Heliyon 5 (2019) 1–8

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.heliyon.com

Heart rate variability in pulmonary hypertension with and without sleep apnea

Carolina Gonzaga Carvalho [a,b], Richard Bresler [a], Ying Xuan Zhi [a], Hisham Alshaer [a], John T. Granton [c,d], Clodagh M. Ryan [a,d,*]

[a] Sleep Research Laboratory, Toronto Rehabilitation Institute, University Health Network, Toronto, ON, Canada
[b] Sleep Laboratory, Dante Pazzanese Institute of Cardiology, Sao Paulo, Brazil
[c] Pulmonary Hypertension Program, University Health Network, Toronto General Hospital, Canada
[d] Department of Medicine, University of Toronto, Canada

ARTICLE INFO

Keywords:
Cardiology
Physiology
Autonomic modulation
Heart rate variability
Sleep apnea
Pulmonary hypertension

ABSTRACT

Objectives: Our aims were to evaluate HRV in pulmonary hypertension (WHO Group 1 and 4) compared to control subjects, and to assess whether the presence of sleep apnea in those with pulmonary hypertension would be deleterious and cause greater impairment in HRV.
Methods: This retrospective case-control study analyzed electrocardiogram segments obtained from diagnostic polysomnography.
Results: Forty-one pulmonary hypertension patients were compared to 41 age, sex and apnea-hypopnea index matched healthy controls. The pulmonary hypertension group had decreased high frequency, very low frequency, low frequency, and percentage of normal R-R intervals that differ by > 50 ms compared to control subjects. Moderate to severe right ventricle dysfunction on echocardiography was a predictor of lower high frequency in pulmonary hypertension patients.
Conclusions: There were no differences in any HRV measures in pulmonary hypertension patients with or without sleep apnea. Impaired HRV was demonstrated in pulmonary hypertension patients however, the presence of sleep apnea did not appear to further reduce vagal modulation.

1. Introduction

Heart rate variability (HRV), the fluctuation in time intervals between consecutive heartbeats, is regulated by both the sympathetic and parasympathetic divisions of the autonomic nervous system at the sino-atrial node and provides an assessment of autonomic nervous system modulation (Pagani et al., 1986; Task Force of ESC/NASPE, 1996). Decreased HRV portends a worse prognosis in the general population (Hillebrand et al., 2013; Maheshwari et al., 2016) and in particular in those with heart failure (Danilowicz-Szymanowicz et al., 2016). Moreover, the clinical importance of low HRV has been recognized for risk stratification for sudden arrhythmic death in the general population (Maheshwari et al., 2016).

Pulmonary artery hypertension (PAH) is a progressive pulmonary vascular disorder characterized by progressive narrowing of the pulmonary vessels (Galie et al., 2016) producing an increase in the afterload to the naïve right ventricle (RV) with subsequent RV failure and death. Due to comorbid conditions including obesity and increased age PAH patients are predisposed to sleep apnea (SA) with high prevalence rates demonstrated in small cohort studies (Minic et al., 2014). Increased sympathetic activation occurs in both PAH (McGowan et al., 2009; Velez-Roa et al., 2004) and obstructive sleep apnea (OSA) (Somers et al., 1995). In PAH it is an independent predictor of clinical deterioration (Ciarka et al., 2010). Furthermore, impaired HRV has been demonstrated in PAH (McGowan et al., 2009; Wensel et al., 2009; Yi et al., 2014) and OSA (Narkiewicz et al., 1998; Wang et al., 2008; Wiklund et al., 2000).

