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ABSTRACT

Purpose: The aim of this study was to investigate the hemodynamic, oxidative stress (OS), and nitric oxide (NO^-) responses to a submaximal isometric exercise session (IES) involving large muscle mass. **Methods***:* Fourteen hypertensive (HTG: age: 35.9 ± 8.1 years, height: 1.73 ± 0.10 m, total body mass: 78.0 ± 15.8 kg) and 10 normotensive (NTG: age: 41.1 ± 9.4 years, height: 1.71 ± 0.12 m, total body mass: 82.3 ± 22.4 kg) participants performed 2 experimental sessions in the leg press and bench press: i) control session and ii) 8 sets x 1 min contraction at 30% maximal voluntary isometric contraction with 2 min rest interval. Blood pressure (BP) was measured at rest and during 60 min postexercise. Blood samples were collected at rest, immediately after the session, and 60 min postexercise. NO^- was obtained through the Griess reaction method. OS parameters were analyzed using commercial kits. A repeated-measures ANOVA with Bonferroni's post hoc was used to analyze all dependent variables. **Results:** A significant decrease in systolic BP was observed only for HTG at 45 min and 60 min postexercise (baseline vs. 45 min: $p = 0.03$, $\Delta\% = 4.44\%$; vs. 60 min: $p = 0.018$, $\Delta\% = 5.58\%$). NO^- increased immediately postexercise only for HTG ($p = 0.008$, $\Delta\% = 16.44\%$). Regarding OS parameters, TBARS presented a significant reduction 60 min after the IES for NTG and HTG; catalase increased in both groups. **Conclusions:** The data showed that only 8 minutes of IES with a large muscle mass elicits an elevated pro-oxidant activity leading to a greater NO^- bioavailability, increases antioxidant reaction, and consequently reduces BP in hypertensive patients.

Key Words*:* Oxidative Stress; Blood Pressure; Postexercise Hypotension; Hypertension; Static Strength.

INTRODUCTION

The use of exercise as a recommendation and ongoing therapy to treat hypertension is well described in the scientific literature (1). Physical training such as aerobic exercise (2) and dynamic strength training (ST) (3) are the most studied and classically recommended. Changes in lifestyle, such as exercise, are associated with increasing nitric oxide (NO^-) bioavailability (4), reduction of oxidative stress (OS) biomarkers (e.g., plasma thiobarbituric acid-reactive substances, TBARS) and increased antioxidant capacity (5).

However, recently, the use of isometric handgrip (IHG, e.g., static ST) exercise in order to treat hypertension has gained attention in the scientific literature (6-9). Recent meta-analyses have suggested the efficacy of isometric exercise (IE) to chronically control and reduce blood pressure (BP) in normotensive, hypertensive, and prehypertensive adults (7, 9, 10).

Nevertheless, only a few studies have evaluated vasodilatation mechanisms for the acute exercise model such as OS (6, 10). Additionally, the peripheral vascular resistance is another factor contributing to hypertension that scientific evidence has been shown to be most affected by biochemical parameters, such as OS and NO^- (11).

A reduction in NO^- bioavailability may lead to endothelial dysfunction, vascular remodeling, and inflammation leading to vascular damage, which is also a major factor to enhance hypertension via increased peripheral vascular resistance. It is suggested that an increase in OS is one of the most important mechanisms associated with the pathophysiology of hypertension (12). In a study by Peters et al. (13), hypertensive patients performed 6 weeks of IHG and the authors reported an improvement in BP and antioxidant capacity and a reduction of reactive oxygen species (ROS) production.

Thus, the chronic IE protocol used in the study reduced OS (i.e., decreased TBARS) and improved antioxidant defense (i.e., increased catalase activity), which allowed a higher release of NO^- during recovery. Nevertheless, several studies have used IE with different acute and chronic protocols but with small muscle mass (handgrip) (6, 8, 14). Studies that compared resistance exercise with different muscle groups (upper and lower limbs), for instance, showed that dynamic resistance exercises that use larger muscle mass promote a greater magnitude in the reduction of BP (15).

However, no known scientific literature exists showing the acute cardiometabolic effects of IE when performed with large muscle mass (e.g., leg press [LEP] and bench press [BEP]) using conventional strength training equipment. This may suggest the versatility of this type of contraction during the treatment of the hypertensive population. Moreover, the influence of larger muscle mass on BP responses and the role of NO^- and redox balance in this process is still unclear.

Therefore, the purpose of this study was to investigate the hemodynamic, OS, and NO^- responses to 1 single bout of submaximal IE involving large muscle mass. The main hypothesis is that a submaximal isometric exercise session (IES) involving large muscle mass improves NO^- bioavailability, increases redox balance, and promotes reduction of BP levels in hypertensive patients.

MATERIALS AND METHODS

Participants

Twenty-four sedentary adult volunteers were recruited and separated, by convenience, into 2 groups: normotensive group (NTG; n = 10; 5 women and 5 men) and hypertensive group (HTG;

n = 14; 7 women and 7men) (Table 1). Volunteers answered a detailed anamnesis to assess history of physical exercise and medical and nutritional information. In addition, an expert cardiologist performed a cardiovascular diagnostic evaluation of the degree of hypertension. The inclusion criteria were (i) hypertension grade I and II (SBP 130-139 mmHg and DBP 80-89 mmHg for grade I; SBP ≥140 mmHg and DBP ≥90 mmHg for grade II) (16); (ii) continuous use of antihypertensive medication for at least 1 year; (iii) able to practice physical exercises; or (iv) not being enrolled in another physical activity program for at least 6 months. Volunteers were excluded if they presented (i) severe hypertension (SBP ≥ 180 mmHg and DBP ≥ 110 mmHg); (ii) any cardiologic changes during the exercise electrocardiogram, such as cardiac arrhythmias, extrasystoles, or other diagnoses by the cardiologist; (iii) target organ damage; (iv) any metabolic changes such as cancer and diabetes mellitus; or (v) bone, muscle, or joint problems that could prevent them from performing the exercises proposed in the study. Participants were oriented to maintain their regular nutritional habits and physical activities throughout the test period and refrain from caffeine, alcohol, and strenuous exercise 24 h prior to the test. All this information was verbally detailed. The university research ethics committee approved this study (#1.269.914/2015), and all subjects read and signed an approved informed consent document.

