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Original article

REPRODUCIBILITY OF HEMODYNAMIC, CARDIAC AUTONOMIC MODULATION AND BLOOD FLOW ASSESSMENTS IN PATIENTS WITH INTERMITTENT CLAUDICATION

Running head: Cardiovascular function reproducibility in IC

Authors

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STRUCTURED ABSTRACT

Objective: To identify, in patients with peripheral artery disease and intermittent claudication (IC), the reproducibility of heart rate (HR), blood pressure (BP), rate pressure product (RPP), heart rate variability (HRV), and forearm and calf blood flow (BF) and vasodilatory assessments.

Methods: Twenty-nine patients with IC underwent test and retest sessions, 8-12 days apart. During each session, HR, BP, HRV, BF and vasodilatory responses were measured by electrocardiogram, auscultation, spectral analysis of HRV (low frequency, LF<sub>R-R</sub>; high frequency, HF<sub>R-R</sub>) and strain gauge plethysmography (baseline BF, post-occlusion BF, post-occlusion area under the curve, AUC). Reproducibility was determined by intraclass coefficient correlation (ICC), typical error, coefficient of variation (CV) and limits of agreement.

Results: The ICC for HR and BP were > 0.8 with CV < 9%. For most HRV measures, ICC were > 0.9 while CV were < 7%, except for LF/HF (ICC = 0.737; CV = 93.8%). The ICC for forearm and calf baseline BF assessments were > 0.9 while CV were < 19%; variable ICC and CV for vasodilatory responses were exhibited for calf (0.653 – 0.770; 35.2 – 37.7%) and forearm (0.169 – 0.265; 46.2 – 55.5%).

Conclusions: In male patients with IC, systemic hemodynamic (HR and BP), cardiac autonomic modulation (LF<sub>R-R</sub> and HF<sub>R-R</sub>) and forearm and calf baseline BF assessments exhibited excellent reproducibility, whereas the level of reproducibility for vasodilatory responses were moderate to poor. Assessment reproducibility has highlighted appropriate clinical tools for the regular monitoring of disease/intervention progression in patients with IC.

Key-words: peripheral artery disease; reliability; agreement; cardiovascular system; hemodynamic variables.
Peripheral artery disease (PAD) affects 200 million people worldwide with this population suffering from a partial or complete artery occlusion, primarily in the lower limbs \(^1\). Due to severe ischemia caused by increased oxygen demand, patients with PAD usually present with pain in the legs during walking that ceases with rest \(^2, 3\); a symptom referred to as intermittent claudication (IC) \(^4\). Patients who experience IC exhibit a reduced walking capacity and quality of life compared to the general healthy population \(^5\).

Apart from limb-specific impairments, patients with IC also present with several traditional cardiovascular risk factors \(^6, 7\), which contribute towards their high rates of cardiovascular events and mortality \(^7, 8\). Recent studies have examined other physiological mechanisms that may be linked with the increased cardiovascular disease burden in PAD. Compared to healthy adults, augmented blood pressure (BP) \(^8, 9\), decreased parasympathetic and increased sympathetic modulations to the heart \(^10\) as well as impairments in blood flow (BF) and vasodilation \(^11\) have been reported in patients with IC. Significant associations between hemodynamic, autonomic and vascular impairments with cardiovascular disease and mortality in PAD \(^12, 13\) have increased the clinical interest in evaluating these impairments to assist with early identification of increased cardiovascular risk for patients. However, the reproducibility of the tools for assessing these impairments must be confirmed before they can be consistently utilised in clinical practice.