The pathophysiological impact of SA on PAH is uncertain, although evidence from studies on those with other cardiac conditions suggests that it would have both an acute and chronic adverse hemodynamic impact (Bradley and Floras, 2003). The long-term prognosis of PAH was dismal prior to the availability of recent therapeutic options for PAH (D'Alonzo et al., 1991). However, with both the increased recognition of, and superior survival in PAH determining the true impact of SA in PAH may be important (Sitbon et al., 2016). HRV provides a non-invasive

* Corresponding author.
E-mail address: clodagh.ryan@uhn.ca (C.M. Ryan).

https://doi.org/10.1016/j.heliyon.2019.e02034
Received 26 November 2018; Received in revised form 1 May 2019; Accepted 28 June 2019

assessment of autonomic modulation and in light of its prognostic value in the general population it may also be of importance in the PAH group. Our hypothesis was that SA would be deleterious to those with pulmonary hypertension (PH) and therefore, greater sympathovagal modulation would occur in those with SA and PH compared to those with isolated PH. The objectives of our study were to evaluate HRV from polysomnographic data 1) in patients with PH compared to healthy individuals, and 2) in PH and control subjects between those with and without SA.

2. Methods

2.1. Study design and population

This is a retrospective case-control study of adults with PH (WHO Group 1 or 4) (Galie et al., 2016) and matched healthy control patients referred as part of their routine care for diagnostic polysomnography (PSG) at the University Health Network, Toronto, Canada, from January 1st, 2008 to November 27th, 2016. PH was defined as per World Health Organization (WHO) guidelines (Galie et al., 2016). Healthy subjects were matched for age, sex, and apnea-hypopnea index (AHI) to the PH group, and were derived from consecutive PSG performed over the study period. Exclusion Criteria included a) pregnancy, b) pulmonary or cardiac disease c) patients on treatment for SA, d) those on amiodarone, beta-blocker or non-dihydropyridine calcium channel blocker medications, e) subjects with atrial fibrillation, excessive premature beats on the PSG electrocardiogram (ECG) ($\geq$5% beats) or cardiac pacemaker and f) a total sleep time <1.5 h on the PSG.

The clinical and medical information was collected within 6 months of the PSG. For those with PH, information collected also included the right heart catheterization data, two-dimensional echocardiography, and six-minute walk distance. PH subjects were stratified into low, intermediate and high risk (Leuchte et al., 2018). The study was approved by the University Health Network Research Ethics Board (15–9401).

2.2. Polysomnography

A diagnostic PSG was performed in all patients. Thoracoabdominal motion was monitored by respiratory inductance plethysmography; nasal airflow by nasal pressure cannula; oxyhemoglobin saturation (SaO_2) by oximetry; and cardiac rhythm was monitored by a 2-lead ECG. Signals were recorded on a computerized sleep recording system (Sandman, Nellcor Puritan Bennett Ltd, Ottawa, ON, Canada). Standard techniques and scoring criteria were used to evaluate subjects for sleep stages and arousal from sleep (Iber et al., 2007).

The severity of SA was assessed by the number of apneas and hypopneas per hour of sleep (AHI). Apnea scoring required a greater than 90% signal drop for at least 10 seconds, and hypopnea scoring required a greater than 30% reduction in nasal pressure signal excursions from baseline and an associated $\geq$3% desaturation or arousal (Berry et al., 2012). Apneas and hypopneas were scored as central or obstructive in the absence or presence of out-of-phase thoracoabdominal motion. The oxygen desaturation index (ODI) was defined as the number of oxygen desaturations per hour $\geq$3% below baseline. The scoring criteria were consistent over time. Subjects were divided into NSA (AHI <5 events/h) and SA (AHI $\geq$5 events/h) groups. Sleep apnea was deemed obstructive (OSA) if $\geq$ 50% of events were obstructive and central (CSA) if > 50% were central in nature. The Epworth Sleepiness Scale (ESS), a subjective measure of sleepiness, was completed on the night of the PSG (Johns, 1991).

2.3. Heart rate variability

Lead II ECG data (sampled at 256 Hz) from the PSG with at least 1 segment $\geq$5 min of stage N2, with no apnea or hypopnea events (AHI = 0) was used to determine the HRV. Each ECG segment had signal quality verified calculating the difference between the R peaks and the mean energy of a normalized filtered ECG signal. Those that did not meet the minimum score (0.1) were excluded. The degree of relationship between respiration (from respitrace) and HRV at a specific frequency was determined by coherence analysis. Only ECG segments with coherence >0.5 were included (MatLab R2015a, The MathWorks Inc., Natick, MA, US).