Study Design

This study was a randomized cross-over design. All participants were instructed to complete all sessions consisting of (i) a session to assess body composition and aerobic capacity; (ii) a session to obtain maximum voluntary isometric contraction (MVIC) in the LEP and BEP; (iii) an IES composed of 4 sets of submaximal contractions for each exercise with 1 min of duration at 30% of MVIC and 2 min of rest interval (hemodynamic variables were recorded at rest and 5, 10, 15, 30, 45, and 60 min postexercise); and (iv) a control session (CS; nonexercise) followed the same

procedures of hemodynamic and biochemical measurements as the exercise session. On the leg press equipment, they just stayed with their feet on the platform without movement or performing strength. While in bench press, participants just stayed lying down without moving; both positions were maintained for the same duration as the exercise session. Afterwards, they went to a quiet room for a 60 min recovery, where the final blood collection was done. Each session had an interval between 48 h and 72 h. For both sessions, the volunteers were instructed to maintain medication schedules and perform 8 h fasting in order to collect venous blood samples (~20 mL). Two more blood samples were collected (immediately after the exercise session and 60 min postexercise) in both sessions. To avoid a possible interference from eating with blood markers, especially in OS variables (17), after the fasting blood sample collection, all participants had a standard breakfast 30 min before every session (exercise and nonexercise), with chocolate milk (185 kcal total – 32 g carbohydrate, 3.9 g protein, and 7.7 g fat) and cereal bar (334 kcal total – 65 g carbohydrate, 7.7 g protein, and 5.6 g fat).

Measurements

Body Composition: Body mass (Filizola, São Paulo, Brazil), height, and body circumferences (Sanny, São Paulo, Brazil) were measured. Body fat was estimated by Jackson and Pollock's 3-sites skinfold protocol for adults (18) using a skinfold caliper (Lange, Santa Cruz, CA, USA).

Aerobic Capacity: A 12-lead electrocardiograph (Micromed Elite, Brasília, DF, Brazil) was used to record the maximal heart rate during the test using the modified Balke protocol, which was performed to rule out cardiovascular diseases. Arterial BP was measured during the test by auscultation. To determine the peak oxygen consumption (VO_2 peak) the Balke protocol was used. The running velocity was selected according to previous familiarization on the treadmill (Imbramed Super ATL, SP, Brazil) based on the individual physical capacity reported. All VO_2

values were recorded by a gas analyzer (Córtex Biophysik, Metalyzer 3B, Germany) every 30 s. Then, the VO_2 peak was calculated based on the plateau of oxygen consumption with increasing work rate and a respiratory exchange ratio greater than 1:1.

Maximal Voluntary Isometric Contraction (MVIC): A Power Din Pro dynamometer (CEFISE®, Campinas, Brazil) was used to measure the MVIC in the LEP 45º (Righetto, Campinas, Brazil). The load cell with an accuracy of 200 kgf (CEFISE®, Campinas, Brazil) and sampling frequency of 100 Hz was coupled in parallel to the load implement axis of the equipment and perpendicular to the plate for LEP. The MVIC in BEP (Biodelta-Portico, São Paulo, Brazil) was performed with an initial load of 90% of 1 RM where the participant should maintain the isometric contraction for 3 s; up to 5 trials were performed with a 10% load increment and 3 min interval at each attempt. A fleximeter (Sanny®, São Paulo, Brazil) was used to adjust the knee angle at 90º for leg press and the elbow angle at 90º for bench press. A warm-up was performed prior the LEP and BEP CVIM tests, consisting of 2 sets of 10-12 repetitions performed at 50% 1 RM with 2 min of rest pause between the sets for activation of muscles and joints involved in the test. After 2 min, all participants performed a set of familiarization with maximal isometric contractions for 5 s and 3 min of pause. For the determination of the MVIC for LEP, participants were instructed to push the LEP and hold the maximal force for 5 s. Tests were repeated 3 times with 3 min of pause, and all contractions were monitored in real time to ensure a good assessment; the major values were considered to MVIC. A researcher (R.R.O.) controlled the correct technique and promoted verbal encouragement during each muscle contraction. The maximal value of MVIC of both tests was used for further analysis.

Measurement of Blood Pressure and Heart Rate: BP and heart rate (HR) were assessed before (10-minat rest in sitting position) and during the session as a safety parameter for the participant,

after each contraction for, and after 5-, 10-, 15-, 30-, 45- and 60-min post sessions in supine position. Oscillometer equipment (Microlife, BP3AC1-1PC, Switzerland) was used to measure BP according to the AHA recommendations (19). HR was also continuously recorded by telemetry using a portable heart monitor (Polar, RS800 CX, Finland). Mean arterial pressure (MAP) was calculated as Moraes et al.(3). Rate-pressure product (RPP) was calculated by multiplying HR by SBP.