Reproducibility is an important methodological psychometric property, especially for tracking changes over time, and crucial for identifying clinical meaningful changes for individuals \(^14\). In healthy \(^15, 16\) and clinical populations, such as those with hypertension \(^17, 18\) and diabetes mellitus \(^19\), electrocardiogram (ECG), auscultation, heart rate variability (HRV) and strain gauge plethysmography have been used reliably to assess heart rate (HR), BP, cardiac autonomic modulation, and BF and vasodilatory response, respectively. However, some of these measures have
been reported to be irreproducible in populations such as cardiac transplant and chronic heart failure patients suggesting that reproducibility may be population-specific. Furthermore, patients with IC usually present with a myriad of cardiometabolic risk factors, which further aggravates hemodynamic, autonomic and vascular dysfunction. This particular disease presentation may lead to unique vascular and cardiac autonomic responses that may affect the reproducibility of assessments, and subsequent clinical relevance of these measures and future cardiovascular risk prediction. Currently, the reproducibility of these important prognostic assessment tools for IC patients is unknown. Thus, the aim of this study was to evaluate the reproducibility of the assessments of HR, BP, rate pressure product (RPP), HRV and upper limb and leg BF and vasodilatory responses in patients with PAD and IC. These assessment tools provide clinicians with reliable indicators for the monitoring of disease progression and cardiovascular risk in patients with IC.

2. MATERIAL AND METHODS

2.1. Participants

Patients with PAD and IC symptoms and enrolled in a tertiary center specialized in vascular disease were invited to participate in this study. Patients were included if they met the following criteria: (1) male; (2) aged ≥ 50 years; (3) diagnosed with Fontaine stage II of PAD; (4) ankle brachial index at rest ≤ 0.90 in at least 1 lower limb; (5) resting systolic and diastolic BP lower than 160 and 105 mmHg, respectively; and (6) not receiving β-blockers or non-dihydropyridine calcium channel antagonists. The study’s protocol was conducted in accordance with the Declaration of Helsinki, registered with the Brazilian Clinical Trials (http://www.ensaiosclinicos.gov.br, RBR-3pq58k), and approved by the Joint Committee on Ethics of Human Research of the institution.
Written informed consent was obtained from all patients prior to study commencement.

2.2. Procedures

Figure 1 shows the timeframe of the four visits to the laboratory completed by participants during study. During the first visit, they were interviewed, and ankle brachial index, anthropometric and resting BP measures were taken. The interview identified age, the presence of cardiovascular disease and risk factors, comorbid conditions, and current medication treatment. Ankle brachial index was evaluated as previously described, and anthropometric measures of body mass and height (Welmy, 110, São Paulo, Brazil) were used to calculate body mass index (BMI). Auscultatory BP was measured in triplicate using a mercury column sphygmomanometer (Unitec, São Paulo, Brazil). Measures were taken after 5 min of rest in the seated position. During the second visit, resting BP was measured again and the mean of the 6 measures (visit 1 and 2) was defined as each participant’s resting BP. For the third and fourth visits (test and retest sessions), HR, BP, HRV, BF and vasodilatory response were measured 8-12 days apart and assessed for reproducibility.

***INSERT FIGURE 1***

2.3. Experimental protocol

The participants were instructed to avoid physical exercise for the previous 48 h, caffeinated and alcoholic beverages for the previous 24 h and smoking on the day of the sessions. They were also instructed to take their medication as prescribed by their physicians. The sessions were conducted throughout the year, with some participants taken part of the study in all the year seasons. Data was collected in a temperature-controlled laboratory (21 – 22°C).
The sessions started at 8:00 a.m and patients initially rested in the supine position for 50 min, including an initial stabilisation period of 10 min. ECG and breathing rate were recorded between 10 and 20 min, while BP and HR were measured in triplicate between 20 and 25 min with the mean value used for analysis. Lower and upper limb BF and vasodilatory response were determined during the last 25 min of supine rest.

2.4. Measurements

ECG (EMG System do Brazil, EMG 030110/00B, Brazil) was continuously monitored while auscultatory BP was measured in the dominant arm using a mercury sphygmomanometer (Unitec, São Paulo, Brazil). Mean BP was calculated as the sum of diastolic BP and one-third of pulse pressure (systolic – diastolic BP), while HR was recorded from the ECG immediately after BP measurement. RPP was calculated as HR x systolic BP.