HRV was then evaluated by Kubios HRV 2.2 (Kubios, Kuopoio, Finland) using both time and frequency domains analysis (Tarvainen et al., 2014). Time analysis involved calculation of the average of R-R intervals (Mean R-R), Standard deviation of R-R intervals (SDNN) the average of heart rate, the root mean square of successive R-R interval differences (RMSSD), the percentage of normal R-R intervals that differ by > 50 ms (pNN50), and the integral of density distribution (histogram) (that is, number of all R-R intervals) divided by the maximum of the density distribution (R-R triangular index) (Lahiri et al., 2008; Stein and Pu, 2012; Task Force of ESC/NASPE, 1996). Frequency spectrum of ECG segments was calculated using the Fast Fourier Transform, from which 3 frequency bands were derived: very low frequency (VLF) from 0.003 to 0.04 Hz; low frequency (LF) from 0.04 to 0.15 Hz, and high frequency (HF) from 0.15 to 0.4 Hz (Lahiri et al., 2008; Stein and Pu, 2012; Task Force of ESC/NASPE, 1996). The total power of frequencies ranges and the LF/HF ratio were also calculated.

2.4. Statistical analysis

PH and SA groups were analyzed as dichotomous variables (i.e., PH or Control and NSA or SA, respectively). Continuous variables were compared using Student t test or one-way analysis of variance (ANOVA), and Mann-Whitney or Kruskal-Wallis test as appropriate. Discrete data was examined by Chi squared or Fisher exact test (for frequencies <5). Correlations were examined by Pearson's or Spearman's rank for normally and non-normally distributed variables, respectively. Univariate analysis for predictors of HF and LF/HF included age, sex, body mass index (BMI), hypertension, smoking, ESS, indicators of SA (arousals, AHI, cumulative percentage time at SaO2 < 90% (CT90)), indicators of PH (mean pulmonary artery pressure, right atrial pressure, mixed venous oxygen saturation, cardiac index, diastolic pressure gradient, right atrial area, RV function (normal-mild dysfunction vs moderate-severe dysfunction), no-or-trivial vs $\geq$ mild pericardial effusion (Klein et al., 2013), B-type natriuretic peptide (BNP), six-minute walk distance, WHO functional class), as independent variables. Any independent variable with an alpha value <0.1 was included in the multivariable models of analysis of covariance (ANCOVA). Possible confounders (age, BMI, hypertension, AHI, mean pulmonary artery pressure) were also tested. Logarithmic transformation was used to convert HF and LF/HF to satisfy the normal distribution assumption. A p-value of <0.05 was considered significant. Analyses were performed by SPSS 20.0 (SPSS Inc, Chicago, IL).

Sample size was calculated assuming an effect size of 0.88 in HF difference between PH and control groups. A two-tailed α of 0.05 and β of 95% were used. The resulting sample size was 35 patients for each group. A p-value of <0.05 was considered significant. Additionally, after the study competition, we calculated the effect size considering the number of PH patients with sleep SA (n = 25) and without SA (n = 16), using a two-tailed α of 0.05 and β of 95%. The resulting effect size was 1.18.

3. Results

Of 66 PH patients initially screened, 25 were excluded (Fig. 1). Group 1 PH was present in 38 and Group 4 PH in 3 subjects. The mean time from diagnosis of PH was 1.8±3.5 years. Of the 41 PH subjects included 58.5% were on pulmonary vasodilators, 17.1% on calcium channel blockers, 39.0% on loop diuretics and 29.3% on anti-coagulation medications. The 41 PH patients included were matched for age, sex, and AHI to the control group. Subjects in both groups were predominantly female,