Biochemical Analysis: Venous blood samples were obtained from the forearm before each session (control and exercise) after fasting for at least 8 h, immediately after the session, and 60 min postexercise. After data collection, the blood samples were immediately processed in a refrigerated centrifuge to obtain plasma and serum (4°C for 15 min at 2500 rpm). Plasma and serum samples were stored in multiple aliquots (Eppendorf, Germany) at -80°C. All assays were performed within 2 months of sample collection, in duplicate, and at the first defrost. Plasma samples were analyzed to determine NO^-, superoxide dismutase activity (SOD), catalase activity (CAT), total antioxidant capacity by the Trolox-equivalent antioxidant capacity (TEAC), lipid peroxidation by the TBARS, glutathione reduced (GSH), and uric acid. The lipid profile was used to estimate total cholesterol, low-density lipoprotein cholesterol (LDL-c), high-density lipoprotein cholesterol (HDL-c), HDL-c/LDL-c ratio and triglycerides using an automated chemistry analyze (COBAS c111 system, Roche Diagnostics, Switzerland).

Nitric Oxide (NO^-): NO^- was measured using a Griess reaction, according to the following protocol. Plasma samples were deproteinized with zinc sulfate (20%). Then 300 µL of each sample was dispensed in duplicate in a 96-well plate, and 100 µL of vanadium chloride, 50 µL sulfinyl amide, and 50 µL of N-(1-Naphthyl) ethylenediamine dihydrochloride were added to the samples, and standard nitrite curves were prepared. After the addition of the reagents, the plates

were homogenized and incubated for 40 min at 37°C. The spectrophotometer reading was made at 540 nm. The coefficients of variability were <6%.

Lipid Peroxidation: Serum samples were diluted in 320 μL MiliQ H2O (1:5), and then 1 mL of 5 trichloroacetic acid (TCA) 17.5%, pH 2.0, and 1mL of thiobarbituric acid 0.6%, pH 2.0, were added, respectively. After homogenization, the samples were kept in a water bath for 30 min at 95°C. The reaction was interrupted with the immersion of the microtubes in ice, the addition of 1 mL of TCA 70%, pH 2.0, and incubation for 20 min at room temperature. After centrifugation (3000 rpm for 15 min), the supernatant was removed and put in new microtubes and read by spectrophotometry at 540 nm. The concentration of lipid peroxidation products was calculated using the molar extinction coefficient equivalent for malondialdehyde (MDA-equivalent = $1.56 \cdot 105\ M^{-1}cm^{-1}$). The coefficients of variability were <6%.

Antioxidant Parameters: The antioxidant parameters used in this study were measured using commercial kits and following the manufacturers' protocols. SOD activity was measured using an assay kit (Sigma Aldrich®, CA, USA) with a final spectrophotometric reading at 450 nm. Total antioxidant capacity was measured with a Trolox-equivalent assay kit (Quanti Chrom® Bio Assay Systems, CA, USA). CAT activity was measured using the Amplex TM Red assay kit (Thermo fisher Scientific®, USA) with a final spectrophotometric reading with 1 min incubation at 560 nm. Glutathione was measured using an assay kit (Sigma Aldrich®, CA, USA) with a final spectrophotometric reading at 490 nm. Serum uric acid was measured following the manufacturer's recommendations (Labtest®, MG, Brazil). Briefly, in a 96-well microplate, 20 μL of sample and 1000 μL of working reagent were added to each well. After homogenizing and incubating at 37°C for 5 min, the absorbance was determined at 505 nm in a microplate reader (ELx800, Bio Tek, USA). For all antioxidant parameters the intraassay coefficient of variation was <4%.

Statistical Analysis

The normality and homogeneity of variances within the data were confirmed with the Shapiro-Wilk and Levene tests, respectively. All data were expressed as mean and (±) standard deviation. Repeated measures ANOVA was used for comparisons between and within groups. The statistical power (1-β) was calculated a priori for all procedures considering an alpha of 5% and an effect size (f = 0.3) for OS and NO^- (1-β = 0.89) and (f = 0.25) for hemodynamic measures (1-β = 0.94), besides a specific statistical model employed (ANOVA for repeated measures with interaction between and within subjects). Cohen's formula for effect size (*d*) was calculated, and the results were based on the following criteria: trivial (<0.2), small (0.2-0.6), moderate (0.6-1.2), large (1.2-2.0), and very large (>2.0) effects (20). An alpha of 5% was used to determine statistical significance. All procedures were performed by the Statistical Package for Social Sciences (SPSS 21.0) and GraphPad Prism 6.0 (San Francisco, CA, USA).

RESULTS

The participants' characteristics were different between groups. On the anthropometrics, HTG showed higher value for waist-to-height ratio than NTG, and no other differences were identified in body composition parameters. For hemodynamic variables, the NTG presented lower values of SBP, DBP, and mean arterial pressure when compared to HTG. Regarding the lipid profile measurements, only LDL presented a significant difference between groups, with higher values for HTG. On the neuromuscular parameters, no differences were identified between groups (Table 1).

Concerning BP responses after CS and IES sessions, an increase in DBP was identified in the NTG after 30 min of the CS. Furthermore, the IES presented a significant reduction in SBP only in the HTG baseline vs. 45 min and 60 min (131.86 ± 2.54 to 126.0 ± 2.59 mmHg, p = 0.03 and 125.93 ± 2.54 mmHg, p = 0.018, respectively) with a moderate effect size for 45-min (d = 0.61) and 60 min (d = 1.01) after IE (Figure 1).