For cardiac autonomic evaluation, R-R intervals from the ECG and respiratory signal from a thoracic piezoelectric belt (UFI, Pneumotrace2, California, USA) were inputted into a data acquisition system (WinDaq, DI-720, Akron, USA) at a sampling rate of 500 Hz/channel. Stationary segments (250 – 300 beats) of the HR series were assessed for HRV via spectral analysis (Heart Scope, version 1.3.0.1, New York, USA), employing an autoregressive method in accordance with Task Force recommendations (24). Low-frequency (LF_{RR}) (0.04 – 0.15 Hz) and high-frequency (HF_{RR}) (0.15 – 0.4 Hz) components of HRV were expressed in normalized units (nu) with these representing the relative contribution of each component within the total power spectrum minus the very low frequency component (24). Total variance (TV_{R-R}) (0 – 0.4 Hz) and LF/HF ratio were also calculated.

Upper and lower limb BF were simultaneously determined in the dominant forearm and the leg with the lowest ankle brachial index. BF was assessed by venous occlusion plethysmography (Hokanson, AI6, Bellevue, USA), as previously described (25). Briefly, BF to the hand and the foot were interrupted by limb occlusion via cuffs inflated to 200 mmHg around the wrist and the ankle,
respectively. Other cuffs were placed on the upper arm and the thigh and were rapidly inflated for 10 s at 40 – 60 mmHg, followed by 10 s of deflation of both cuffs. Increases in limb volumes were detected by mercury strain gauges positioned at the forearm and calf using specialised software (NIVP3; Hokanson, Bellevue, WA). Measurements were taken during 4 min (12 x 20 s cycles) and the mean was used to determine BF. Vascular resistance (VR) was determined as the ratio between mean BP and BF in each limb. Vasodilatory response to reactive hyperaemia was also assessed.

After BF determination, BF to each limb was totally occluded for 5 min by inflating the thigh and arm cuffs to 200 mmHg. The cuff was then released and post-occlusion BF was measured for 4 min as described above. The vasodilatory response was calculated by: 1) the first BF measured immediately after cuff release (post-occlusion BF) and 2) the differences in area under the curve (AUC) between post- and pre-hyperaemia BF measurements.

2.5. Statistical Analysis

Data normality was confirmed by Shapiro-Wilk test while the presence of heteroscedasticity was verified by significant correlations between the test-retest mean values and their absolute differences. Variables violating these assumptions were transformed via natural logarithm prior to further analysis.

Reproducibility was examined using the following parameters: 1) presence of systematic bias; 2) inter-test reliability; 3) within individual variation; and 4) bias with limits of agreement. Systematic bias was assessed as the difference between test and retest mean values using a paired t-test. Inter-test reliability was evaluated by the intraclass correlation coefficient (ICC) with values < 0.40 considered poor, 0.40 – 0.75 considered moderate to good, and > 0.75 considered excellent.

Within-individual variation was evaluated by the typical error (TE) between test and retest values, expressed both in absolute and relative terms (e.g. coefficient of variation – CV) with CV < 20% considered desirable and > 30% undesirable. Lastly, agreement was evaluated by the mean bias ±
95% limits of agreement (LOA) as previously described by Bland & Altman. For the logarithm-transformed data, the ratio bias */± 95% LOA was calculated as previously recommended.

3. RESULTS

Twenty-nine male patients with IC volunteered for this study and their characteristics are summarized in Table I. In general, patients were elderly (> 60 years), had an ankle brachial index between 0.45 and 0.90 (i.e. mild to moderate PAD), presented with several cardiovascular risk factors, and were taking a range of medications such as aspirin, statins, and anti-hypertensives.

***INSERT TABLE I***

Mean values of systemic hemodynamic variables measured at test and retest, and their reproducibility indices are shown in Table II. There were no significant differences between the mean values of any variable (P > 0.05), while ICCs ranged from 0.833 (diastolic BP) to 0.908 (HR) and CVs ranged from 5.0 (mean BP) to 8.3% (RPP). For each variable, the TE and mean/ratio bias were small while the LOA varied (Table II).