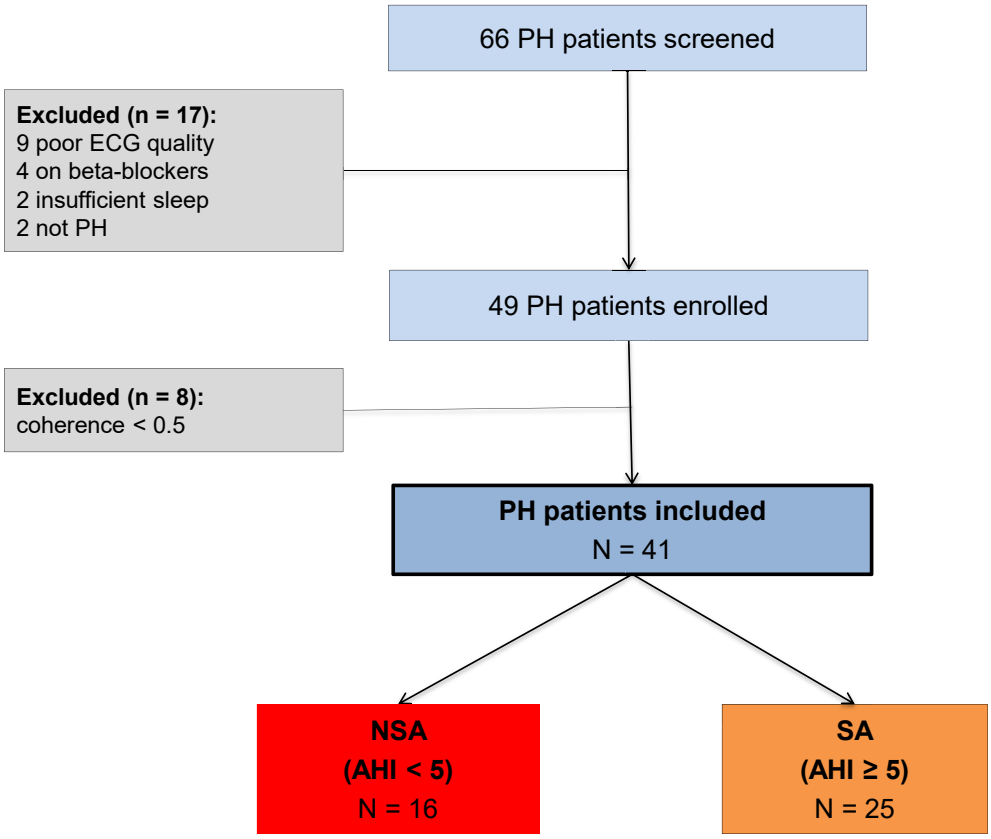

Fig. 1. Flow diagram. Abbreviations: PH = pulmonary hypertension, ECG = electrocardiogram, NSA = no sleep apnea, SA = sleep apnea, AHI = apnea-hypopnea index.

middle aged, and overweight (Table 1). PH patients had a higher prevalence of dyslipidemia and hypertension compared to the control group. Subjects were not subjectively sleepy as measured by the ESS (7.1±4.2). Compared to controls, the PH subjects had a significantly greater frequency of CSA and lower mean nocturnal SaO_2.

Table 1
Baseline characteristics.

	Control N = 41	PH N = 41	p-value
Age, years	48 (13)	47 (15)	0.745
Female sex n (%)	30 (73)	31 (76)	0.800
Body mass index, kg/m^2	29 (6)	30 (7)	0.319
Dyslipidemia n (%)	2 (5)	8 (19)	0.043
Hypertension (%)	0	8 (19)	0.005
Smoking n (%)	0	1 (2)	0.500
Epworth Sleep Scale	7 (4)	7 (4)	0.524
Apnea-hypopnea index, n/h	15 (21)	15 (21)	0.985
Central index, n/h	1 (1)	4 (12)	0.556
Obstructive index, n/h	14 (20)	10 (15)	0.584
Central sleep apnea n (%)	0	6 (15)	0.025
Total sleep time, min	337 (56)	307 (65)	0.027
Stage N1, %	9 (7)	9 (8)	0.933
Stage N2, %	61 (10)	65 (11)	0.091
Stage N3,%	13 (10)	13 (10)	0.963
REM Stage, %	18 (6)	13 (7)	0.003
Arousal index, n/h	22 (14)	22 (19)	0.481
Mean SaO_2, %	95 (2)	90 (4)	<0.001
CT90, %	3 (9)	45 (39)	<0.001
ODI, n/h	12 (18)	16 (17)	0.036
Supplemental oxygen use n (%)	0	4 (10)	0.116