Regarding HR and rate-pressure product, statistical differences in the within-group comparisons were identified during recovery in the CS and after IES (Figure 2), showing a significant reduction only for NTG in HR.

Individual data and delta values for SBP in HTG and NTG after CS and IES are presented in Figure 3.

Results from biochemical analysis showed that the HTG (126.21 ± 23.34 μM) presented higher values of NO^- at baseline when compared to NTG (103.17 ± 6.07 μM) with a very large effect size (d = 1.33), and a significant increase after IE (146.96 ± 31.67 μM) with a moderate effect size (d = 0.75). Both groups showed a significant decrease in TBARS after 60 min post-IE when compared with baseline (2.54 ± 0.81 to 1.89 ± 0.7 nmol/L with moderate effect size d = 0.745) and (2.61 ± 1.1 to 1.64 ± 0.37 nmol/L with very large effect size d=1.193) for NTG and HTG, respectively. Furthermore, NTG had a significant increase in CAT immediately after IES (34.72 ± 10.77 to 97.80 ± 8.33 $U \cdot mL^{-1}$, p=0.000 and d=6.55) and after 60 min (83.38 ± 17.78 $U \cdot mL^{-1}$, p = 0.000 and d = 3.31), and HTG had a similar increase immediately after IE (42.95 ± 12.88 to 78.36 ± 17.55 $U \cdot mL^{-1}$, p = 0.000 and d = 2.30) and after 60 min (79.87 ± 17.78 $U \cdot mL^{-1}$, p = 0.000 and d = 2.66), with higher values for NTG than HTG (Figure 4).

The NTG presented higher values of Trolox-equivalent antioxidant capacity than HTG at baseline and immediately after IE. The glutathione activity presented a significant increase at 60 after min of recovery only for NTG (Table 2).

DISCUSSION

The purpose of the present study was to investigate the effects of IES on cardiovascular and biochemical parameters in normo- and hypertensive individuals. A previous study showed that IES using a small muscle volume (i.e., handgrip) reduced the BP of elderly hypertensive patients (6). Nevertheless, the involvement of a larger muscle volume in this process on the hemodynamic behavior of normotensive and hypertensive individuals remains unclear. The main finding was the OS responses and NO^- bioavailability from both groups (NTG and HTG) were quite similar, but only in HTG was post-isometric exercise hypotension observed.

Isometric muscle contractions cause increased vascular resistance, and sessions have been prescribed with a great variety of training variables, such as volume, intensity, and rest between sets (6, 14, 21). These protocols promoted important systemic hemodynamic changes, such as autonomic control (22), improved local blood flow dilatation (23), and improved endothelial function (24). A previous study pointed out that IES involving a large muscle mass (e.g., leg) increased the release of lactate (25). It is known that lactate is involved in NO^- release, so such an event may have occurred in the present study, and is related to a decrease in BP (26). This process of isometric muscular contractions accompanied by rest intervals between sets induces ischemia-reperfusion in the adjacent region (25). This process may be involved in the pro- and antioxidant adjustments.

In this scenario, the combination of physical training with vascular occlusion induces ischemia-reperfusion and improves and alters the metabolism of NO^- (27). In general, all these observations presumably indicated that such mechanisms were involved in the increase of CAT activity (28), in what might reverberate in an increase in plasma NO^- and PIEH. It has been suggested that the decrease in BP immediately after IE may be due to greater perfusion of the muscular blood flow generated by the release of vasodilator metabolites (25, 29); such inferences occurred partially in the present study supported by the increase in NO^- release and CAT activity. A low NO^- contributes to the development and maintenance of hypertension and is related to endothelial dysfunction (30). However, HTG showed higher values of NO^- when compared to NTG, and IES induced increases in NO^- bioavailability. This may have occurred as a compensatory way to maintain the pressure state taking into consideration the age of all subjects of this study (young participants) (31). Additionally, the increase of NO^- could be explained by 2 major factors: (i) some pharmacological treatments for hypertension that may increase NO^- (30); and (ii) an exaggerated metabolic response to produce NO^- in order to control BP and to scavenge superoxide to reduce oxidative parameters as a defense mechanism (32).

Furthermore, increases in basal NO^- production may represent a better OS, which was observed after IHG training (13), and it is plausible to infer that the reduction of OS may lead to an increase in NO^- (24). In that matter, lipid peroxidation was augmented in both groups after IES, and since the first antioxidant response (SOD) did not increase with exercise, it is reasonable to infer that the ROS produced during exercise was blunted, especially by NO^- scavenging and cytosolic antioxidants. Indeed, CAT and GSH increased after IES (GSH only in the NTG), as well as the total antioxidant capacity.

The amount of NO^- available is determined by a balance between its formation and scavenging superoxide anion, which is the major responsible for the latter process; its roles in hypertension have been investigated and are well documented (33). In fact, NO^- produced in a pro-oxidant environment is abducted by free radicals, decreasing its plasma concentration. Thus, the IES used in the present study reduced OS (i.e., decrease in TBARS) and improved antioxidant defense (i.e., increased CAT activity), allowing a better release of NO^- during the recovery time in hypertensive subjects.

Some studies also investigated the acute effects of aerobic exercise (large muscle mass) on OS parameters (34, 35). The study by Farney et al. (34) was conducted with acute aerobic exercise and reported an increase in antioxidant activity immediately after exercise and during recovery. Furthermore, the study by Barili et al. (35) was conducted with 16 hypertensive women who performed 3 protocols of aerobic exercise, including 1 with blood flow restriction in a low-intensity. The authors reported elevated levels of lipid peroxidation, as well as SOD and glutathione-s-transferase 30 min after exercise, which partially corroborates with the present results.

