatti, A., Nunes, R. B., Saur, L., Rodrigues, M., … Dall'Ago, P. (2011). Neuromuscular electrical stimulation improves GLUT-4 and morphological characteristics of skeletal muscle in rats with heart failure. *Acta Physiologica*, *201*(2), 265–273. https://doi.org/10.1111/j.1748-1716.2010.02176.x

Del Rio, R., Marcus, N. J., & Schultz, H. D. (2013). Carotid chemoreceptor ablation improves survival in heart failure: Rescuing autonomic control of cardiorespiratory function. *Journal of the American College of Cardiology*. https://doi.org/10.1016/j.jacc.2013.07.079

Durigan, J. L. Q., Delfino, G. B., Peviani, S. M., Russo, T. L., Ramírez, C., Da Silva Gomes, A. D. B., & Salvini, T. F. (2014). Neuromuscular electrical stimulation alters gene expression and delays quadriceps muscle atrophy of rats after anterior cruciate ligament transection. *Muscle & Nerve*, *49*(1), 120–128. https://doi.org/10.1002/mus.23883

Eckberg, D. L., Drabinsky, M., & Braunwald, E. (1971). Defective Cardiac Parasympathetic Control in Patients with Heart Disease. *New England Journal of Medicine*. https://doi.org/10.1056/NEJM197110142851602

Gallinat, S., Busche, S., Raizada, M. K., & Sumners, C. (2000). The angiotensin II type 2 receptor: An enigma with multiple variations. *American Journal of Physiology-Endocrinology and Metabolism*.

Gronda, E., Seravalle, G., Brambilla, G., Costantino, G., Casini, A., Alsheraei, A., … Grassi, G. (2014). Chronic baroreflex activation effects on sympathetic nerve traffic, baroreflex function, and cardiac haemodynamics in heart failure: A proof-of-concept study. *European Journal of Heart Failure*. https://doi.org/10.1002/ejhf.138

Head, G. A., & McCarty, R. (1987). Vagal and sympathetic components of the heart rate range and gain of the baroreceptor-heart rate reflex in conscious rats. *Journal of the Autonomic Nervous System*. https://doi.org/10.1016/0165-1838(87)90023-3

Hentschke, Vítor S., Jaenisch, R. B., Schmeing, L. A., Cavinato, P. R., Xavier, L. L., & Dal Lago, P. (2013). Low-level laser therapy improves the inflammatory profile of rats with heart failure. *Lasers in Medical Science*, *28*(3), 1007–1016. https://doi.org/10.1007/s10103-012-1190-4

Hentschke, Vítor Scotta, Capalonga, L., Rossato, D. D., Perini, J. L., Alves, J. P., Quagliotto, E., … Dal Lago, P. (2017). Functional capacity in a rat model of heart failure: impact of myocardial infarct size. *Experimental Physiology*. https://doi.org/10.1113/EP086076

Hentschke, Vítor Scotta, Capalonga, L., Rossato, D. D., Perini, J. L., Alves, J. P., Stefani, G. P., … Lago, P. D. (2017). Maximal oxygen uptake and exercise tolerance are improved in rats with heart failure subjected to low-level laser therapy associated with resistance training. *Lasers in Medical Science*, *32*(1), 73–85. https://doi.org/10.1007/s10103-016-2088-3

Jaenisch, R. B., Hentschke, V. S., Quagliotto, E., Cavinato, P. R., Schmeing, L. A., Xavier, L. L., & Dal Lago, P. (2011). Respiratory muscle training improves hemodynamics, autonomic

function, baroreceptor sensitivity, and respiratory mechanics in rats with heart failure. *Journal of Applied Physiology*, *111*(6), 1664–1670. https://doi.org/10.1152/japplphysiol.01245.2010