Abbreviations: PH = pulmonary hypertension, REM = rapid-eye movement, SaO_2 = Oxygen saturation; CT90, cumulative percentage time at SaO_2 < 90%, ODI = Oxygen desaturation index.
Results are shown as Mean (SD) or n (%).

3.1. HRV in PH compared to control subjects

In PH patients, there were significantly lower mean R-R, SDNN, pNN50, R-R triangular index, VLF, LF, HF, total power, and higher heart rate compared to control subjects (Table 2). LF/HF did not differ between groups.

3.2. Effect of SA on HRV

When subjects were stratified according to the presence of SA, the control subjects with SA (control-SA, n = 21) had similar demographics

Table 2
Heart rate variability in PH and control subjects.

	Control N = 41	PH N = 41	p-value
Mean R-R, ms	972 (138)	831 (108)	<0.001
SDNN, ms	50 (18)	35 (23)	0.001
Mean heart rate, 1/min	63 (9)	74 (11)	<0.001
RMSSD, ms	41 (22)	31 (29)	0.091
pNN50, %	18 (19)	9 (14)	0.001
R-R triangular index	12 (4)	8 (4)	<0.001
VLF Power, ms^2	1075 (858)	569 (614)	<0.001
LF Power, ms^2	801 (691)	382 (614)	<0.001
HF Power, ms^2	723 (774)	467 (1003)	<0.001
Total Power, ms^2	2600 (1849)	1395 (1825)	<0.001
LF/HF ratio	2 (2)	4 (8)	0.581

Abbreviations: PH = pulmonary hypertension, Mean R-R = average of R-R intervals, SDNN = standard deviation of R-R intervals, RMSSD = root mean square of successive R-R interval differences, pNN50 = percentage of normal R-R intervals that differ by > 50 ms, R-R triangular index = number of all R-R intervals divided by the maximum of the density distribution, VLF = very low frequency, LF = low frequency, HF = high frequency.
Results are shown as Mean (SD).

to the control subjects without SA (control-NSA, n = 20) (Table 3). Polysomnographic characteristics were similar other than an elevated AHI (26 vs 3 events/h, p < 0.001), ODI (25 vs 3/h, p < 0.001) and arousal index (29 vs 16/h, p = 0.047) in the control-SA compared to the control-NSA group. In the control subjects no significant differences between groups were demonstrated for HRV (Fig. 2). However, when control subjects with moderate to severe SA (AHI ≥15 events/h) were compared to control-NSA patients, the mean R-R, RMSSD, pNN50, and HF were significantly lower (Table 4). LF/HF was similar among all groups.

The 25 PH patients with SA (PH-SA) were older, had higher systolic blood pressure (123 ± 14 vs 112 ± 17 mmHg, p = 0.024) and lower BNP (70 ± 94 vs 275 ± 317 ng/L, p = 0.020) compared to those without SA (PH-NSA) (Table 5). Although, the PH risk stratification score was similar between groups. A mild degree of SA was present in the PH−SA group and they had a significantly higher arousal index than the PH−NSA group. However, the mean SaO2, ODI, CT90 and sleep architecture were similar between those with PH−SA and PH−NSA (Table 5). HRV parameters were similar in PH patients with or without SA, irrespective of SA severity (Fig. 2).

