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# Variation in Heart Rate Variability between Sitting and Standing Postures under Different Ambient Temperatures

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**Abstract Purpose:** The purpose of this study was to investigate the effects of two different postures (sitting and standing) and three different ambient temperatures  $(10^{\circ}, 20^{\circ})$ , and  $30^{\circ}$ ) on heart rate variability (HRV) among healthy young adults. Methods: Twelve young adult volunteers (males 6, females 6) were recruited. Following acclimatization to any the room temperature  $(10^{\circ}\text{C}, 20^{\circ}\text{C} \text{ or } 30^{\circ}\text{C})$ , 5-min measurements of HRV were conducted in sitting and standing postures of the subjects. **Results:** Compared to the sitting posture, measurements obtained in the standing posture revealed a significant decrease in high-frequency power/HF, root mean square of successive differences between RR intervals, standard deviation of Poincaré plot perpendicular to the line-of-identity or SD1 and SD1/standard deviation of Poincaré plot along the line-of-identity or SD2, and a significant increase in lowfrequency power/LF and LF/HF under all experimental conditions (p<0.05 to 0.005). Majority of HRV parameters showed significant differences while the values obtained under 10°C were compared with 20°C and 30°C conditions, respectively (p<0.05 to 0.001). **Conclusions:** Our findings suggest the predominance of sympathetic tone in the standing compared with sitting posture. Furthermore, colder conditions caused a predominance of the parasympathetic activity in both sitting and standing postures, and such effects of ambient temperature on the sympathovagal balance were stronger in the latter posture.

Key words: heart rate variability, posture, ambient temperature

# Introduction

Non-invasive evaluation of heart rate variability (HRV) has become a subject of clinical and research interest as a key indicator of the activity of the autonomic nervous system (ANS). High or improved levels of HRV indicate good ANS homeostasis characterizing a healthy individual; in contrast, low or reduced levels of HRV often indicate an imbalance in ANS activity.<sup>1</sup> Such a decrease in HRV can be caused by an increase in sympathetic activity and/or a reduction in parasympathetic activity,<sup>2</sup> and can be a predictor of cardiovascular, metabolic, and other adverse health outcomes.<sup>3-5</sup> Therefore, the noninvasive measurement of HRV has become a topic of growing interest in biomedical, clinical and epidemiological research.

For proper evaluation and/or identification of any health impairments reflected by the changes in HRV, accurate and standardized measurements of the latter is important. However, in the published literature, the relevant study protocols for the measurements of HRV vary widely among the investigators. Moreover, it has been demonstrated that various physiological and environmental factors can affect the measurements of HRV.<sup>2</sup> Therefore, these issues need to be considered in designing precise research protocols, and for proper measurements and interpretations of the relevant data.

As observed in the published studies, the measurements of HRV have been performed at different body postures of the study subjects (supine, sitting, and standing) under different ambient temperatures (ranging between  $10^{\circ}$ C and  $30^{\circ}$ C).<sup>6-9</sup> However, the measurements of HRV can be strongly influenced by such factors like body posture and environmental conditions.<sup>10-12</sup> In the literature, a good number of studies exists that compared the HRV values obtained at supine and standing postures.  $^{\scriptscriptstyle 13\text{-}15}$  On the other hand, only a few studies compared those values obtained for sitting and standing postures. The situation has been complicated by the fact that the relevant findings of those studies conducted among healthy subjects are inconsistent. For instances, a number of studies suggested a decrease in HRV in the standing posture compared to the sitting posture,<sup>2,16</sup> whereas others could not find any such differences in HRV.<sup>17</sup> Due to such inconsistent findings, the effects of body posture on HRV remain inconclusive. Furthermore, there is a severe lack of studies assessing the effects of ambient temperature on HRV for different body postures, especially between the sitting and standing postures of study subjects.

Improved knowledge on the influence of body posture with simultaneous exposure to different ambient temperatures, on the patterns of changes in HRV, should help to establish a suitable and standard protocol for the measurements of HRV. It would also assist to better understand the possibility of inducing positive changes in the latter.