Karavidas, A., Driva, M., Parissis, J. T., Farmakis, D., Mantzaraki, V., Varounis, C., … Filippatos, G. (2013). Functional electrical stimulation of peripheral muscles improves endothelial function and clinical and emotional status in heart failure patients with preserved left ventricular ejection fraction. *American Heart Journal*, *166*(4), 760–767. https://doi.org/10.1016/j.ahj.2013.06.021

La Rovere, M. T., Bigger, J. T., Marcus, F. I., Mortara, A., & Schwartz, P. J. (1998). Baroreflex sensitivity and heart-rate variability in prediction of total cardiac mortality after myocardial infarction. *Lancet*. https://doi.org/10.1016/S0140-6736(97)11144-8

La Rovere, M. T., Pinna, G. D., & Raczak, G. (2008). Baroreflex sensitivity: Measurement and clinical implications. *Annals of Noninvasive Electrocardiology*. https://doi.org/10.1111/j.1542-474X.2008.00219.x

Lazzarotto Rucatti, A., Boemo Jaenisch, R., Dalcin Rossato, D., Poletto Bonetto, J. H., Ferreira, J., Xavier, L. L., … Lago, P. D. (2015). Skeletal muscle electrical stimulation improves baroreflex sensitivity and heart rate variability in heart failure rats. *Autonomic Neuroscience*, *193*, 92–96. https://doi.org/10.1016/j.autneu.2015.08.008

Lima, J. W., Hentschke, V. S., Rossato, D. D., Quagliotto, E., Pinheiro, L., Almeida, E., … Lukrafka, J. L. (2015). Chronic electroacupuncture of the ST36 point improves baroreflex function and haemodynamic parameters in heart failure rats. *Autonomic Neuroscience: Basic and Clinical*. https://doi.org/10.1016/j.autneu.2015.05.002

Macova, M., Pavel, J., & Saavedra, J. M. (2009). A peripherally administered, centrally acting angiotensin II AT2 antagonist selectively increases brain AT1 receptors and decreases brain tyrosine hydroxylase transcription, pituitary vasopressin and ACTH. *Brain Research*. https://doi.org/10.1016/j.brainres.2008.11.006

Perini, J. L., Scotta Hentschke, V., Sonza, A., & Dal Lago, P. (2016). Long-term low-level laser therapy promotes an increase in maximal oxygen uptake and exercise performance in a dose-dependent manner in Wistar rats. *Lasers in Medical Science*. https://doi.org/10.1007/s10103-015-1849-8

Pfeffer, M. A., Pfeffer, J. M., Fishbein, M. C., Fletcher, P. J., Spadaro, J., Kloner, R. A., & Braunwald, E. (1979). Myocardial infarct size and ventricular function in rats. *Circulation Research*. https://doi.org/10.1161/01.RES.44.4.503

Ponikowski, P., A. Voors, A., D. Anker, S., Bueno, H., G. F. Cleland, J., J. S . Coats, A., … Van der Meer, P. (2017). 2016 ESC GUIDELINES FOR THE DIAGNOSIS AND TREATMENT OF ACUTE AND CHRONIC HEART FAILURE. *Russian Journal of Cardiology*, *141*(1), 7–81. https://doi.org/10.15829/1560-4071-2017-1-7-81

Poole, D. C., Hirai, D. M., Copp, S. W., & Musch, T. I. (2012). Muscle oxygen transport and utilization in heart failure: implications for exercise (in)tolerance. *AJP: Heart and Circulatory Physiology*. https://doi.org/10.1152/ajpheart.00943.2011

Quagliotto, E., Casali, K. R., Dal Lago, P., & Rasia-Filho, A. A. (2015). Neuropeptides in the posterodorsal medial amygdala modulate central cardiovascular reflex responses in awake male rats. *Brazilian Journal of Medical and Biological Research*. https://doi.org/10.1590/1414-431X20144095

Quagliotto, E., Neckel, H., Riveiro, D. F., Casali, K. R., Mostarda, C., Irigoyen, M. C., … Rasia-Filho, A. A. (2008). Histamine in the posterodorsal medial amygdala modulates cardiovascular reflex responses in awake rats. *Neuroscience*. https://doi.org/10.1016/j.neuroscience.2008.09.053

