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The impact of breathing on HRV measurements: Implications for the longitudinal follow-up of athletes

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Abstract
The purpose of the present work was to compare daily variations of heart rate variability (HRV) parameters between controlled breathing (CB) and spontaneous breathing (SB) sessions during a longitudinal follow-up of athletes. HRV measurements were performed daily on 10 healthy male runners for 21 consecutive days. The signals were recorded during two successive randomised 5-minutes sessions. One session was performed in CB and the other in SB. The results showed significant differences between the two respiration methods in the temporal, nonlinear and frequency domains. However, significant correlations were observed between CB and SB (higher than 0.70 for RMSSD and SD1), demonstrating that during a longitudinal follow-up, these markers provide the same HRV variations regardless of breathing pattern. By contrast, independent day-to-day variations were observed with HF and LF/HF frequency markers, indicating no significant relationship between SB and CB data over time. Therefore, we consider that SB and CB may be used for HRV longitudinal follow-ups only for temporal and nonlinear markers. Indeed, the same daily increases and decreases were observed whatever the breathing method employed. Conversely, frequency markers did not provide the same variations between SB and CB and we propose that these indicators are not reliable enough to be used for day-to-day HRV monitoring.

Keywords: *Endurance training, monitoring, heart rate variability, breathing, spontaneous controlled*

Introduction

For the last three decades, heart rate variability (HRV) has been widely used for quantifying the activity of the autonomic nervous system (Akselrod et al., 1981; Task-Force, 1996). Several authors have demonstrated the utility of HRV for the follow up of diseases (La Rovere et al., 2003; Tsuji et al., 1996). In addition, because the autonomic nervous system plays an important role in exercise training responses, HRV can be used to prevent fatigue or overtraining syndromes (Baumert et al., 2006; Hedelin, Wiklund, Bjerle, & Henriksson-Larsen, 2000). Thus, HRV is now commonly used to guide and individualise training load (Kiviniemi et al., 2010; Kiviniemi, Hautala, Kinnunen, & Tulppo, 2007) and monitor fatigue during longitudinal follow-ups (Plews, Laursen, Kilding, & Buchheit, 2012).

Many studies have shown that HRV is modulated by respiratory activity (Eckberg, 2003; Penttila et al., 2001). This cardio-respiratory interaction is called respiratory sinus arrhythmia (RSA) (Hirsch & Bishop, 1981; Larsen, Tzeng, Sin, & Galletly, 2010). It is currently widely admitted that the respiration cycle affects HRV results and some studies have demonstrated that RSA causes a spectral power peak at breathing frequency (BF; Schipke, Pelzer, & Arnold, 1999). With slow and deep breathing (Strano et al., 1998), the RSA of trained athletes is stronger than that of sedentary subjects (De Meersman, 1992). Thus respiration may greatly disturb HRV results, especially for athletes whose BF is close to 0.15 Hz [i.e. the threshold between low frequency (LF) and high frequency (HF); Middleton & De Vito, 2005; Saboul, Pialoux, & Hautier, in press].

To rule out this influence of respiration, many authors use metronomic controlled breathing (CB) to perform HRV measurements on populations of athletes (Gregoire, Tuck, Yamamoto, & Hughson,

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1996; Melanson & Freedson, 2001). In addition, using CB improves the reproducibility and reliability of test findings and facilitates quantitative inter-individual comparisons, thereby avoiding misclassification between LF and HF power (Ori, Monir, Weiss, Sayhouni, & Singer, 1992; Rajendra Acharya, Paul Joseph, Kannathal, Lim, & Suri, 2006). Conversely, the LF/HF ratio is significantly modified by CB (Saboul et al., in press), and some authors have postulated that sympathovagal balance can only be represented by spontaneous breathing (SB) (Pagani et al., 1986). Consequently, SB and CB continue to be used indifferently in sports studies making it impossible to compare the results of different publications.

To address this problem, several studies have compared and modelled the influence of CB and SB on instantaneous HRV analyses (Ben Lamine et al., 2004; Bernardi et al., 2000). The effects of ventilation on HRV markers are controversial and generally limited to sedentary and patient populations (Bloomfield et al., 2001). More globally, it is impossible to determine whether CB or SB is the most suitable breathing pattern to monitor fatigue since these two methods usually provide very different results (Brown, Beightol, Koh, & Eckberg, 1993), especially with athletes (Middleton & De Vito, 2005).

To our knowledge, all the studies comparing the impact of these two breathing methods on HRV results were performed for short periods during a single day (Bernardi et al., 2000; Brown et al., 1993). Indeed, none of them focused on the differences between CB and SB over several days during a longitudinal follow-up of athletes, raising the question of whether HRV measures performed in CB or SB provide the same variations from one day to another? From a practical and methodological point of view, no information exists on the impact of breathing (i.e. spontaneous or controlled) on HRV marker variation during a longitudinal follow-up. However, this respiratory choice is essential given the specific characteristics of well-trained subjects (i.e. slow breathing and strong RSA). It is noteworthy that both SB and CB are still used and there is no gold standard for HRV analysis during an extended period of endurance training (Kiviniemi et al., 2007; Melanson & Freedson, 2001). For example, a difference between the two methods is generally measured during a single day, but although this difference is constant throughout monitoring, it may have no impact on the interpretation of HRV variations during longitudinal studies.