Considering the above-mentioned issues, in the present study, we attempted to explore and clarify the effects of two different postures (sitting and standing) and three different ambient temperatures ( $10^{\circ}$ C,  $20^{\circ}$ C, and  $30^{\circ}$ C) on HRV among healthy young adults.

### Materials and Methods

#### Selection of subjects

The current study is part of a larger research project the purpose of which was to examine the acute effects of intervention with moderate exercise protocol under three different ambient temperatures  $(10^{\circ}\text{C}, 20^{\circ}\text{C})$ and  $30^{\circ}$  on various physiological parameters including HRV. For this study, young healthy adult subjects were recruited from Yamaguchi University School of Medicine via poster advertisements and word of mouth. The selection criteria for the study subjects were as follows: no reported neurological, musculoskeletal, cardiovascular or connective tissue disorders or any other known diseases that would prohibit exposure to exercise, no history of surgery within a year, not habitual cigarette smokers etc. A total of 12 young adult volunteers (males 6, females 6) were invited to participate in the study, and all of them were able to complete all the experimental sessions. The median with 25th and 75th percentiles for age and BMI of the study subjects were as follows: males, 21.0 (20.8 to 22.3) years and 19.3 (17.9 to 20.2) kg/m<sup>2</sup>, respectively; females, 21.0 (20.8 to 22.0) years and 18.9

# $(17.7 \text{ to } 21.7) \text{ kg/m}^2$ , respectively.

The subjects were instructed to refrain from eating and drinking tea or coffee for at least 3 h and smoking, alcohol drinking and strenuous physical activity for at least 12 h prior to the beginning of the test. They entered the experiment room after voiding. The subjects were instructed to wear light indoor clothing (two each for the upper and lower parts of the body) and were barefoot in the laboratory.

#### Experimental design

To account for the effects of circadian rhythms on the physiological responses,<sup>18</sup> we carried out the three experimental sessions randomly on three different days separated by at least 24 h, approximately at the same time for each subject between 9:00 am and 5:00 pm, during the fall season (October 2021 - November 2021).

Upon arrival, the subjects underwent acclimatization for a period of 15 min in the experiment room with a room temperature maintained at one of the three conditions,  $10^{\circ}$ C,  $20^{\circ}$  or  $30^{\circ}$  ( $\pm 1^{\circ}$ ), while seated comfortably on a height-adjustable chair without physiological or psychological stress. While seated, they positioned their feet on the wooden floor, and both hands on respective thighs. At the end of the acclimatization period, initial measurements of HRV were conducted for 5 min. This was followed by baseline measurements of vibration perception threshold (not reported in this study), after which the participants were asked to stand on a treadmill platform in complete upright posture and to look forward. During this time, all subjects positioned their both arms hanging down in a relaxed manner and in close contact to the upper body. In this standing posture, after a rest period of 1 min, the measurements of HRV were conducted for 5 min.

## Equipment and measurements

HRV data were recorded using a portable battery-operated 2-channel heart rate device (CheckMyHeart, DailyCare BioMedical, Inc., Taiwan). The device was connected via electrode cables with disposable Ag/AgCl circular surface electrodes (Bioload SDC-H, GE Healthcare, Japan), and the latter were placed on ventral forearms.

## Data processing and statistical analyses

The sampling frequency of HRV data was 250 Hz.<sup>19</sup> The data were visually inspected for noise or ectopic beats using the HRV analysis software (DailyCare BioMedical, Inc., Taiwan).