Rigatto, K., Casali, K. R., Shenoy, V., Katovich, M. J., & Raizada, M. K. (2013). Diminazene aceturate improves autonomic modulation in pulmonary hypertension. *European Journal of Pharmacology*. https://doi.org/10.1016/j.ejphar.2013.04.017

Struthers, A. D., & MacDonald, T. M. (2004). Review of aldosterone- and angiotensin II-induced target organ damage and prevention. *Cardiovascular Research*. https://doi.org/10.1016/j.cardiores.2003.11.037

Szczepanska-Sadowska, E., Czarzasta, K., & Cudnoch-Jedrzejewska, A. (2018). Dysregulation of the Renin-Angiotensin System and the Vasopressinergic System Interactions in Cardiovascular Disorders. *Current Hypertension Reports*. https://doi.org/10.1007/s11906-018-0823-9

Triposkiadis, F., Karayannis, G., Giamouzis, G., Skoularigis, J., Louridas, G., & Butler, J. (2009). The Sympathetic Nervous System in Heart Failure. Physiology, Pathophysiology, and

Clinical Implications. *Journal of the American College of Cardiology*. https://doi.org/10.1016/j.jacc.2009.05.015

Zhang, D. Y., & Anderson, A. S. (2014). The sympathetic nervous system and heart failure. *Cardiology Clinics*. https://doi.org/10.1016/j.ccl.2013.09.010

Table 1. Photobiomodulation therapy (PBMTT) and Neuromuscular Electrical Stimulation (NMES) protocols

Protocols	
Photobiomodulation therapy (PBMT)	GaAlAs (850nm)
Frequency	Continuous output
Optical output	50 mW
Application area or spot size	0,2 cm^2
Power density	0,25 W/cm^2
Energy (per point)	3 J
Energy density (per point)	15 J/cm^2
Application time (per point)	60 s
Number of irradiation points per muscle group	2
Total energy delivered per muscle group	6 J
Application mode	LED device positioned at 90º angle in skin contact without moving and with light pressure
	Device parameters
NMES (FES VIF 995, Quark, Piracicaba, Brazil)	I = 70 mA; T = 40 µs - 250 µs; F = 4 Hz - 200 Hz; Alternating Current
Frequency (F)	30 Hz
Application time (t)	30 min
Duty cycle	8 s ON – 4 s OFF
Pulse duration (T)	250 µs
Waveform	asymmetric bipolar
Intensity (I)	Enough to generate visible muscle contraction

Table 2. Body, heart and ventricular weight ratio, tissue and infarct characteristics of Sham-operated rats and rats with Heart Failure

Measurement	Sham (n=6)	Control-HF (n=5)	NMES-HF (n=6)	PBMT-HF (n=6)	NMES + PBMT-HF (n=6)	*P* (GLM)
Initial Body Mass (g)	258 ± 18.2	248 ± 19.1	255.0 ± 25.9	232.0 ± 28	252 ± 9.67	0.27
Final Body Mass (g)	311 ± 7.97	306 ± 20.9	310 ± 36	291 ± 34	319 ± 11.5	0.427
Infarcted Area (%) §	---------	25.1 ± 4.55	36.2 ± 6.75 *	24.9 ± 2.59	27.1 ± 2.13	0.001
H/BM (mg/g)	3.45 ± 0.65	3.43 ± 0.62	3.86 ± 0.63	3.44 ± 0.36	3.76 ± 0.46	0.531
LV/BM (mg/g)	2.52 ± 0.36	2.55 ± 0.46	2.8 ± 0.35	2.62 ± 0.24	2.56 ± 0.16	0.606
RV/BM (mg/g)	0.94 ± 0.4	0.88 ± 0.21	1.06 ± 0.39	0.98 ± 0.16	1.2 ± 0.45	0.577
Pulmonary Congestion (%)	73.2 ± 1.6	75.6 ± 2.88	77.4 ± 1.62 †	75.6 ± 1.58	76.7 ± 1.35 †	0.005
Hepatic Congestion (%)	72.8 ± 1.79	72.3 ± 0.18	73.1 ± 0.9	72.1 ± 0.73	71.7 ± 1.18	0.26