The results of this study demonstrated that IES did not alter the activity of SOD and GSH, but increased CAT activity during recovery. Such increase is important because CAT participates in the reduction of hydrogen peroxide (H_2O_2) to water and carbon dioxide, thereby reducing the pro-oxidant potential of the H_2O_2 (36). Thus our data indicate that IES with greater muscle mass at 30% of MVIC increases the activity of blood catalase, which corroborates with the evidence that exercise improves plasma (37) and muscle CAT activity (38).

In addition, the increased activity of antioxidant enzymes appears to be influenced by muscle group size involved during exercise (39). It is possible to think that the greater the muscle mass

involved during exercise, the higher BP will be. This would allow us to deduce that greater muscle mass would increase tissue hypoxia. Therefore, it can be presumed that the increase of the antioxidant activity could be a protective effect against the stimulus of the tissue hypoxia. It is reported that increased CAT activity in plasma after periods of occlusion increase the levels of H_2O_2 induced by ischemia-reperfusion. These data from the literature suggest that even brief periods of elevated intraluminal pressure raise H_2O_2 concentration, which provides a compensatory dilator mechanism with NO^- participation (40). It is likely that ischemia-reperfusion processes induced by IE improve vascular function due to increased NO^- bioavailability related to increased CAT activity.

Scientific literature is limited regarding the acute effects of IES on OS parameters, especially in hypertensive individuals. In a study by Alessio et al. (41) 12 subjects underwent an exhaustive aerobic exercise and IE under the hypothesis that the anaerobic nature of IE might have presented lower central stress and less ROS production. The authors reported an increase in lipid hydroperoxides only after the isometric session and elevated protein carbonyls and antioxidant capacity only after the aerobic session, with no significant changes in malondialdehyde levels. In addition, the authors used an IHG exercise that involves much less muscle mass than the leg press used in the present study, which makes it difficult to reach accurate direct comparisons and the involved mechanisms. Regarding the present results, it is important to note that the postexercise blood samples were taken in a fed state, which could possibly lead to biased OS results. However, both control and exercise sessions were performed in the same conditions, and a fed state could increase lipid peroxidation, acutely increasing the OS, and the antioxidant effect of exercise was even more prominent.

Additionally, this isometric program does not require qualified instruction in exercise technique, and only two upper and lower body exercise equipment can easily be structured in a small space and with low cost in relation to isokinetic dynamometers or a fitness center. This possibility of adjustment has a very important application for those living in confined spaces and for other patients with limited mobility. Therefore, it is of practical value for coaches and health professionals to apply this model of strength exercise as an adjuvant strategy to improve muscular strength and hypertension treatment.

In conclusion, results of this study showed the first evidence that submaximal IES with greater muscle mass reduced BP levels for at least 1 h after the 8 min session and was well tolerated by the hypertensive patients. Furthermore, that model of exercise elicits an elevated pro-oxidant activity leading to a greater NO^- bioavailability, increases antioxidant reaction, and consequently reduces BP in hypertensive individuals. Further studies could increase the period of BP monitoring as well as the analysis of other OS parameters.

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Author contributions

The contributions of each author will be described below

RRO, TSR, PHM, and MRM conceived and designed the research.

RRO, TSR, RVN, LHS, TBR, BRS, IRS, LAD, and CVS conducted experiments. (maximal effort tests and experimental tests)

RRO, TSR, RVN, LHS, TBR, BRS, IRS, LAD, HGS and CVS conducted the laboratory tests (biochemistry and oxidative stress)

RRO, TSR, HGS, PHM, CVS and MRM contributed to all data analysis.

RRO and TSR wrote the manuscript.

RRO, TSR, PHM and MRM Significant manuscript reviewer/reviser

All authors read and approved the manuscript.

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Figure captions

Figure 1. Blood pressure responses to control session (A) and submaximal isometric exercise session (B) in normotensive (NTG) and hypertensive (HTG) adults; SBP: systolic blood pressure; DPB: diastolic blood pressure; MAP: mean arterial pressure; †: statistical difference from baseline (p<0.05); *: statistical difference between groups (p<0.05). Data expressed as mean and (±) standard deviation.

Figure 2. Heart rate (HR) and rate pressure product (RPP) responses to control (A) and submaximal isometric exercise session (B) in normotensive (NTG) and hypertensive (HTG) adults; †: statistical difference from baseline (p<0.05); *: statistical difference between groups (p<0.05). Data expressed as mean and (±) standard deviation.

Figure 3. Individual data and delta values of systolic blood pressure (SBP) after control session and isometric exercise for hypertensive (HTG) and normotensive (NTG) subjects. A: control session for NTG; B: submaximal isometric exercise for NTG; C: control session in HTG; D: submaximal isometric exercise for HTG.

*Figure 4.*Nitric Oxide (NO^-), thiobarbituric acid-reactive substances (TBARS) and catalase (CAT) responses to control (A) and isometric exercise session (B) in normotensive (NTG) and hypertensive (HTG) adults; †: statistical difference from baseline (p<0.05); *: statistical difference between groups (p<0.05).Data expressed as mean and (±) standard deviation.