Thus it would be interesting to study whether the day-to-day variations (i.e. increase or decrease) of HRV markers are similar for CB and SB methods. Therefore, the aim of this methodological work was to observe whether or not the two breathing methods provide the same HRV variations during longitudinal studies of healthy trained subjects.

Methods

Subjects

Ten healthy male runners were recruited from a running and athletics club (Age: 29.3 ± 4.8; Weight: 74.5 ± 8 kg; height: 182.1 ± 6.6 cm; $\dot{V}O_{2\ max}$: 65.5 ± 5.8 $ml \cdot min^{-1} \cdot kg^{-1}$). Subjects receiving medical treat mentor with asthma or cardiovascular disorders were excluded. By design, throughout the experiment, the subjects were asked not to change their lifestyles or, in particular, their training loads. This resulted in the day-to-day HRV variations necessary to satisfy the objective of our methodological study.

Protocol

The experiment lasted a total of three weeks. The subjects performed daily HRV measurements for the 21 days of the period. The signals were recorded at home in the morning, on waking. Everyday, the subjects performed two successive HRV measurements in supine position (Buchheit, Al Haddad, Laursen, & Ahmaidi, 2009) in a quiet environment (low brightness) and with an empty stomach. One HRV measurement was performed in CB (during 5 min) and the second in SB (during 5 min) for a total duration of 10 min per day (Schroeder et al., 2004; Togo & Takahashi, 2009). The order of the sessions (CB or SB) was randomised for each subject and maintained throughout the experiment. A soundtrack (metronome) was used to fix the pace of the CB session. The data were collected and recorded using a Suunto T6d Heart Rate Monitor (Suunto, Oy, Finland). This system records RR intervals at a sampling frequency of 1000 Hz and provided reliable results comparable to other ECG recording systems (Weippert et al., 2010). The protocol was approved by the ethical committee of Lyon Sud-Est II and was in accordance with the guidelines set by the Declaration of Helsinki. All the subjects gave their written informed consent.

Choice of controlled breathing frequency (CBF)

In order to determine the CBF, we performed a preliminary study to measure the mean spontaneous breathing frequency (SBF) of our sample population (Mean $= 0.124 \pm 0.026$ Hz). Thus, the CBF was fixed at 0.125 Hz (7.5 breath/min). We conducted a visual control of each HRV power spectrum during the CB session to check whether the energy peak was

centred around 0.125 Hz, in order to verify that BF had been correctly maintained.

HRV parameters

For each session, the signal was analysed in the temporal, non-linear and frequency domains (Task-Force, 1996). For the temporal domain, the mean heart rate (HR), the SDNN (the standard deviation of normal-to-normal RR intervals) and the RMSSD (the square root of the mean squared differences between successive normal-to-normal RR intervals) were considered. Resting HR is a health indicator used by all general practitioners; SDNN corresponds to total variability and RMSSD to short-term variability (Task-Force, 1996). Two markers of the non-linear domain were also measured: SD1 and SD2. The SD1 values characterise short-term HRV and SD2 describes long-term HRV (Tulppo, Makikallio, Takala, Seppanen, & Huikuri, 1996). The frequency domain was assessed by calculating the low-frequency power (LF) (from 0.04 to 0.15 Hz), high-frequency power (HF) (from 0.15 to 0.4 Hz), total power (TP) and the LF/HF ratio (Task-Force, 1996). It is commonly accepted that LF are influenced by both sympathetic and parasympathetic systems, whereas HF is largely thought to reflect parasympathetic modulation (Carney et al., 2001; Task-Force, 1996). Similarly, some authors consider that TP corresponds to total HRV variability and that the LF/HF ratio represents the sympathovagal balance reflecting autonomic nervous system activity (Pagani et al., 1986; Task-Force, 1996). The frequency content of the signals was analysed using custom-made software programmed in C# (Saboul et al., in press). After ectopic removal, we performed a Discrete Fourier Transform with Adjusted Rectangular Windowing on the signals re-sampled at 4 Hz by cubic spline interpolation. When either the CB or SB file was unusable (e.g. unusable non stationary HRV curve explained by subject movement or environmental disturbance), the file concerned (i.e. CB or SB) was removed from the analysis ($n = 28$ files on a total of 420).

Statistical analysis

The results of all the HRV markers obtained for each subject in SB and CB during the 21-day experiment are expressed as means and standard deviations. The relative difference between the two respiration modes was calculated for each marker using Equation 1. One-way (SB vs. CB) repeated-measurement (time effect: 21 days) analysis of variance (ANOVA) followed by a post hoc test was used for the multiple comparison of the HRV data. The relative differences may not totally represent the systematic individual differences that can exist between the two respiration modes throughout the experiment (i.e. one subject may present SB values systematically higher than CB values over the 21 days, whereas another subject may present SB values systematically lower than CB ones). Consequently, we also analysed the difference between the breathing methods that provided the highest (BMsup) and the lowest (BMinf) HRV values for each subject (BMsup vs. BMinf; see Table II for the individual determination of BMsup and BMinf).