In the current study, we analyzed the following HRV indices which have been commonly used in the previous studies while investigating the effects of physical and environmental factors on HRV:11,20-25 1) lowfrequency power (LF, 0.04 to 0.15 Hz); 2) highfrequency power (HF, 0.15 to 0.40 Hz); 3) the ratio of LF-to-HF power (LF/HF); 4) standard deviation of normal-to-normal intervals (SDNN); 5) root mean square of successive differences between RR intervals (RMSSD); 6) standard deviation of Poincaré plot perpendicular to the line-of-identity (SD1); 7) standard deviation of Poincaré plot along the line-of-identity (SD2); 8) the ratio of SD1-to-SD2 (SD1/SD2). As shown in Figure 1, the Poincaré plot is a scatter plot in which all RR intervals are plotted against each previous interval, and SD1 and SD2 are calculated by fitting ellipses to this plot.<sup>23</sup> Among the

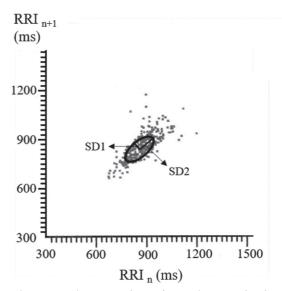


Fig. 1 Poincaré plot of RR intervals derived from HRV data during standing posture of a healthy subject under 20°C exposure condition.

RRI, RR interval; SD1, standard deviation of Poincaré plot perpendicular to the line-of-identity; SD2, standard deviation of Poincaré plot along the line-of-identity. indices, LF, HF and LF/HF are classified as frequency-domain, SDNN and RMSSD as time-domain, and SD1, SD2 and SD1/SD2 as non-linear indices of HRV.

The frequency-domain indices of HRV were calculated for the detrended values of regular RR interval data, using the fast - Fourier - transformation. Furthermore, the values of LF and HF were normalized (expressed in normalized unit or nu) as follows: (1) normalized LF = LF  $\div$  (total power - VLF); and (2) normalized HF = HF  $\div$  (total power - VLF), where VLF indicates very low frequency power (between 0.0033 to 0.04 Hz) of HRV.<sup>23</sup>

The continuous variables of this study were expressed as median with 25th and 75th percentiles in the text and figures. The differences between two-related samples were assessed by Wilcoxon signed-rank tests. Friedman test was used for examination of the differences between k-related samples, with subsequent application of Wilcoxon signed-rank tests for the variables showing significant differences by the former test. Bonferroni corrections were applied for multiple comparisons as necessary.

All statistical tests were considered as twotailed, and the significance level was set at P<0.05. The software package SPSS version 22 for Windows (SPSS Inc., Chicago, IL, USA) was used to perform the statistical analyses.

## Ethical statement

All subjects provided written informed consent before they participated in this study. The study was conducted in accordance with the Declaration of Helsinki, and the protocol was approved by the relevant institutional review board of Yamaguchi University School of Medicine (approval no. H2021-104, dated 22-09-2021).

#### Results

#### Influence of posture

Figure 2 displays the values of LF (ms<sup>2</sup> and nu), HF (ms<sup>2</sup> and nu) and LF/HF obtained in sitting and standing postures under each of the three ambient temperature conditions. Data analysis revealed that all the parameters significantly differed between the two postures, except LF (ms<sup>2</sup>) for 10°C and 20°C.

Compared to the sitting posture, measurements obtained in the standing posture revealed a significant increase in LF (nu) and LF/HF (p<0.05 to 0.01) and a significant decrease in HF (ms<sup>2</sup> and nu) (p<0.01 to 0.005) under all ambient temperature conditions. For LF (ms<sup>2</sup>), a significant decrease was observed in the standing posture, compared with the sitting posture, only under the 30°C condition (p<0.05).

Figure 3 represents the values of SDNN and RMSSD obtained in sitting and standing postures under each of the three ambient temperature conditions. The values for both parameters were significantly different between the two postures with the exception of SDNN obtained under 20°C condition. Compared to the sitting posture, measurements in the standing posture showed a significant decrease in SDNN under 10°C (p<0.05) and 30°C (p<0.005) conditions, and for RMSSD, under all ambient temperature conditions (p<0.005).

Figure 4 shows the values of SD1, SD2 and SD1/SD2 obtained in sitting and standing postures under three different ambient temperature conditions. The values for all parameters were significantly different between the two postures with the exception for SD2 under 10°C and 20°C conditions. Compared to the sitting posture, measurements in the standing posture demonstrated a significant decrease in SD1 and SD1/SD2 under all experimental conditions (p<0.05 to 0.005), and in SD2, under only 30°C condition (p<0.005).