***INSERT TABLE II***

Mean values of all HRV variables were not significantly different between test and retest (P > 0.05, Table III) with ICCs ranging from 0.929 (ln TVR-R) to 0.986 [HF_{R,R} (nu)], and CVs from 4.9 (ln TV_{R,R}) to 93.8% (LF/HF). For each variable, the TE and mean/ratio bias were small while the LOA varied (Table III).
The results for BF variables are shown in Table IV. The mean values of most variables did not significantly differ between test and retest with the exception of calf BF that was significantly higher at retest (P = 0.004). Reproducibility of the forearm BF variables varied with ICCs ranging from 0.169 (post-occlusion BF) to 0.982 (BF), and CVs from 8.2 (BF) to 55.5% (post-occlusion BF). For the calf BF variables, ICCs ranged between 0.653 (AUC) to 0.928 (VR), and CVs between 17.8 (VR) and 37.7% (AUC). In addition, the TE and mean bias were small while the LOA varied each forearm and calf BF variable (Table IV).

4. DISCUSSION

The main findings of this study were that in patients with PAD and IC, systemic hemodynamic, cardiac autonomic modulation and baseline BF assessments exhibited excellent reproducibility as reflected by ICC, CV and mean bias ± LOA. In contrast, reproducibility of the vasodilatory response was moderate for calf and poor for forearm. These assessment tools provide clinicians with reliable indicators for the monitoring of disease progression and cardiovascular risk in PAD patients with IC.

4.1. Systemic hemodynamic variables

Few studies have evaluated the reproducibility of resting BP, HR and RPP in clinical populations. This is surprising given the utility of these measures in guiding disease management and their positive relationship with cardiovascular morbidity and mortality.
Previous studies have reported good to excellent reliability for auscultatory systolic and diastolic BP in patients with borderline hypertension (ICC: 0.61 – 0.83) and untreated adults with elevated BP (ICC: 0.76 – 0.77). Similarly, the current study identified excellent reproducibility (ICC > 0.80 and CV < 9%) for systolic BP, diastolic BP, HR and RPP in patients with IC. These results confirm the utility of these clinical measures to assist with cardiovascular risk screening, and monitoring of disease progression and treatment in patients with PAD and IC.

4.2. Cardiac autonomic modulation

HRV has been used as a non-invasive method to quantify parasympathetic and sympathetic modulations to the heart. HRV alterations, mainly characterized by decreased TVR and HFR, and increased LFR and LF/HF, have been reported in patients with PAD and IC compared to healthy adults with this reduce HRV associated with greater mortality. However, to the best of our knowledge, the current study has been the first to verify the reproducibility of resting HRV in patients with IC. It is well known that reproducibility of HRV is population-specific with lower reproducibility generally obtained in clinical populations compared to healthy adults. However, the present results showed excellent reproducibility (ICC > 0.90 and CV < 9%) for almost all indices, except for LF/HF. Previous studies of patients with a history of myocardial infarction (ICC: 0.77 – 0.81) and type 2 diabetes mellitus (ICC: 0.58 – 0.71) reported poor reliability for the HRV indices. In addition, the current study demonstrated good within-individual variation for TVR, LFR and HFR (CV < 9.0%). Previous studies have reported greater variation for these variables, even in healthy adults (CV: 20.1 – 23.0%) Differences for the within-individual variation of HRV may reflect differences in resting HRV levels with healthy individuals exhibiting greater HRV compared to those with chronic diseases. Regardless, the current study has demonstrated excellent reproducibility (i.e. excellent ICC and desirable CV) for some common HRV measures used for patients with IC. On the other hand, the LF/HF exhibited a high CV (93.8%) that
might be related to the low test and retest mean values with changes producing greater variation in
the measure compared to other HRV variables. Subsequently, LF/HF should be cautiously used for
the monitoring of PAD patients with IC due to its poorer reproducibility.