$$\%\ \text{diff}\ (\text{RMSSD}) = 100 \times \left(\frac{\overline{\text{RMSSD}}_{\text{SB}} - \overline{\text{RMSSD}}_{\text{CB}}}{(\overline{\text{RMSSD}}_{\text{SB}} - \overline{\text{RMSSD}}_{\text{CB}})/2} \right) \quad (1)$$

To explore respiratory interaction over the 21 days, linear regressions were performed between SB and CB for every HRV marker. Indeed, the variation of differences between the two types of breathing may be constant over time. Thus, for each subject, R Pearson coefficients were calculated individually with all the daily measurements obtained in SB and CB. For each HRV marker, the means and standard deviations of the correlation coefficients obtained with all the subjects were calculated to compare the influence of respiration. The significance of R coefficients was tested with a Bravais-Pearson table. A Z Fisher transformation was performed to obtain a normal distribution followed by a paired student's t-test, in order to compare the R coefficients.

The data were analysed using StatSoft software (Statistica 7.1, StatSoftinc., USA) and the statistical significance threshold was set at $p < 0.05$.

Results

The mean and standard deviation of each HRV marker are presented in Table I. Only two markers, RMSSD and SD1 are not significantly different between SB and CB while all the frequency markers present significant differences. HR, SDNN, SD2, LF, TP and the LH/HF ratio are significantly higher when using CB compared to SB, whereas HF is lower in CB.

Table II gives very significant differences between the two breathing methods of each subject throughout the 21 days. The sign (minus or plus) of the relative difference was directly related to the position of SBF compared to CBF (i.e. an SBF higher than CBF provides negative results while an SBF lower than CBF provides positive results). When calculated from the absolute difference between SB and

Table I. Relative differences between spontaneous and controlled breathing for each HRV marker from 10 subjects, 21 days, $n=182$.

HRV markers	Spontaneous breathing Mean ± SD	Controlled breathing mean ± SD	Mean difference (%)	Mean |difference| (%)
Temporal domain				
HR (bpm)	48.4 ± 9.4	50.3 ± 9.5	+3.82⋆	3.82⋆⋆
SDNN (ms)	110 ± 50	118 ± 51	+6.92⋆	20.7⋆⋆
RMSSD (ms)	104 ± 67	102 ± 61	−1.64	23.3⋆⋆
Non-linear domain				
SD1 (ms)	73 ± 48	72 ± 43	−1.64	23.3⋆⋆
SD2 (ms)	134 ± 59	148 ± 61	+9.96⋆	22.0⋆⋆
Frequency domain				
LF (ms^2/Hz)	2734 ± 3016	4868 ± 4192	+56.16⋆	64.0⋆⋆
HF (ms^2/Hz)	1773 ± 2201	958 ± 1263	−59.72⋆	64.8⋆⋆
TP (ms^2/Hz)	6769 ± 6160	7691 ± 6275	+12.75⋆	39.7⋆⋆
LF/HF	2.7 ± 3.0	9.3 ± 7.3	+110⋆	110⋆⋆

Spontaneous Breathing vs. Controlled Breathing: ⋆$p<0.05$.
Breathing Method inferior vs. Breathing method superior: ⋆⋆$p<0.05$
Mean difference was calculated as follows: 100 × ((SB-CB)/ ((SB-CB)/2))
Mean of absolute value of the individual difference (|Difference|) was calculated as follows: 100 × |(SB-CB)/ ((SB-CB)/2)|

CB, all the HRV markers were significantly affected by the breathing method (fourth column of Table I).

The day-to-day variation of RMSSD and the LF/HF ratio in SB and CB during the 21-day experiment for a typical subject is shown in the left panel of Figure 1. Linear regressions calculated between SB and CB values are represented in the right panel of Figure 1. Despite a significant difference between RMSSD values in SB and CB (−24.0%; $p<0.01$), we found a very significant correlation between the two breathing methods ($R=0.78$; $p<0.01$). We also found a significant difference between the LF/HF ratio during the 21-day experiment (82%; $p<0.01$), but the correlation coefficients obtained for this marker were very poor ($R=-0.04$; $p=0.9$).

The correlation coefficient calculation was performed for all the HRV markers of each subject, as shown in Figure 1 which represents the data for one subject. The distributions of correlation coefficients are shown in Figure 2. HR, RMSSD and SD1 are clearly consistent and present a good correlation of the means (0.81 ± 0.12; 0.73 ± 0.12; 0.73 ± 0.13, respectively). In addition, for these 3 HRV markers all the subjects presented a significant R coefficient between SB and CB (i.e. the number of significant correlation coefficients is presented at the bottom of Figure 2 for each HRV markers). We observed a more heterogeneous distribution of SDNN, SD2, LF and TP correlation coefficients (0.54 ± 0.22; 0.47 ± 0.22; 0.50 ± 0.20; 0.51 ± 0.20, respectively). Moreover, these markers were significantly correlated for only 6 and 7 subjects (out of 10). The HF and LF/HF markers were distributed very heterogeneously (0.40 ± 0.36; 0.14 ± 0.34, respectively). Only five subjects presented a significant correlation coefficient for HF while two subjects presented a significant correlation coefficient for the LF/HF ratio.