#### Influence of ambient temperature

Comparisons of the values of HRV between three ambient temperature conditions for each body posture revealed that there were significant differences in all measured parameters with the exception for LF  $(ms^2)$ (p<0.05 to 0.001; Table 1). The values of HRV parameters showed a decreasing trend with an increase in the ambient temperature, except for LF (nu) and LF/HF which showed the opposite trend in their corresponding values (Figures 2-4). Next, those significant variables were compared between the ambient temperature conditions by multiple comparisons. As the results revealed, all HRV parameters showed significant differences while the corresponding values were compared between

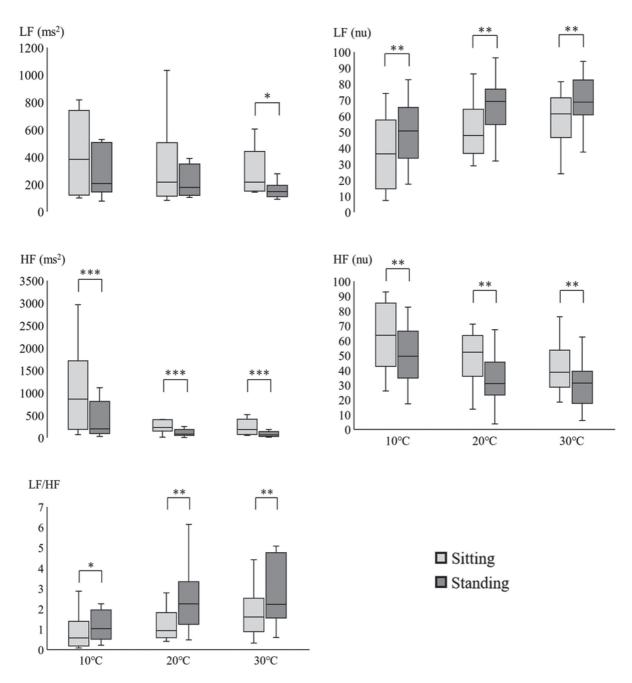


Fig. 2 Boxplots displaying median with 25th and 75th percentiles for measured HRV parameters (frequency-domain) in two different postures (sitting and standing) under three different ambient temperature conditions (10°C, 20°C and 30°C). Levels of significant differences between sitting and standing postures: \*p<0.05, \*\*p<0.01, \*\*\*p<0.005.

LF (ms<sup>2</sup>), absolute power of the low-frequency band; LF (nu), relative power of the low-frequency band in normal units; HF (ms<sup>2</sup>), absolute power of the high-frequency band; HF (nu), relative power of the high-frequency band in normal units; LF/HF, ratio of LF-to-HF power.

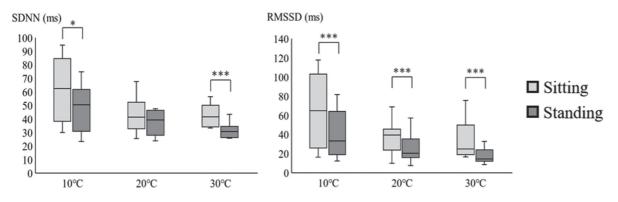


Fig. 3 Boxplots displaying median with 25th and 75th percentiles for measured HRV parameters (time-domain) in two different postures (sitting and standing) under three different ambient temperature conditions ( $10^{\circ}$ C,  $20^{\circ}$ C and  $30^{\circ}$ C). Levels of significant differences between sitting and standing postures: \*p<0.05, \*\*\*p<0.005.

SDNN, standard deviation of normal-to-normal intervals; RMSSD, root mean square of successive differences between RR intervals.