It is important to highlight that a range of factors may influence reproducibility (reliability
and agreement) of HRV variables. Differences in internal (e.g. physiological state, mood, alertness,
mental activity, etc) and external (e.g. time of day, temperature, etc) factors have been reported to
influence cardiac autonomic modulation \(^{24, 40}\), and may have an impacted on the reproducibility of
test-retest measurements. A rigorous set of procedures had been implemented in the current study
(e.g. control of physical activity, time of day, laboratory temperature and medication) to minimise
any potential influencing factors, which may have resulted in better reproducibility indicators than
that seen previously \(^{19, 38, 39}\). Subsequently, a standardised procedure, such as that adopted in the
current study, may be beneficial and vital to ensure reproducible and accurate monitoring of patients
with IC. Future studies may confirm the advantages of these standardised procedures for other
clinical populations.

4.3. Peripheral hemodynamic variables

Patients with IC have varying degrees of arterial occlusion that consequently result in several
physical limitations due to reduced BF in the limbs \(^{1-3}\). Assessment of BF via strain gauge
plethysmography has been considered the gold standard method for measure distal limb BF \(^{41}\) with a
previous study reporting similar reliability (ICC > 0.90) to that of the current study for baseline
forearm and calf BFAs in healthy adults \(^{42}\). Additionally, within-individual variations for baseline
forearm (8.2%) and calf BF (18.0%) were similar to those previously reported in healthy individuals
\(^{42, 43}\). Others \(^{15, 44}\) though have reported higher CV for these variables with differences between
studies likely a result of different populations and experimental session conditions, such as
positioning of the cuff, strain gauge, assessed limb and physiological circadian rhythm. In addition,
forearm and VR also presented excellent reproducibility (ICC > 0.90 and CV < 20%) as compared with a previous study.\(^1^\)

Considering vasodilatory response, a poor to moderate reproducibility was observed for post-occlusion BF and AUC for the forearm and calf. In contrast, Altenkirch et al \(^4^\) and Thijssen et al \(^5\) reported better reproducibility via CV for forearm post-occlusion BF (10.5% and 8.6%, respectively) and calf post-occlusion BF (7.1% and 6.4%, respectively) in healthy individuals. Differences in reproducibility between studies are most likely due to the presence of vascular disease for PAD patients with post-occlusion vasodilatory responses influenced by several regulating factors (e.g. NO, prostaglandins and others).\(^4^\) Repeated ischemia and reperfusion during ambulation may alter these regulating factors as well as enhance oxidative stress and inflammation\(^4^\),\(^4^\) which contributes to the poorer reproducibility of vasodilatory response in PAD patients with IC.

4.4. Practical applications

The current findings have important applications for both research and clinical settings. In general, excellent reproducibility (ICC > 0.75 and CV < 20%) was identified for systemic hemodynamics, cardiac autonomic modulation and baseline BF and VR in patients with IC that confirms their valuable and practical use to monitor disease and/or intervention progression in clinical settings. However, vasodilatory responses (i.e. post-occlusion BF and AUC) for calf (ICC > 0.40 and CV > 30%) and forearm (ICC < 0.40 and CV > 30%) were moderate-poor that questions the practical relevance of these measures in patients with IC. Finally, the current study determined the specific TE for each variable that should be considered when examining smallest worthwhile changes in PAD patient’s progression and for calculating minimum sample sizes for research studies with IC patients. Moreover, the specific LOA of each variable could be employed as a cut-off/minimal threshold to identify mean changes when evaluating interventions within this specific population.\(^4^\)
4.5. Limitations