Discussion

The main objective of this study was to compare two respiration methods during the HRV monitoring of

Table II. Relative differences between spontaneous and controlled breathing for each subject from RMSSD markers for 21 days.

Subject	SB mean ± SD (ms)	CB mean ± SD (ms)	Relative difference (%)	SBF (Hz)	BMsup
1 ($n=20$)	219 ± 25	198 ± 23	10.3⋆	0.10	Spontaneous
2 ($n=21$)	135 ± 48	85 ± 40	45.6⋆	0.11	Spontaneous
3 ($n=21$)	75 ± 23	97 ± 29	−24.8⋆	0.14	Controlled
4 ($n=21$)	141 ± 29	179 ± 42	−24.0⋆	0.16	Controlled
5 ($n=13$)	134 ± 115	74 ± 61	57.4⋆	0.11	Spontaneous
6 ($n=18$)	113 ± 24	128 ± 22	−12.2⋆	0.13	Controlled
7 ($n=16$)	45 ± 15	46 ± 14	−2.2	0.12	Controlled
8 ($n=21$)	35 ± 4	34 ± 4	0.9	0.12	Spontaneous
9 ($n=21$)	62 ± 14	86 ± 20	−31.6⋆	0.17	Controlled
10 ($n=10$)	60 ± 11	54 ± 11	11.6⋆	0.08	Spontaneous

Spontaneous Breathing vs. Controlled Breathing: ⋆$p<0.05$
SB, Spontaneous Breathing; CB, Controlled Breathing; SBF, Spontaneous Breathing Frequency; BMsup, Breathing Method Superior (which provides the greatest RMSSD value).

athletes. Although there were significant differences between spontaneous and CB for all HRV markers, this work showed that day-to-day variations (i.e. HRV variations) were correlated between SB and CB measurement for the RMSSD, SD1 and HR indices. These markers provided similar results during a longitudinal follow-up, regardless of breathing pattern. Conversely, spectral HRV markers like HF and the LF/HF ratio were not significantly correlated and independent day-to-day variations were observed between SB and CB data over time.

The critical choice of the CBF

Although the choice of the CB frequency was one of the central features of this work, the literature provided a very wide range of imposed respiratory frequencies and there is no "gold standard" for this critical choice (Bloomfield et al., 2001; Brown et al., 1993). Nevertheless, the CBF can be predefined according to the type of population studied (Hayano et al., 1994). For example, a CBF of 0.25 Hz is typically used for sedentary subjects (Bernardi et al., 2000) and some studies have used a CBF ranging between 0.10 and 0.166 Hz for athletes (De Meersman, 1993; Melanson & Freedson, 2001). As suggested by several authors, we conducted pilot testing to determine the mean SBF of our athlete population (mean $=0.124 \pm 0.026$ Hz) in order to set the CBF at 0.125 Hz. This choice of frequency may appear very low but it was in line with other studies that also focused on athlete populations (e.g. 0.125 Hz or 0.10 Hz (Boutcher & Stein, 1995; De Meersman, 1993)). In addition, during the SB session a large part of the spectral energy was already naturally located in the LF band, a phenomenon probably caused by the low SBF and strong RSA of our subjects (De Meersman, 1992). Therefore, a CBF close to 0.25 Hz may have caused misallocation of all the energy in the HF band and may have influenced the regulatory mechanisms brought into play.

Influence of training sessions

We acknowledge that a decrease of HRV is usually observed the day after a training session and, conversely, HRV increases after one day of recovery (Aubert, Seps, & Beckers, 2003; Kiviniemi et al., 2010). More specifically, HRV variations observed during longitudinal follow-up may reflect a complex cumulative effect of fatigue during the training cycle rather than the sole result of the training load of the preceding day (Pichot et al., 2000; Plews et al., 2012). Nevertheless, during the 21 days of the experiment, each subject was asked not to change their lifestyle or, in particular, their training loads. By design, we deliberately chose to ensure they maintained their training habits in order to produce significant day-to-day variations of HRV. In our methodological work, these daily HRV variations were necessary to study whether the decrease or increase of HRV from one day to another can be observed in both SB and CB during longitudinal monitoring of athletes.

Differences between SB and CB

As observed previously, the overall analysis of the HRV values presented in Table I demonstrated that the temporal, non-linear and frequency domains are