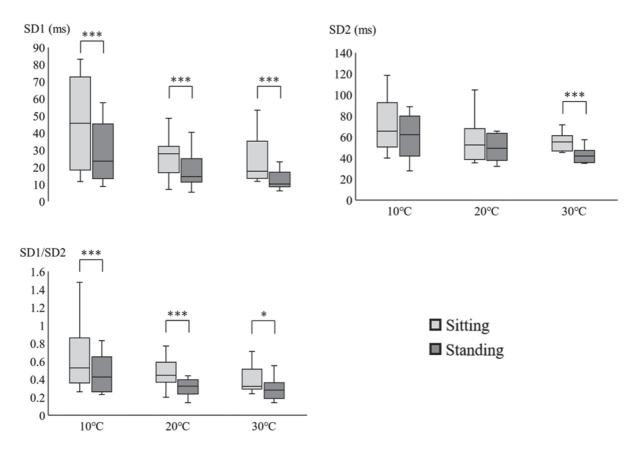


Fig. 4 Boxplots displaying median with 25th and 75th percentiles for measured HRV parameters (non-linear measurements) in two different postures (sitting and standing) under three different ambient temperature conditions ( $10^{\circ}$ C,  $20^{\circ}$ C and  $30^{\circ}$ C). Levels of significant differences between sitting and standing postures: \*p<0.05, \*\*\*p<0.005.

SD1, standard deviation of Poincaré plot perpendicular to the line-of-identity; SD2, standard deviation of Poincaré plot along the line-of-identity; SD1/SD2, ratio of SD1 to SD2.

HRV	Sitting/Standing	p-value			
Component	Posture	Overall	10°C vs 20°C	20°C vs 30°C	10℃ vs 30℃
LF (ms <sup>2</sup> )	Sitting	0.127	—	—	_
	Standing	0.076	_	—	_
LF (nu)	Sitting	0.013	0.045	0.627	0.036
	Standing	0.002	0.018	0.627	0.009
HF (ms <sup>2</sup> )	Sitting	0.002	0.012	0.924	0.009
	Standing	< 0.001	0.006	0.123	0.006
HF (nu)	Sitting	0.013	0.045	0.627	0.036
	Standing	0.002	0.018	0.627	0.009
LF/HF	Sitting	0.013	0.102	1.000	0.057
	Standing	0.002	0.018	1.000	0.015
SDNN (ms)	Sitting	0.018	0.015	1.000	0.030
	Standing	0.028	0.069	0.213	0.024
RMSSD (ms)	Sitting	0.001	0.015	0.351	0.015
	Standing	< 0.001	0.015	0.018	0.006
SD1 (ms)	Sitting	0.001	0.015	0.351	0.015
	Standing	< 0.001	0.015	0.018	0.006
SD2 (ms)	Sitting	0.046	0.018	1.000	0.102
	Standing	0.028	0.084	0.252	0.024
SD1/SD2	Sitting	0.002	0.033	0.273	0.009
	Standing	0.005	0.030	0.030	0.021

Table 1 Comparison of heart rate variability between three ambient temperature conditions in sitting and standing postures

P values indicate the levels of significant differences for the corresponding values under three ambient temperature conditions by Friedman test (overall) and subsequent analysis by Wilcoxon signed ranks test for paired comparisons with adjustments for multiple comparisons by Bonferroni corrections.

LF (ms<sup>2</sup>), absolute power of the low-frequency band; LF (nu), relative power of the low-frequency band in normal units; HF (ms<sup>2</sup>), absolute power of the high-frequency band; HF (nu), relative power of the high-frequency band in normal units; LF/HF, ratio of LF-to-HF power; SDNN, standard deviation of normal-to-normal intervals; RMSSD, root mean square of successive differences between RR intervals; SD1, standard deviation of Poincaré plot perpendicular to the line-of-identity; SD2, standard deviation of Poincaré plot along the line-ofidentity; SD1/SD2, ratio of SD1 to SD2.

 $10^{\circ}$  and  $20^{\circ}$  conditions except LF/HF (sitting), SDNN (standing) and SD2 (standing), and between  $10^{\circ}$  C and  $30^{\circ}$  C with the exception for LF/HF and SD2 in the sitting posture. On the other hand, there were significant differences between  $20^{\circ}$  and  $30^{\circ}$  conditions only for three HRV parameters in the standing posture: RMSSD, SD1, and SD1/SD2.