This study has some limitations, which should be acknowledged. Firstly, the current study was conducted with men only, which restricts extrapolation of the results to women. Secondly, although patients with PAD usually present other cardiovascular diseases, none of the participants of the study presented heart failure. As reproducibility parameters has been shown to be poor in heart failure\cite{21}, results might be different in patients with this comorbidity. Third, different medications might affect cardiovascular reproducibility. The current study included medication regimes typically used in patients with PAD excepted for β-blockers and non-dihydropyridine calcium channel antagonists because these drugs directly affect the assessment of heart rate variability. Therefore, the results were not applicable to patients receiving these specific drugs. Finally, as seasonal variations may impact on cardiovascular parameters and test and retest had been conducted at the same season of the year, the reproducibility indices obtained in the current study are valid for within season measures but may be different if test and retest were conducted at different seasons. The limitations of the present study suggest that future study should investigate the reproducibility indices in patients with PAD who are women, have heart failure, and are receiving β-blockers or non-dihydropyridine calcium channel antagonists. In addition, future studies should address reproducibility between different seasons of the year.

5. CONCLUSIONS

Systemic hemodynamic (HR and auscultatory BP), cardiac autonomic modulation (TV_{R-R}, LF_{R-R} and HF_{R-R}) and baseline BF assessments exhibited excellent reproducibility at rest for male patients with IC with these tools highly appropriate for clinical monitoring of patients. However, the level of reproducibility for vasodilatory responses (post-occlusion BF and AUC) of the calf and
forearm was moderate to poor that highlighted the limitations of these tools for regular monitoring of disease/intervention in PAD patients.

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The authors declare no conflicts of interest to disclose.

### 6. REFERENCES


47. Signorelli SS, Mazzarino MC, Di Pino L, et al. High circulating levels of cytokines (IL-6 and TNFalpha), adhesion molecules (VCAM-1 and ICAM-1) and selectins in patients with peripheral arterial disease at rest and after a treadmill test. Vasc Med. 2003;8:15-9.


Figure 1. Timeframe of the 4 visits. BMI – Body Mass Index; ABI – Ankle Brachial Index; BP – Blood Pressure; HR – Heart Rate; HRV – Heart Rate Variability; BF – Blood Flow.
Table I. Clinical characteristics of patients

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<table>
<thead>
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</thead>
<tbody>
<tr>
<td>n</td>
<td>29</td>
</tr>
<tr>
<td>Age, y</td>
<td>67 ± 11</td>
</tr>
<tr>
<td>Height, m</td>
<td>1.67 ± 0.07</td>
</tr>
<tr>
<td>Weight, kg</td>
<td>71.0 ± 10.1</td>
</tr>
<tr>
<td>Body mass index, kg/m²</td>
<td>25.3 ± 3.3</td>
</tr>
<tr>
<td>Ankle brachial index</td>
<td>0.62 ± 0.13</td>
</tr>
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</table>

**Cardiovascular measures**

<p>| | |</p>
<table>
<thead>
<tr>
<th></th>
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</thead>
<tbody>
<tr>
<td>Systolic blood pressure (mmHg)</td>
<td>136 ± 15</td>
</tr>
<tr>
<td>Diastolic blood pressure (mmHg)</td>
<td>82 ± 9</td>
</tr>
<tr>
<td>Heart rate (bpm)</td>
<td>72 ± 11</td>
</tr>
</tbody>
</table>

**Cardiovascular risk factors and comorbidities**

<p>| | |</p>
<table>
<thead>
<tr>
<th></th>
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</thead>
<tbody>
<tr>
<td>Obesity, %</td>
<td>6.9</td>
</tr>
<tr>
<td>Hypertension, %</td>
<td>69.0</td>
</tr>
<tr>
<td>Diabetes mellitus, %</td>
<td>27.6</td>
</tr>
<tr>
<td>Dyslipidaemia, %</td>
<td>93.1</td>
</tr>
<tr>
<td>Current smoker, %</td>
<td>34.5</td>
</tr>
<tr>
<td>Heart disease, %</td>
<td>17.2</td>
</tr>
</tbody>
</table>