Figure 1

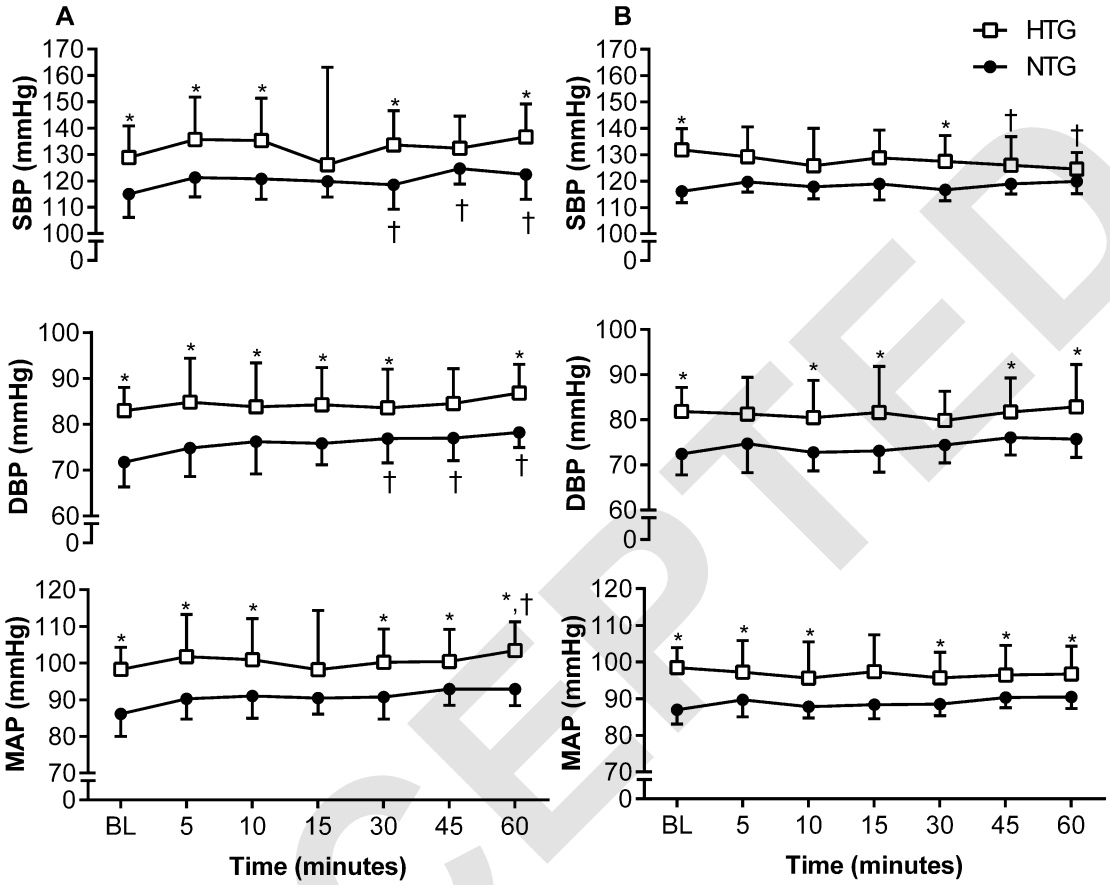

Figure 2

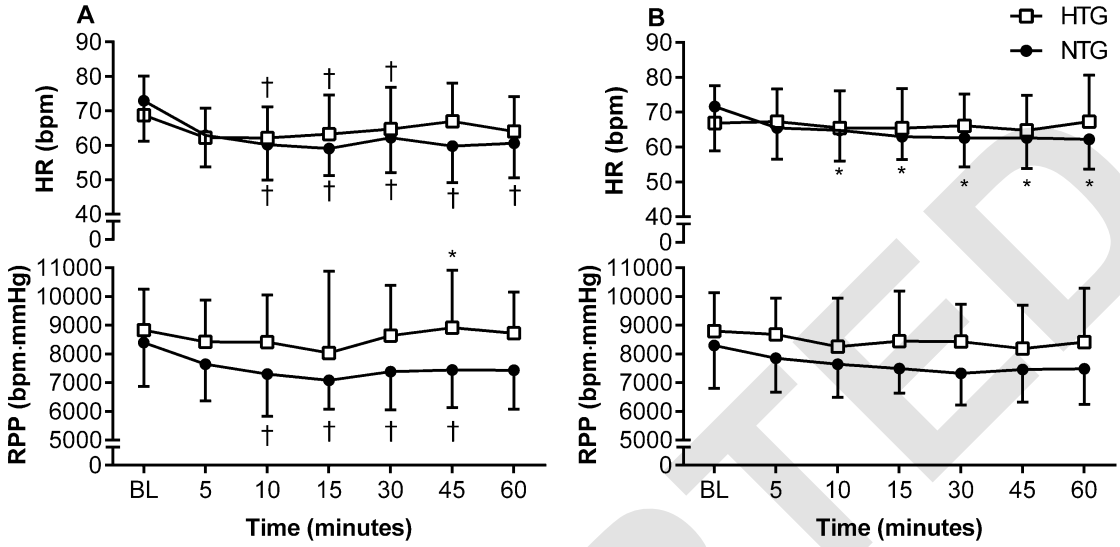

Figure 3

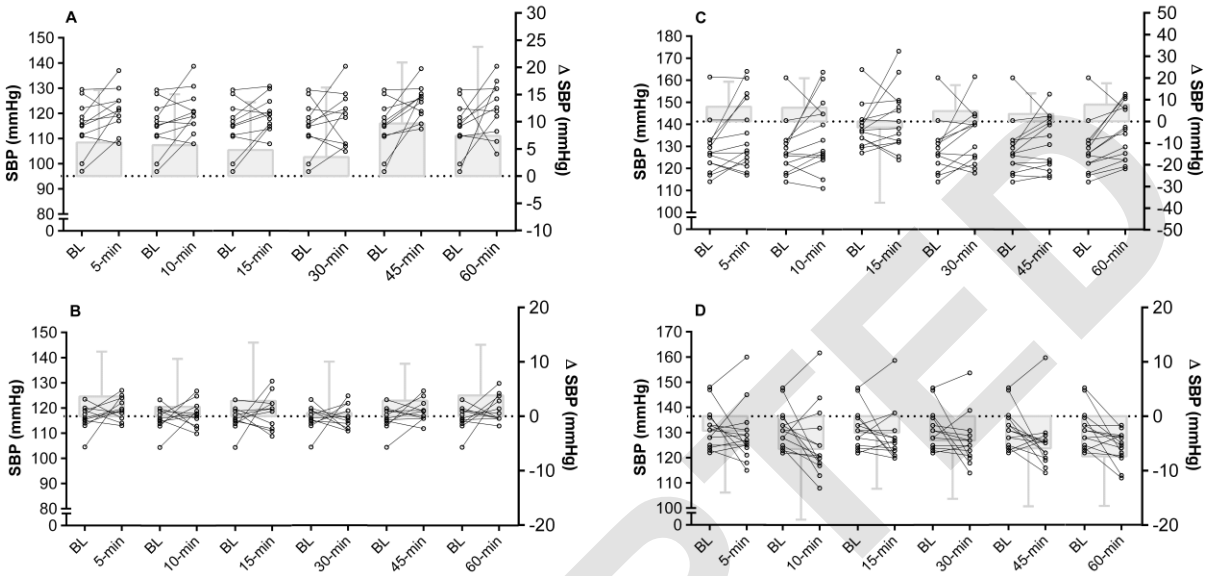

Figure 4

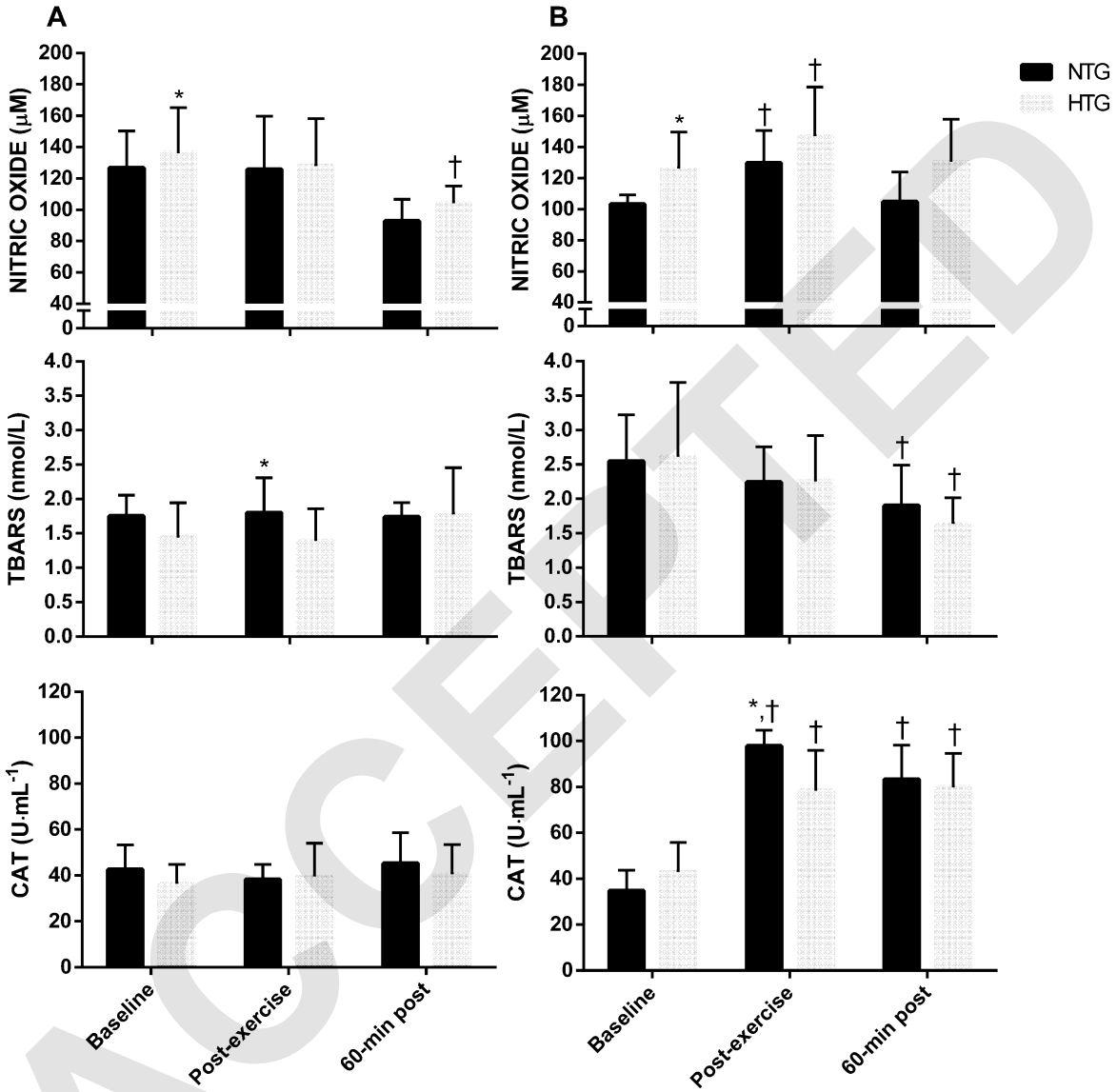

Table 1. Age, body composition, hemodynamics, neuromuscular and number of medication data for normotensive and hypertensive adults. Data expressed as mean and standard deviation (± SD).