Values are means ± SD. Control-HF = Heart Failure Control group; NMES - HF = Neuromuscular Electrical Stimulation Heart Failure group; PBMT-HF = Photobiomodulation Therapy Heart Failure group; NMES + PBMT-HF = Electrical Stimulation plus Photobiomodulation Therapy Heart Failure group. H/BM: heart-to-body mass ratio; LV/BM: left ventricular-to-body mass ratio; RV/BM: right ventricular-to-body mass ratio. GLM: General Liner Model followed by Bonferroni post hoc test. § The Sham group was not included in the analysis. * $P \leq 0.01$ compared to Control-HF, PBMT-HF, and NMES + PBMT-HF; † $P < 0.05$ compared to Sham;

Table 3. Power spectral analysis of cardiovascular parameters in awake rats

Measurement	Sham (n=6)	Control-HF (n=5)	NMES-HF (n=6)	PBMT-HF (n=6)	NMES + PBMT-HF (n=6)	*P* (GLM)
HRV, ms²	45.8 ± 14.5	44.6 ± 14.2	56.6 ± 18.7	33.5 ± 15.5	47.7 ± 39.9	0.557
$f_{LF,\ Hz}$	0.25 ± 0.05	0.22 ± 0.04	0.23 ± 0.05	0.23 ± 0.08	0.27 ± 0.12	0.87
LF, ms²	8.9 ± 1.85	10.3 ± 5.17	9.15 ± 4.8	7.22 ± 4.57	9.77 ± 9.91	0.923
Lfnu	0.42 ± 0.17	0.52 ± 0.16	0.32 ± 0.04	0.42 ± 0.15	0.4 ± 0.06	0.174
$f_{HF,\ Hz}$	1.5 ± 0.43	1.26± 0.25	1.53 ± 0.28	1.53 ± 0.75	1.35 ± 0.54	0.846
HF, ms²	16 ± 11.8	8.6 ± 2.16	20.9 ± 11.3	10.2 ± 6.04	18.1 ± 23.3	0.502
Hfnu	0.58 ± 0.17	0.48 ± 0.16	0.68 ± 0.04	0.58 ± 0.15	0.60 ± 0.06	0.174
LF/HF	1 ± 0.83	1.34 ± 0.71	0.48 ± 0.1	0.87 ± 0.4	0.77 ± 0.2	0.129
VLF, ms²	20.6 ± 5.03	25.4 ± 10.4	26.4 ± 7.23	15.8 ± 8.27	19.6 ± 7.71	0.161

Values are means ± SD. Control-HF = Heart Failure Control group; NMES-HF = Neuromuscular Electrical Stimulation Heart Failure group; LEDT - HF = Light Emitting Diode Therapy Heart Failure group; NMES + PBMT-HF = Neuromuscular Electrical Stimulation plus Photobiomodulation Therapy Heart Failure group. HRV: Heart Rate Variability; LF: Low Frequency Component; HF: High Frequency Component; LF/HF: Low and High Frequency Ratio; VLF: Very Low Frequency. GLM: General Liner Model followed by Bonferroni post hoc test.