**Medications**

<p>| | |</p>
<table>
<thead>
<tr>
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<tbody>
<tr>
<td>Aspirin, %</td>
<td>93.1</td>
</tr>
<tr>
<td>Statin, %</td>
<td>93.1</td>
</tr>
<tr>
<td>Angiotensin-converting enzyme inhibitor, %</td>
<td>44.8</td>
</tr>
<tr>
<td>Diuretic, %</td>
<td>24.1</td>
</tr>
<tr>
<td>Dihydropyridine calcium channel antagonist, %</td>
<td>20.7</td>
</tr>
</tbody>
</table>
Continuous data are expressed as mean ± SD. Categorical data are expressed as percentage of occurrence (%). Obesity was defined as body mass index ≥ 30 kg/m. Hypertension, diabetes mellitus, dyslipidaemia and heart disease were defined by previous physician diagnosis.
Table II. Mean ± SD values and reproducibility parameters of systemic hemodynamic variables

<table>
<thead>
<tr>
<th>Variable</th>
<th>Test</th>
<th>Retest</th>
<th>P value</th>
<th>ICC</th>
<th>TE</th>
<th>CV %</th>
<th>Bias ± 95% LOA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Systolic BP (mmHg)</td>
<td>134 ± 13</td>
<td>132 ± 16</td>
<td>0.409</td>
<td>0.863</td>
<td>7.2</td>
<td>5.4</td>
<td>1.6 ± 20.0</td>
</tr>
<tr>
<td>Diastolic BP (mmHg)</td>
<td>78 ± 8</td>
<td>77 ± 9</td>
<td>0.362</td>
<td>0.833</td>
<td>4.8</td>
<td>6.2</td>
<td>1.1 ± 13.2</td>
</tr>
<tr>
<td>Mean BP (mmHg)</td>
<td>96 ± 9</td>
<td>95 ± 10</td>
<td>0.306</td>
<td>0.845</td>
<td>4.8</td>
<td>5.0</td>
<td>1.3 ± 13.2</td>
</tr>
<tr>
<td>HR (bpm)</td>
<td>66.6 ± 9.1</td>
<td>64.6 ± 8.8</td>
<td>0.062</td>
<td>0.908</td>
<td>3.7</td>
<td>5.6</td>
<td>2.0 ± 10.2</td>
</tr>
<tr>
<td>RPP (mmHg.bpm)</td>
<td>8879 ± 1347</td>
<td>8494 ± 1520</td>
<td>0.060</td>
<td>0.856</td>
<td>720</td>
<td>8.3</td>
<td>385 ± 1994</td>
</tr>
</tbody>
</table>

BP – blood pressure; HR – heart rate; RPP – rate pressure product; ICC – intraclass correlation coefficient; TE – typical error; CV – coefficient of variation; LOA – limits of agreement
Table III. Mean ± SD values and reproducibility parameters of autonomic variables

<table>
<thead>
<tr>
<th></th>
<th>Test</th>
<th>Retest</th>
<th>P value</th>
<th>ICC</th>
<th>TE</th>
<th>CV %</th>
<th>Bias ± or */÷ 95% LOA</th>
</tr>
</thead>
<tbody>
<tr>
<td>ln TV&lt;sub&gt;R-R&lt;/sub&gt; (ms&lt;sup&gt;2&lt;/sup&gt;)</td>
<td>6.8 ± 1.0</td>
<td>6.9 ± 0.9</td>
<td>0.459</td>
<td>0.929</td>
<td>0.3</td>
<td>4.9</td>
<td>0.9 */÷ 2.6</td>
</tr>
<tr>
<td>LF&lt;sub&gt;R-R&lt;/sub&gt; (nu)</td>
<td>47.8 ± 19.9</td>
<td>47.5 ± 18.8</td>
<td>0.831</td>
<td>0.979</td>
<td>4.0</td>
<td>8.3</td>
<td>0.3 ± 11.0</td>
</tr>
<tr>
<td>HF&lt;sub&gt;R-R&lt;/sub&gt; (nu)</td>
<td>44.7 ± 20.7</td>
<td>45.5 ± 19.4</td>
<td>0.535</td>
<td>0.986</td>
<td>3.3</td>
<td>7.3</td>
<td>-0.7 ± 9.1</td>
</tr>
<tr>
<td>ln LF/HF</td>
<td>0.18 ± 0.87</td>
<td>0.17 ± 0.88</td>
<td>0.737</td>
<td>0.982</td>
<td>0.16</td>
<td>93.8</td>
<td>1.0 */÷ 1.6</td>
</tr>
</tbody>
</table>