	NTG (n = 10)	HTG (n = 14)	*p*-value
Anthropometrics			
Age (years)	35.9 (8.1)	41.1 (9.4)	0.182
Height (m)	1.73 (0.10)	1.71 (0.12)	0.620
Total body mass (kg)	78.0 (15.8)	82.3 (22.4)	0.601
BMI (kg/m^2)	26.4 (6.6)	28.2 (5.7)	0.512
Lean mass (kg)	29.0 (6.1)	32.2 (8.7)	0.334
Fat mass (kg)	20.8 (12.9)	21.8 (10.3)	0.849
Body fat (%)	25.8 (10.7)	25.3 (8.4)	0.918
Waist-to-hip ratio	0.81 (0.07)	0.88 (0.10)	0.131
Waist-to-height ratio	0.47 (0.04)	0.53 (0.07)	*0.019**
Neck circumference (cm)	35.8 (3.3)	38.0 (4.5)	0.223
Hemodynamic			
SBP (mmHg)	115 (10.65)	128.89 (11.95)	*0.007**
DBP (mmHg)	71.75 (6.51)	82.96 (5.08)	*0.000**
MAP (mmHg)	86.17 (7.38)	98.27 (9.31)	*0.000**
HR (bpm)	72.95 (14.16)	70.35 (9.31)	0.593
RPP (mmHg·bpm)	8391.05 (835.97)	9034.27 (1193.66)	0.308
Number of medications (%)			
Inhibitors of angiotensin-converting enzyme		14.29	
Angiotensin II receptor antagonist		21.43	

Diuretics		7.14	
Beta blocker		14.29	
Combination		14.29	
Don't use medications		28.57	
Lipid Profile			
Triglycerides (mg/dL)	82.34(41.66)	81.86 (45.68)	0.979
Cholesterol (mg/dL)	161.56 (28.23)	181.86 (31.10)	0.115
HDL (mg/dL)	46.02 (16.22)	41.76 (12.74)	0.478
LDL (mg/dL)	99.08 (21.71)	123.73 (31.71)	*0.045**
TG/HDL ratio	2.02 (1.3)	2.37 (1.92)	0.627
Cho/HDL ratio	3.74 (0.98)	4.82 (2.0)	0.130
Neuromuscular			
Bench Press			
MVIC (Kg)	42 (21.23)	46.46 (22.23)	0.598
MVIC relative (Kg/BM)	1.71 (0.94)	1.63 (0.75)	0.832
MVIC relative (Kg/LM)	2.24 (1.88)	2.12(1.32)	0.837
Leg Press			
MVIC (Kgf)	139.06 (51.23)	164.76 (71.27)	0.341
MVIC relative (Kgf/BM)	1.84 (0.72)	1.89 (0.66)	0.685
MVIC relative (Kgf/LM)	4.71 (1.19)	4.86 (1.37)	0.578

BMI: body mass index; SBP: systolic blood pressure; DBP: diastolic blood pressure; MAP: mean arterial pressure; HR: heart rate; RPP: rate pressure product; HDL: high density lipoprotein; LDL: low density lipoprotein; TG: triglycerides; Cho: Cholesterol; MVIC: maximal voluntary isometric contraction; BM: body mass; LM: lean mass; *: statistical significant (p<0.05).

Table 2. Oxidative stress parameters responses of control session and isometric exercise in normotensive and hypertensive adults. Data expressed as mean and standard deviation (± SD).

	Control Session						*Isometric Exercise*					
	NTG			*HTG*			*NTG*			*HTG*		
	BL	Post	60' Rec	BL	Post	60' Rec	BL	Post	60' Rec	BL	Post	60' Rec
GSH	6.61	6.26	5.89	5.51	5.90	5.95	5.61	6.21	6.77	6.38	6.35	6.00
(µM)	(1.03)*	(0.94)	(1.07)	(0.40)	(0.62)	(1.20)	(0.93)	(0.97)	(0.56) †	(1.10)	(0.94)	(1.19)
TEAC	1.44	1.66	1.46	1.40	1.61	1.64	1.88	1.94	1.92	1.64	1.61	1.64
(mM)	(0.45)	(0.48)	(0.47)	(0.48)	(0.44)	(0.49)	(0.52) *	(0.41) *	(0.36)	(0.31)	(0.2)	(0.45)
SOD	39.04	60.58	78.01	42.3	63.3	79.3	79.8	70.3	58.5	76.5	80.9	64.7
(U/mL)	(13.83)	(24.59)†	(5.16) †	(14.3)	(20.0) †	(6.9) †	(15.9)	(21.1)	(11.8)	(19.7)	(15.6)	(23.1)
Uric Acid	6.41	6.79	6.73	6.36	6.74	6.81	7.00	7.01	7.22	7.29	6.70	6.78
(mg/dL)	(0.59)	(0.98)	(0.98)	(1.25)	(1.01)	(1.21)	(1.51)	(1.88)	(2.01)	(1.72)	(1.0)	(1.36)

NTG: normotensive group; HTG: hypertensive group; BL: baseline; GSH: Glutathione; TEAC: trolox-equivalent antioxidant capacity; SOD: superoxide dismutase; * Statistical difference for between groups comparison(p<0.05); † Statistical difference from baseline for within-group comparison(p<0.05).