Table 4. Hemodynamic variable

Measurement	Sham (n=6)	Control-HF (n=5)	NMES-HF (n=6)	PBMT-HF (n=6)	NMES + PBMT-HF (n=6)	*P* (GLM)
SAP (mmHg)	132 ± 7.97	120 ± 8.14	117 ± 5.31 †	123 ± 10.2	113 ± 5.39 *	0.003
DAP (mmHg)	90 ± 6.22	85.9 ± 5.79	85.9 ± 5.79	87.9 ± 5.83	81.7 ± 5.16	0.147
MAP (mmHg)	109 ± 6.54	101 ± 6.71	99.2 ± 6.07	104 ± 7.41	95.7 ± 5.37 *	0.025
HR (bpm)	458 ± 21.4	527 ± 52.1	446 ± 39.6 ¥	498 ± 38.2	444 ± 35.4 ¥	0.004
LVEDP (mmHg)	4.42 ± 1.07	19.8 ± 5.86 †	23.2 ± 3.19 †	22.7 ± 6.21 †	21 ± 7.55 †	<0.001
LVSP (mmHg)	120 ± 27.8	129 ± 28.2	115 ± 20.7	115 ± 22.2	105 ± 14.6	0.548
+dP/dtmax (mmHg/s)	6441 ± 1997	5403 ± 1339	4640 ± 582	4943 ± 1179	4357 ± 1172	0.089
-dP/dtmax (mmHg/s)	-4238 ± 1098	-3813 ± 1270	-2861 ± 283.8	-3208 ± 1162	-2803 ± 680.4	0.07

Values are means ± SD. Control-HF = Heart Failure Control group; NMES - HF = Neuromuscular Electrical Stimulation Heart Failure group; PBMT-HF = Photobiomodulation Therapy Heart Failure group; NMES + PBMT-HF = Neuromuscular Electrical Stimulation plus Photobiomodulation Therapy Heart Failure group. SAP: Systolic Arterial Pressure; DAP: Diastolic Arterial Pressure; MAP: Median Arterial Pressure; HR: Heart Rate; LVEDP: Left ventricular end-diastolic pressure; LVSP: Left ventricular systolic pressure; $+dP/dt_{max}$: Left ventricular maximum change in pressure over time; $-dP/dt_{max}$: Left ventricular minimum change in pressure over time. GLM: General Liner Model followed by Bonferroni post hoc test. † $P<0.01$ compared to Sham; * $P<0.05$ compared to Sham; ¥ $P<0.05$ compared to Control-HF; # $P<0.05$ compared to NMES + PBMT-HF.

Table 5. Effect of NMES and PBMT on baroreflex curve parameters in awake rats

Measurement	Sham (n=6)	Control-HF (n=5)	NMES-HF (n=6)	PBMT-HF (n=6)	NMES + PBMT-HF (n=6)	*P* (GLM)
Upper plateau (bpm)	495 ± 57.3	494 ± 26.7	451 ± 39.9	484.5 ± 15.3	438 ± 54.1	0.093
Lower plateau (bpm)	313 ± 31	320 ± 61.2	344 ± 56.4	320 ± 28.2	333 ± 28	0.725
HR range (bpm)	182 ± 80.2	175 ± 65.2	107± 46.9	164 ± 23.8	105 ± 48.2	0.055
MAP_{50} (mmHg)	103 ± 3.52	100 ± 6.72	90.8 ± 12.5	104 ± 5.1 *	95.6 ± 7.45	0.043
Maximum gain (beats·min^{-1}·mm Hg^{-1})	-9.07 ± 4.91	-7.69 ± 4.17	-12.5 ± 5.86	-6.8 ± 1.56	-9.83 ± 4.78	0.27

Values are means ± SD. Control-HF = Heart Failure Control group; NMES-HF = Neuromuscular Electrical Stimulation Heart Failure group; PBMT-HF = Photobiomodulation Therapy Heart Failure group; NMES + PBMT-HF = Neuromuscular Electrical Stimulation plus Photobiomodulation Therapy Heart Failure group. MAP_{50}, MAP that corresponds to the value found at half of the HR range evoked by the baroreflex response. GLM: General Liner Model followed by Bonferroni post hoc test. * Compared to NMES-HF, not significant difference; p = 0.075.