ln – natural logarithm; TV – total variance; LF – low frequency component; HF – high frequency component; nu – normalized units; ICC – intraclass correlation coefficient; TE – typical error; CV – coefficient of variation; LOA – limits of agreement
Table IV. Mean ± SD values and reproducibility forearm and calf blood flows and vasodilatory response variables

<table>
<thead>
<tr>
<th></th>
<th>Test</th>
<th>Retest</th>
<th>P value</th>
<th>ICC</th>
<th>TE</th>
<th>CV %</th>
<th>Bias ± 95% LOA</th>
</tr>
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<tbody>
<tr>
<td><strong>Forearm</strong></td>
<td></td>
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</tr>
<tr>
<td>BF (ml.100 ml(^{-1}).min(^{-1}))</td>
<td>1.42 ± 0.63</td>
<td>1.41 ± 0.59</td>
<td>0.872</td>
<td>0.982</td>
<td>0.12</td>
<td>8.2</td>
<td>0.00 ± 0.32</td>
</tr>
<tr>
<td>VR (U)</td>
<td>75 ± 35</td>
<td>74 ± 32</td>
<td>0.627</td>
<td>0.978</td>
<td>7.0</td>
<td>9.3</td>
<td>1 ± 19</td>
</tr>
<tr>
<td>Post-occlusion BF (ml.100 ml(^{-1}).min(^{-1}))</td>
<td>11.4 ± 7.0</td>
<td>11.2 ± 6.1</td>
<td>0.938</td>
<td>0.169</td>
<td>6.3</td>
<td>55.5</td>
<td>0.15 ± 17.37</td>
</tr>
<tr>
<td>Post-occlusion AUC BF (U)</td>
<td>1294 ± 676</td>
<td>1085 ± 507</td>
<td>0.232</td>
<td>0.265</td>
<td>550</td>
<td>46.2</td>
<td>209 ± 1524</td>
</tr>
<tr>
<td><strong>Calf</strong></td>
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<td></td>
</tr>
<tr>
<td>BF (ml.100 ml(^{-1}).min(^{-1}))</td>
<td>1.69 ± 0.72</td>
<td>2.06 ± 0.95</td>
<td>0.004</td>
<td>0.913</td>
<td>0.34</td>
<td>18.0</td>
<td>-0.37 ± 0.94</td>
</tr>
<tr>
<td>VR (U)</td>
<td>60 ± 25</td>
<td>57 ± 31</td>
<td>0.362</td>
<td>0.928</td>
<td>10.0</td>
<td>17.8</td>
<td>3 ± 29</td>
</tr>
<tr>
<td>Post-occlusion BF (ml.100 ml(^{-1}).min(^{-1}))</td>
<td>5.73 ± 3.18</td>
<td>5.05 ± 3.04</td>
<td>0.297</td>
<td>0.770</td>
<td>1.9</td>
<td>35.2</td>
<td>0.68 ± 5.27</td>
</tr>
<tr>
<td>Post-occlusion AUC BF (U)</td>
<td>999 ± 467</td>
<td>1081 ± 616</td>
<td>0.553</td>
<td>0.653</td>
<td>392</td>
<td>37.7</td>
<td>-82 ± 1088</td>
</tr>
</tbody>
</table>

BF – blood flow; VR – vascular resistance; AUC – area under the curve; ICC – intraclass correlation coefficient; TE – typical error; CV – coefficient of variation; LOA – limits of agreement.