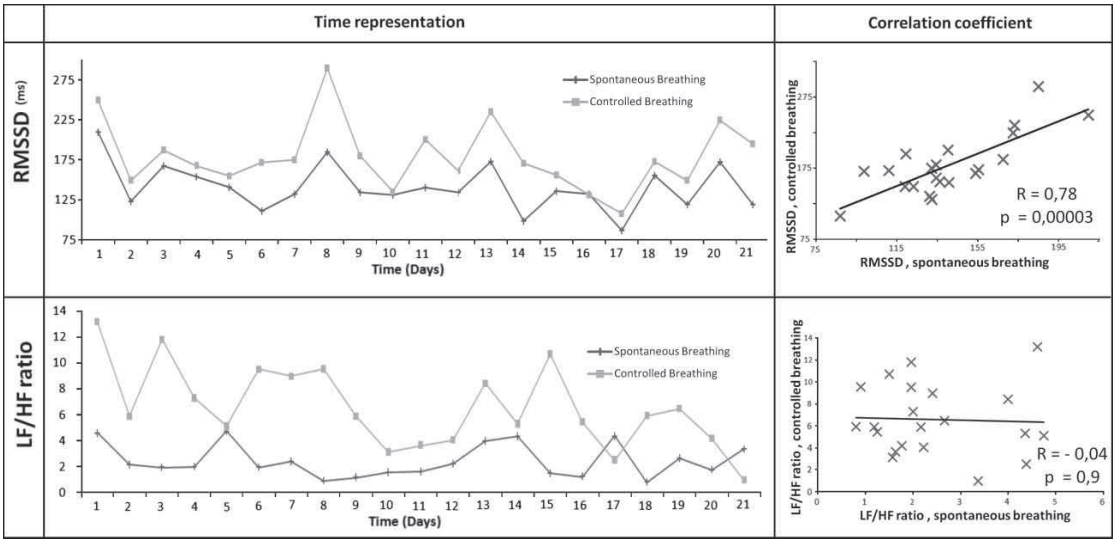

Figure 1. Daily variation of two selected HRV markers for 21 days and the relationship between spontaneous and controlled breathing HRV data for a typical subject.
RMSSD, root mean square of successive difference; LF/HF ratio, low frequency/high frequency ratio.

affected by the mode of respiration (Brown et al., 1993; Eckberg, 2003; Hirsch & Bishop, 1981). Modifications caused by CB conditions can be attributed to the cognitive task of correctly controlling breathing and thus cause higher sympathetic activation (Bernardi et al., 2000). In addition, during CB the strong influence of RSA disturbed the HRV frequency markers which mechanically concentrate most of the spectral energy around the CBF (Saboul et al., in press).

It is noteworthy that inter-individual differences exist for most HRV indices and especially for the RMSSD and SD1 markers. Indeed, as highlighted in Table II, subjects with an SBF lower than the CBF (i.e. 0.125 Hz) exhibited positive RMSSD and SD1 differences between SB and CB during the 21 days of the experiment. Conversely, subjects with an SBF higher than the CBF exhibited negative RMSSD and SD1 differences between SB and CB. In brief, the vagal tone and the global activity of the autonomic nervous system were disturbed by respiration and our results were in line with previous works (Bernardi et al., 2000; Eckberg, 2003).

Monitoring the athletes' training using HRV measures: SB vs. CB

Absolute HRV values are never used during longitudinal follow-ups and many authors prefer to focus on the relative day-to-day variation which can indicate athlete fatigue (Kiviniemi et al., 2010, 2007; Plews et al., 2012). For example, the work performed by Kiviniemi et al. (2010), illustrates this daily follow-up: increase or no change of HRV (i.e. compared to the previous day) resulted in the prescription for high intensity training on that day. By contrast, low-intensity training or rest was prescribed if HRV decreased. Consequently, the main goal of our study was to determine whether HRV measurements performed during CB or SB can provide similar variations from one day to another.

As illustrated in Figures 1 and 2, the significant correlations between CB and SB for the RMSSD, SD1 and HR indices suggested that HRV variations are likely to be similar regardless of breathing pattern and demonstrate that the two respiratory methods provided the same results (i.e. HRV variations) for these 3 markers. By contrast, HF and the LF/HF ratio provided very heterogeneous variations with SB and CB trends that do not follow the same variations throughout the experiment (Figure 1). This lack of correlation between the two respiration modes raises a major issue. Indeed, the LF/HF ratio is one of the main physiological markers of the sympathovagal balance and it had been used for detecting overtraining in certain previous studies (Mourot et al., 2004; Pagani et al., 1986). In addition, some authors performed HRV measurements in SB (Kiviniemi et al., 2007) while others in CB (Melanson & Freedson, 2001). Consequently, during a longitudinal follow-up, the two breathing patterns do not provide the same spectral HRVvariations and the interpretation made by the coach can lead to serious errors such as the non-detection of fatigue or overtraining syndrome. To conclude, SB or CB may be used indifferently and reliably to monitor training and fatigue with the RMSSD and SD1 markers. On the contrary, it is impossible to determine which breathing pattern is the most reliable for studying frequency markers since they present very different day-to-day variations.

Reliability of spectral HRV markers in athletes

Athletes are characterised by a low SBF (close to 0.15 Hz) and a strong RSA (De Meersman, 1992; Middleton & De Vito, 2005). Thus, regardless of breathing method, spectral energy is concentrated mechanically around the BF and the remaining spectral energy on either side (i.e. in the HF band) can be considered as residual information (Brown et al., 1993). Thus the impact of breathing mechanically cancels all the physiological manifestations of fatigue and the LF/HF ratio does not seem to provide a faithful representation of the sympathovagal balance (Saboul et al., in press). This finding is confirmed by our results which show that frequency HRV markers are not reliable, even during the longitudinal follow-up of an athlete. Indeed, we observed a strong day-to-day variation which has never been reported in the literature (e.g. in Figure 1, between day 17 and day 18, the LF/HF ratio ranged from 4 to 0.5 in SB and from 3 to 6 in CB). Moreover, the information provided by this ratio with the two breathing methods is totally