FIGURE LEGENDS

Figure 1. Flowchart of design

Figure 2. Experimental timeline of the study

Figure 3. Plots of logistic analysis of the sigmoidal baroreceptor curve, showing the mean values of the ratio between mean arterial pressure (MAP) and heart rate (HR) of rats Sham group; Control-HF = Heart Failure Control group; NMES-HF = Neuromuscular Electrical Stimulation Heart Failure group; PBMT-HF = Photobiomodulation Therapy Heart Failure group; NMES + PBMT-HF = Electrical Stimulation plus Photobiomodultaion Therapy Heart Failure group. In the upper right corner, they are showing the plot of baroreflex sensitivity between the same experimental groups. Data are presented as mean ± SD. GLM: General Liner Model followed by Bonferroni post hoc test.

Figure 4. Delta of blood pressure and heart rate during baroreflex sensitivity analysis of Sham group; Control-HF = Heart Failure Control group; NMES-HF = Neuromuscular Electrical Stimulation Heart Failure group; PBMT-HF = Photobiomodulation Therapy Heart Failure group; NMES + PBMT-HF = Electrical Stimulation plus Photobiomodulation Therapy Heart Failure group. Data are presented as mean ± SD.

Figure 1. Flowchart of design

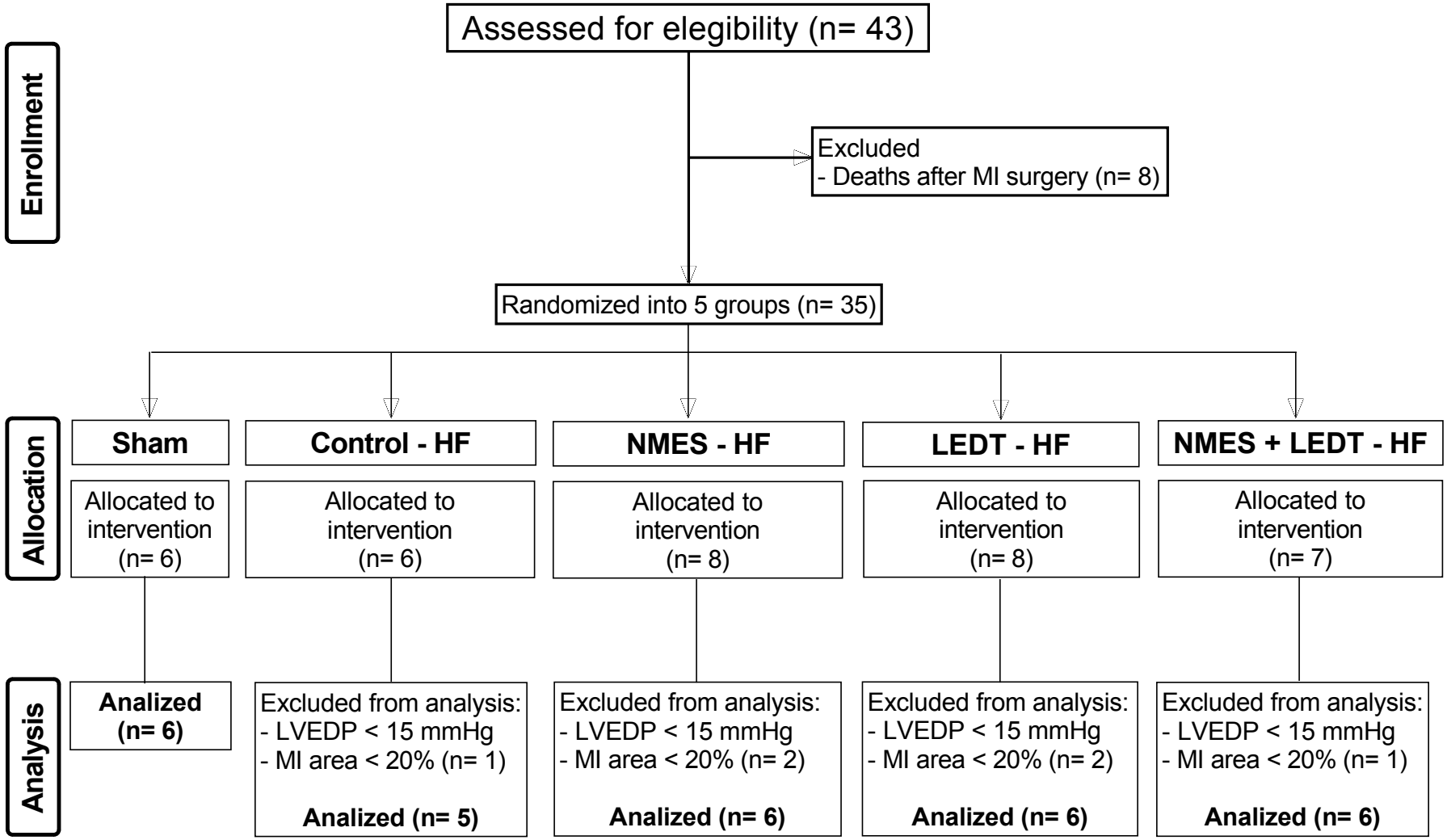

Figure 2. Experimental timeline of the study

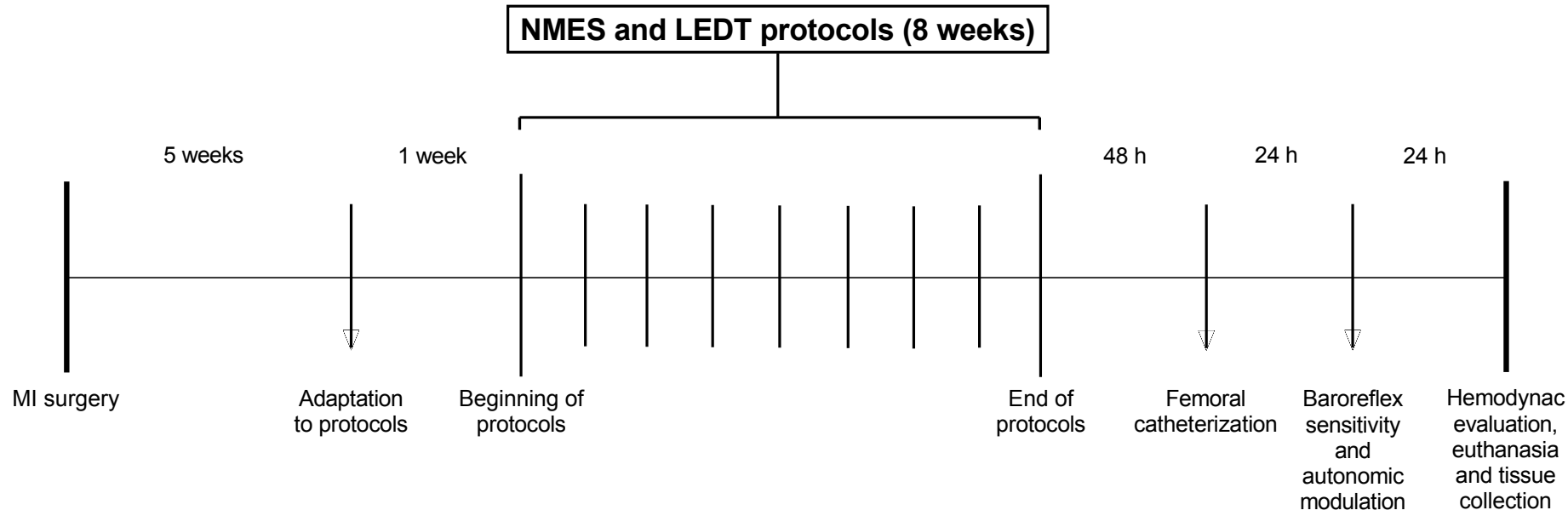

Figure 3. Baroreflex sensitivity

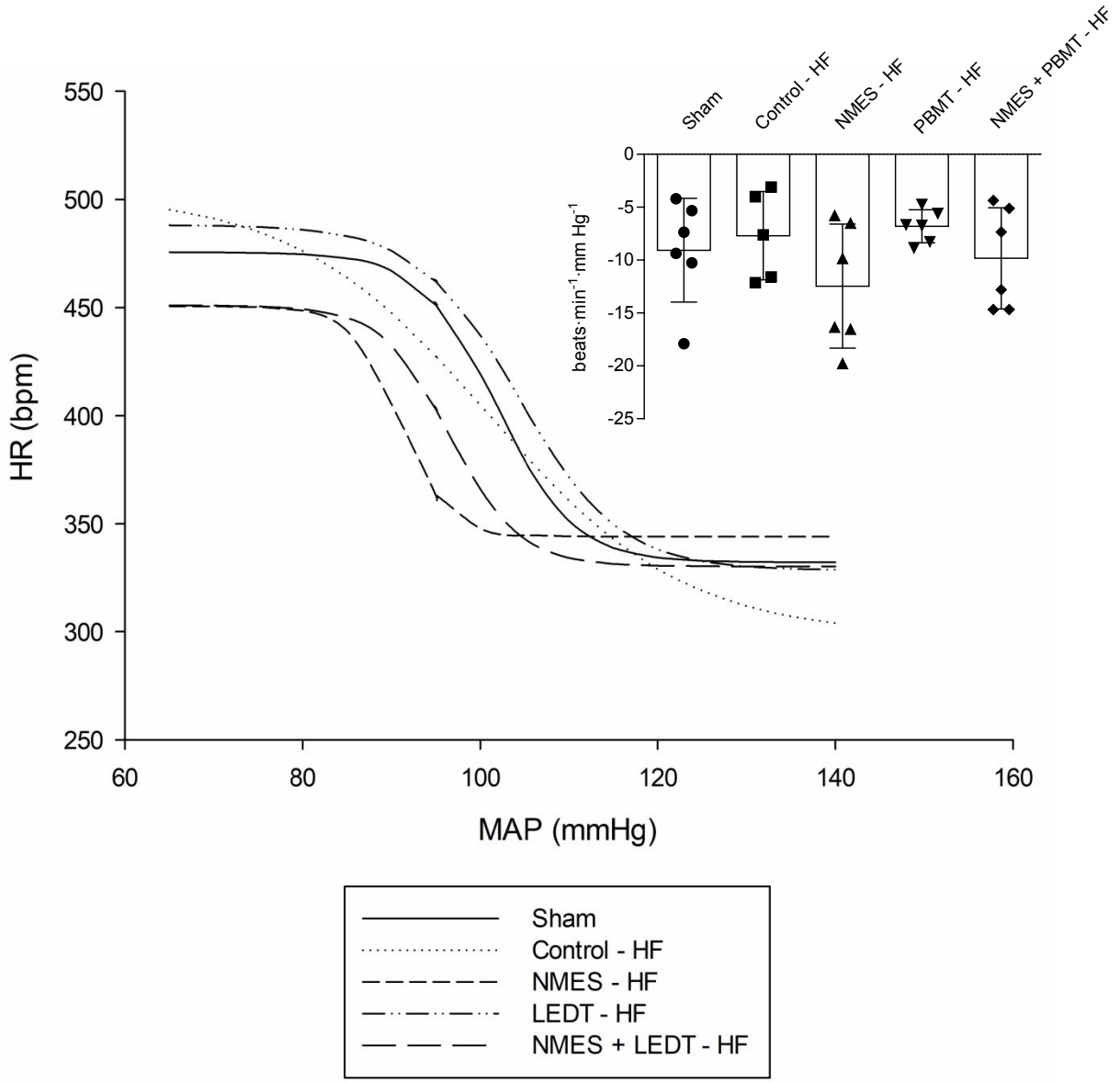

Figure 4. Delta of blood pressure and heart rate during baroreflex sensitivity analysis