## Discussion

effects of two different postures (sitting and standing) and three different ambient temperature conditions ( $10^{\circ}$ C,  $20^{\circ}$ C, and  $30^{\circ}$ C) on HRV in young healthy subjects. We conducted short 5-min measurements of HRV, which is recognized as an appropriate standard option for relevant data analysis.<sup>23</sup>

#### Influence of posture

The frequency-domain measure allows quantification of the cyclic variation in the In the present study, we characterized the RR intervals.<sup>26</sup> Among the parameters related to this measure used in the present study, LF is an index reflecting both sympathetic and parasympathetic nervous tone, HF represents purely parasympathetic tone, and LF/HF is an index showing sympathovagal balance.<sup>27</sup> As recommended, LF and HF need to be presented in both absolute (ms<sup>2</sup>) and normalized (nu) values which would provide a complete view of the power distribution, emphasizing the balanced changes in the ANS and minimizing the effects of changes in total power on the values of LF and HF.<sup>27,28</sup> In fact, both LF  $(ms^2)$  and HF  $(ms^2)$  are known to be affected by reduction in total power, especially the former one, which may not be properly reflected in situations with sympathetic excitation.<sup>28</sup> Therefore, in this study, we also used LF (nu) and HF (nu), which are widely used as measures of sympathetic nervous system (SNS) and parasympathetic nervous system (PNS), respectively.<sup>29</sup> It should be noted here that corrected values for LF and HF were used in this study when calculating LF/HF.<sup>30</sup> Considering these and the fact that compared to the sitting posture, the standing posture resulted in a significant decrease in  $HF (ms^2 and nu)$  and a significant increase in LF (nu) and LF/HF under all ambient temperature conditions, we postulate that the PNS tone was remarkably reduced and the SNS tone was significantly elevated with a relative predominance of the sympathetic activity. Our findings are comparable to those of Bloomfield et al. (1997) and Young et al. (2011), who also measured the HRV and reported similar changes in the autonomic balance in the standing posture compared to the head-up tilt or sitting posture of study subjects.<sup>16,31</sup> As we observed an opposite trend in the values of LF  $(ms^2)$  and LF (nu) for both postures, this might have been caused by a decrease in total power, as mentioned above, and corresponds with the findings reported previously in the literature.<sup>16</sup>

The time-domain measure quantifies the variability of the normal-to-normal (normal RR) interval time series, which is considered to be broadly sensitive to changes in various determinants of HRV.<sup>27,32</sup> Among the relevant parameters that we measured in this study, SDNN is a measure of overall variability, while RMSSD varies mainly due

to the effects of the PNS.<sup>28</sup> A significant decrease in RMSSD for standing posture under all temperature conditions indicates that the parasympathetic tone was reduced, compared to the sitting posture. On the other hand, we observed a significant decrease in SDNN at standing posture for 10°C and 30°C test conditions. Similarly, Nepal and Paudel (2012) also examined the changes in HRV among school children and reported significant decreases in RMSSD and SDNN in the standing posture in comparison with the corresponding values in the supine and/or sitting postures. They explained such a decrease in PNS as a reduction in HRV in the standing posture.<sup>33</sup> All these findings indicate that the standing posture caused a notable decrease in HRV under  $10^{\circ}$ C and  $30^{\circ}$  test conditions whereas exposure to 20°C test condition was probably less stressful to the study participants.

Non-linear measurement quantifies the complexity of cardiovascular dynamics based on the unpredictability of the time series of factors associated with HRV.<sup>23,34</sup> In our study, we applied an approach based on the Poincaré plot, which is the most popular method used for the purpose and provides a visually comprehensible summary of cardiac behavior.<sup>35</sup> Among the measured non-linear parameters of HRV, SD1 and SD2 are comparable to RMSSD and LF, reflecting vagal and overall ANS modulation, respectively.<sup>36,37</sup> Significant reductions in SD1 and SD1/SD2 in standing posture correspond to our relevant findings of frequency- and time-domain measures mentioned above, and also indicate a remarkable reduction in PNS tone. On the other hand, a significant decrease in SD2 at 30°C test condition during standing, probably indicate a stronger decrease in HRV under this higher temperature.