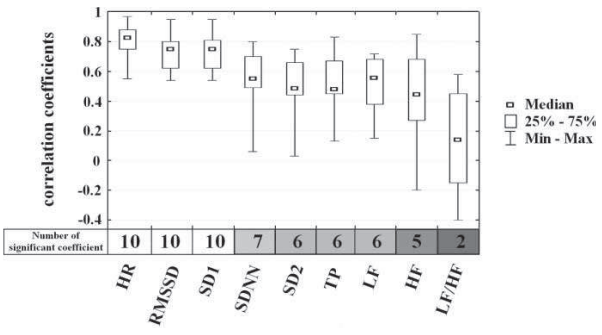

Figure 2. Distribution of correlation coefficients of 10 subjects for each HRV marker.
HR, heart rate; RMSSD, root mean square of successive difference; SD1, standard deviation 1; SDNN, standard deviation of N-N interval; SD2, standard deviation 2; TP, total power; LF, low frequency; HF, high frequency; LF/HF ratio, low frequency/high frequency ratio.

contradictory (increase with CB and decrease with SB). Thus we believe that in well-trained subjects presenting an SBF close to 0.15 Hz, the LF/HF ratio strongly depends on the respiratory frequency in SB and CB, which may obscure the influence of fatigue status (Middleton & De Vito, 2005; Sinnreich, Kark, Friedlander, Sapoznikov, & Luria, 1998).

Consequently, to perform more relevant longitudinal monitoring, the authors now prefer to use RMSSD or SD1 markers which are related to athletes' fatigue (Kiviniemi et al., 2010; Plews et al., 2012) and provide the same variations whatever the breathing pattern.

Limitations of the study

Tidal volume was not controlled during this experiment, although its effect on the HRV power spectrum has been reported previously (Brown et al., 1993). Nevertheless, for both SB and CB, recent works have not detected significant differences between the LF/HF ratio by artificially increasing or decreasing the baseline tidal volume (Poyhonen, Syvaoja, Hartikainen, Ruokonen, & Takala, 2004). In addition, the measurement of tidal volumes using uncomfortable mouthpieces may affect HRV data as have been shown during mental stress (Bernardi et al., 2000). Finally, our goal was to compare SB and CB during a longitudinal follow-up under real conditions and in this context, for technical and practical reasons, the athletes measured their HRV at home using a simple HR monitor without controlling tidal volume (Kiviniemi et al., 2007).

Conclusion

As in previous studies, we found that there were differences between HRV markers obtained with SB and CB during a single measurement. Nevertheless, the present findings show that these differences remain stable over time for HR, RMSSD and SD1 during a longitudinal follow-up. Thus these markers can be used regardless of breathing method since each of them provides exactly the same tendency through time. It is noteworthy that none of the frequency markers provided the same day-to-day variation with the two breathing patterns, while HRV spectral indices were strongly mediated by respiration, especially in athletes. In this context and in agreement with recent works, we suggest using RMSSD or SD1 markers to perform relevant and reliable longitudinal HRV monitoring in order to prevent overtraining.

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References

Akselrod, S., Gordon, D., Ubel, F. A., Shannon, D. C., Berger, A. C., & Cohen, R. J. (1981). Power spectrum analysis of heart rate fluctuation: A quantitative probe of beat-to-beat cardiovascular control. *Science*, *213*(4504), 220–222. doi:10.1126/science.6166045

Aubert, A. E., Seps, B., & Beckers, F. (2003). Heart rate variability in athletes. *Sports Medicine*, *33*(12), 889–919. doi:10.2165/00007256-200333120-00003

Baumert, M., Brechtel, L., Lock, J., Hermsdorf, M., Wolff, R., Baier, V., & Voss, A. (2006). Heart rate variability, blood pressure variability, and baroreflex sensitivity in overtrained athletes. *Clinical Journal of Sport Medicine*, *16*(5), 412–417. doi:10.1097/01.jsm.0000244610.34594.07

Ben Lamine, S., Calabrese, P., Perrault, H., Dinh, T. P., Eberhard, A., & Benchetrit, G. (2004). Individual differences in respiratory sinus arrhythmia. *American Journal of Physiology – Heart Circulatory Physiology*, *286*(6), H2305–2312. Retrieved from http://ajpheart.physiology.org/content/286/6/H2305.short; http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=14751864

Bernardi, L., Wdowczyk-Szulc, J., Valenti, C., Castoldi, S., Passino, C., Spadacini, G., & Sleight, P. (2000). Effects of controlled breathing, mental activity and mental stress with or without verbalization on heart rate variability. *Journal of the American College of Cardiology*, *35*(6), 1462–1469. doi:10.1016/S0735-1097(00)00595-7

Bloomfield, D. M., Magnano, A., Bigger, J. T., Jr., Rivadeneira, H., Parides, M., & Steinman, R. C. (2001). Comparison of spontaneous vs. metronome-guided breathing on assessment of vagal modulation using RR variability. *American Journal of Physiology – Heart Circulatory Physiology*, *280*(3), H1145–1150. Retrieved from http://ajpheart.physiology.org/content/280/3/H1145.short; http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=11179058

Boutcher, S. H., & Stein, P. (1995). Association between heart rate variability and training response in sedentary middle-aged men. *European Journal of Applied Physiology and Occupational Physiology*, *70*(1), 75–80. doi:10.1007/BF00601812

Brown, T. E., Beightol, L. A., Koh, J., & Eckberg, D. L. (1993). Important influence of respiration on human R-R interval power spectra is largely ignored. *Journal of Applied Physiology*, *75*(5), 2310–2317. Retrieved from http://jap.physiology.org/content/75/5/2310.short; http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=8307890

Buchheit, M., Al Haddad, H., Laursen, P. B., & Ahmaidi, S. (2009). Effect of body posture on postexercise parasympathetic reactivation in men. *Experimental Physiology*, *94*(7), 795–804. doi:10.1113/expphysiol.2009.048041

Carney, R. M., Blumenthal, J. A., Stein, P. K., Watkins, L., Catellier, D., Berkman, L. F., ... Freedland, K. E. (2001). Depression, heart rate variability, and acute myocardial infarction. *Circulation*, *104*(17), 2024–2028. doi:10.1161/hc4201.097834

De Meersman, R. E. (1992). Respiratory sinus arrhythmia alteration following training in endurance athletes. *European Journal of Applied Physiology and Occupational Physiology*, *64*(5), 434–436. doi:10.1007/BF00625063

De Meersman, R. E. (1993). Heart rate variability and aerobic fitness. *American Heart Journal*, *125*(3), 726–731. doi:10.1016/0002-8703(93)90164-5

Eckberg, D. L. (2003). The human respiratory gate. *Journal of Physiology*, *548*(Pt 2), 339–352. Retrieved from http://jp.physoc.org/content/548/2/339.short; http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=12626671

Gregoire, J., Tuck, S., Yamamoto, Y., & Hughson, R. L. (1996). Heart rate variability at rest and exercise: Influence of age, gender, and physical training. *Canadian Journal of Applied Physiology*, *21*(6), 455–470. doi:10.1139/h96-040

Hayano, J., Mukai, S., Sakakibara, M., Okada, A., Takata, K., & Fujinami, T. (1994). Effects of respiratory interval on vagal modulation of heart rate. *American Journal of Physiology*, *267*(1 Pt 2), H33–40. Retrieved from http://ajpheart.physiology.org/content/267/1/H33.short; http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=7914066

Hedelin, R., Wiklund, U., Bjerle, P., & Henriksson-Larsen, K. (2000). Cardiac autonomic imbalance in an overtrained athlete. *Medicine and Science in Sports & Exercise*, *32*(9), 1531–1533. Retrieved from http://europepmc.org/abstract/MED/10994900; http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=10994900

Hirsch, J. A., & Bishop, B. (1981). Respiratory sinus arrhythmia in humans: How breathing pattern modulates heart rate. *American Journal of Physiology*, *241*(4), H620–629. Retrieved from http://ajpheart.physiology.org/content/241/4/H620.short; http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=7315987

Kiviniemi, A. M., Hautala, A. J., Kinnunen, H., Nissila, J., Virtanen, P., Karjalainen, J., & Tulppo, M. P. (2010). Daily exercise prescription on the basis of HR variability among men and women. *Medicine and Science in Sports & Exercise*, *42*(7), 1355–1363. Retrieved from http://www.ncbi.nlm.nih.gov/pubmed/20575165

Kiviniemi, A. M., Hautala, A. J., Kinnunen, H., & Tulppo, M. P. (2007). Endurance training guided individually by daily heart rate variability measurements. *European Journal of Applied Physiology*, *101*(6), 743–751. doi:10.1007/s00421-007-0552-2

La Rovere, M. T., Pinna, G. D., Maestri, R., Mortara, A., Capomolla, S., Febo, O., … Cobelli, F. (2003). Short-term heart rate variability strongly predicts sudden cardiac death in chronic heart failure patients. *Circulation*, *107*(4), 565–570. doi:10.1161/01.CIR.0000047275.25795.17

Larsen, P. D., Tzeng, Y. C., Sin, P. Y., & Galletly, D. C. (2010). Respiratory sinus arrhythmia in conscious humans during spontaneous respiration. *Respiratory Physiology & Neurobiology*, *174*(1–2), 111–118. doi:10.1016/j.resp.2010.04.021

Melanson, E. L., & Freedson, P. S. (2001). The effect of endurance training on resting heart rate variability in sedentary adult males. *European Journal of Applied Physiology*, *85*(5), 442–449. doi:10.1007/s004210100479

Middleton, N., & De Vito, G. (2005). Cardiovascular autonomic control in endurance-trained and sedentary young women. *Clinical Physiology and Functional Imaging*, *25*(2), 83–89. doi:10.1111/j.1475-097X.2004.00594.x

Mourot, L., Bouhaddi, M., Perrey, S., Cappelle, S., Henriet, M. T., Wolf, J. P., … Regnard, J. (2004). Decrease in heart rate variability with overtraining: assessment by the Poincare plot analysis. *Clinical Physiology and Functional Imaging*, *24*(1), 10–18. doi:10.1046/j.1475-0961.2003.00523.x