#### Influence of ambient temperature

Among the frequency-domain parameters of HRV, we observed significant differences for HF (ms<sup>2</sup> and nu), LF (nu), and LF/HF (in standing posture) when the values under  $10^{\circ}$ exposure condition were compared with the those obtained under  $20^{\circ}$  and  $30^{\circ}$  exposure conditions, respectively. Among these, a significant increase in HF (ms<sup>2</sup> and nu) and a significant decrease in the other two parameters suggest that compared to heat stress, cold stress has a greater effect on HRV which tends to shift ANS toward parasympathetic dominance. Our observation is consistent with those of previous studies which suggested that acute whole-body cold stimulation leads to the vagal activation.<sup>38,39</sup> As we did not observe any significant difference in LF/HF in the sitting posture between the exposure temperature conditions, it might suggest that compared to the sitting posture, the effects of ambient temperature on sympathovagal balance are stronger in the standing posture.

Overall, the time-domain parameters of HRV showed significant or remarkable differences when the values under  $10^{\circ}$ C exposure condition were compared with those obtained under  $20^{\circ}$ C and  $30^{\circ}$ C exposure conditions. There was an increasing trend in the values of SDNN and RMSSD with a decreasing ambient temperature, in both sitting and standing postures. It is difficult to compare our findings with the existing literature due to the lack of studies with concomitant investigation of postural effects and ambient temperature on HRV. However, our abovementioned findings correspond to the observations discussed above for frequency-domain parameters, and indicate the promotion of the activity of the PNS by exposure to cold stimulation.<sup>38-40</sup> For RMSSD, a significant difference in the standing posture between  $20^{\circ}$  and  $30^{\circ}$  exposure conditions might have been caused by a stronger effect of the ambient temperature in this body position, compared to the sitting posture.

Among the non-linear parameters of HRV, the significant changes observed for SD2 indicate that cold stimulation may activate the PNS as well as the SNS.<sup>39</sup> However, the changes in SD1 and SD1/SD2 observed in both sitting and standing postures under different temperature conditions implies that although cold exposure can cause activation of both SNS and PNS, it induces a dominance of the parasympathetic tone over the sympathetic tone. In addition, an increase in SD1/SD2 under the colder temperatures suggests an abnormal distribution of Poincaré plots indicating an increase in heart rate unpredictability under cold environments.<sup>41</sup> Moreover, as SD1/ SD2 showed a significant difference between  $20^{\circ}$ C and  $30^{\circ}$ C in the standing posture and not in the sitting posture, it suggests that this metric of HRV in the standing posture is probably more affected by changes in the ambient temperature conditions. It also corresponds well with our relevant observations for the frequency- and time-domain parameters of HRV.

## Limitations

This study has several potential limitations and caution should be exercised in interpreting the current results. The generalization of the findings is somewhat limited as this study was conducted amongst a group of young people. Another limitation might be the fact that sample size of this study was relatively small. Also, due to this limited number of subjects, we did not attempt to investigate the sex differences in the observed findings of our study. However, we believe that such a difference in the sex of participants would not alter the trends in our findings as female HRV is also reported to be characterized by a relative dominance of the vagal activity.<sup>42</sup> In this study, we conducted short-term (5-min) measurements of HRV. The current findings need to be confirmed in future studies with long-term measurements of HRV including subjects with different age groups.

#### Conclusions

The findings of our study with frequencydomain, time-domain, and non-linear analysis of short-term HRV in young healthy subjects indicate the predominance of SNS in the standing compared with sitting posture. Furthermore, colder conditions caused a predominance of the parasympathetic activity in both sitting and standing postures, and such effects of ambient temperature on the sympathovagal balance were stronger in the latter posture. Therefore, body posture of participants and ambient temperature need to be carefully considered when designing research studies with measurements of HRV.

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# Conflict of Interest

The authors declare no conflict of interest.

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