Ori, Z., Monir, G., Weiss, J., Sayhouni, X., & Singer, D. H. (1992). Heart rate variability. Frequency domain analysis. *Cardiology Clinics*, *10*(3), 499–537. Retrieved from http://www.ncbi.nlm.nih.gov/pubmed/1504981

Pagani, M., Lombardi, F., Guzzetti, S., Rimoldi, O., Furlan, R., Pizzinelli, P., … Malliani, A. (1986). Power spectral analysis of heart rate and arterial pressure variabilities as a marker of sympatho-vagal interaction in man and conscious dog. *Circulation Research*, *59*(2), 178–193. doi:10.1161/01.RES.59.2.178

Penttila, J., Helminen, A., Jartti, T., Kuusela, T., Huikuri, H. V., Tulppo, M. P., … Scheinin, H. (2001). Time domain, geometrical and frequency domain analysis of cardiac vagal outflow: Effects of various respiratory patterns. *Clinical Physiology*, *21*(3), 365–376. doi:10.1046/j.1365-2281.2001.00337.x

Pichot, V., Roche, F., Gaspoz, J. M., Enjolras, F., Antoniadis, A., Minini, P., … Barthelemy, J. C. (2000). Relation between heart rate variability and training load in middle-distance runners. *Medicine & Science in Sports & Exercise*, *32*(10), 1729–1736. doi:10.1097/00005768-200010000-00011

Plews, D. J., Laursen, P. B., Kilding, A. E., & Buchheit, M. (2012). Heart rate variability in elite triathletes, is variation in variability the key to effective training? A case comparison. *European Journal of Applied Physiology*, *112*(11), 3729–3741. doi:10.1007/s00421-012-2354-4

Poyhonen, M., Syvaoja, S., Hartikainen, J., Ruokonen, E., & Takala, J. (2004). The effect of carbon dioxide, respiratory rate and tidal volume on human heart rate variability. *Acta Anaesthesiologica Scandinavica*, *48*(1), 93–101. doi:10.1111/j.1399-6576.2004.00272.x

Rajendra Acharya, U., Paul Joseph, K., Kannathal, N., Lim, C. M., & Suri, J. S. (2006). Heart rate variability: A review. *Medical & Biological Engineering & Computing*, *44*(12), 1031–1051. doi:10.1007/s11517-006-0119-0

Saboul, D., Pialoux, V., & Hautier, C. (in press). The breathing effect of the LF/HF ratio in the heart rate variability measurements of athletes. *European Journal of Sport Science*, 1–7.

Schipke, J. D., Pelzer, M., & Arnold, G. (1999). Effect of respiration rate on short-term heart rate variability. *Journal of Clinical & Basic Cardiology*, *2*(1), 92–95. Retrieved from http://www.kup.at/journals/summary/39.html

Schroeder, E. B., Whitsel, E. A., Evans, G. W., Prineas, R. J., Chambless, L. E., & Heiss, G. (2004). Repeatability of heart rate variability measures. *Journal of Electrocardiology*, *37*(3), 163–172. doi:10.1016/j.jelectrocard.2004.04.004

Sinnreich, R., Kark, J. D., Friedlander, Y., Sapoznikov, D., & Luria, M. H. (1998). Five minute recordings of heart rate variability for population studies: Repeatability and age-sex characteristics. *Heart*, *80*(2), 156–162. Retrieved from http://heart.bmj.com/content/80/2/156.short

Strano, S., Lino, S., Calcagnini, G., Di Virgilio, V., Ciardo, R., Cerutti, S., … Caselli, G. (1998). Respiratory sinus arrhythmia and cardiovascular neural regulation in athletes. *Medicine &amp Science in Sports &amp Exercise*, *30*(2), 215–219. doi:10.1097/00005768-199802000-00007

Task-Force. (1996). Heart rate variability. Standards of measurement, physiological interpretation, and clinical use. Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology. *European Heart Journal*, *17*(3), 354–381. doi:10.1093/oxfordjournals.eurheartj.a014868

Togo, F., & Takahashi, M. (2009). Heart rate variability in occupational health – a systematic review. *Industrial Health*, *47*(6), 589–602. doi:10.2486/indhealth.47.589

Tsuji, H., Larson, M. G., Venditti, F. J., Jr., Manders, E. S., Evans, J. C., Feldman, C. L., & Levy, D. (1996). Impact of reduced heart rate variability on risk for cardiac events. The Framingham Heart Study. *Circulation*, *94*(11), 2850–2855. doi:10.1161/01.CIR.94.11.2850

Tulppo, M. P., Makikallio, T. H., Takala, T. E., Seppanen, T., & Huikuri, H. V. (1996). Quantitative beat-to-beat analysis of heart rate dynamics during exercise. *American Journal of Physiology*, *271*(1 Pt 2), H244–252. Retrieved from http://ajpheart.physiology.org/content/271/1/H244.short; http://www.

ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=8760181

Weippert, M., Kumar, M., Kreuzfeld, S., Arndt, D., Rieger, A., & Stoll, R. (2010). Comparison of three mobile devices for measuring R-R intervals and heart rate variability: Polar S810i, Suunto t6 and an ambulatory ECG system. *European Journal of Applied Physiology*, *109*(4), 779–786. doi:10.1007/s00421-010-1415-9