3.3. Predictors of HRV

In control subjects, SA severity represented by the AHI and CT90 were negatively correlated with both the RMSSD (r = - 0.324, p = 0.039; r = -0.450, p = 0.003) and pNN50 (r = - 0.316, p = 0.044; r = -0.473, p = 0.002), respectively. HF was inversely correlated to arousals (r = - 0.321, p = 0.041) and CT90 (r = - 0.477, p = 0.002). LF was associated with CT90 (r = - 0.373; p = 0.016), while LF/HF was associated with arousals (r = 0.311, p = 0.048), but not with CT90 (r = 0.151, p = 0.345).

Among PH subjects, there were a number of weak associations of uncertain significance (Table 6). Diastolic pressure gradient was inversely correlated with HRV parameters including the pNN50 (r = -0.401, p = 0.015), RMSSD (r = -0.456, p = 0.005). Of the available markers of PH severity the presence of pericardial effusion (r = 0.387, p = 0.014) and BNP > 50 ng/L (r = −0.237, p = 0.042) were predictors of LF/HF. Moderate to severe RV dysfunction on echocardiography had a weak correlation with lower HF (r = -0.156, p = 0.005), although, - in multivariable models it was an independent predictor of lower HF among PH patients. There was no independent predictor associated with LF/HF (Table 7). The PH risk stratification score was not associated with HRV measures.

4. Discussion

Our study demonstrates that the polysomnographic HRV was significantly decreased in PH compared to control subjects. Secondly, the influence of SA on HRV among PH patients was explored, and contrary to our expectations no additional autonomic modulation was demonstrated.

HRV has been utilized as a tool for the non-invasive assessment of the autonomic nervous system regulation of the sinoatrial node (Shaffer and Ginsberg, 2017; Task Force of ESC/NASPE, 1996). A pathological state is usually indicated by reductions in HRV. However, biological systems are complex and this is reflected by the dynamic interplay between the sympathetic and parasympathetic nervous systems. In the usual healthy state at rest the balance of activity is parasympathetic. The time domain measure of RMSSD and pNN50 are influenced by parasympathetic activity as is HF, the frequency domain which reflects respiratory sinus arrhythmia. Both parasympathetic and sympathetic activity contribute to SDNN, however, in short duration recordings the variability primarily reflects parasympathetic activity. LF is modulated by both the parasympathetic and sympathetic outflow, while the LF/HF is considered to reflect the sympathovagal balance (Shaffer and Ginsberg, 2017). However, increased sympathetic nervous activity may both suppress and augment parasympathetic nervous activity to maintain homeostasis (Tulppo et al., 2005). Understanding this interplay is important when interpreting HRV.

Sleep is a period of quiescence and in non-rapid eye movement sleep there is usually autonomic stability with a reduction in sympathetic nervous activity and a predominance of parasympathetic nervous activity

Table 3
Clinical Characteristics and Polysomnography of Both Control and Pulmonary Hypertension subjects stratified according to the presence of Sleep Apnea.

	C-NSA N = 20	C-SA N = 21	p-value	PH-NSA N = 16	PH-SA N = 25	p-value	Between group p-value
Age, years	44 (15)	52 (11)	0.395	39 (12)[a]	52 (15)	0.004	0.006
Male sex n (%)	5 (25)	6 (29)	0.796	2 (12)	8 (32)	0.265	0.585
Body mass index, kg/m^2	26 (4)	31 (7)	0.056	27 (8)	32 (6)[b]	0.068	0.003
Diabetes n (%)	1 (5)	0	0.488	2 (12)	5 (20)	0.685	0.099
Dyslipidemia n (%)	0	2 (9)	0.488	4 (25)	4 (16)	0.689	0.109
Hypertension n (%)	0	0		2 (12)	6 (24)[a,b]	0.448	0.003
Smoking n (%)	0	0		0	1 (4)	0.390	0.195
Coronary artery disease n (%)	0	0		1 (6)	3 (12)	0.488	0.191
Stroke/TIA n (%)	0	0		2 (12)	1 (4)	0.550	0.150
Epworth Sleepiness Scale	8 (5)	7 (4)	0.999	7 (5)	7 (4)	0.999	0.760
Apnea-hypopnea index, n/h	3 (2)	26 (24)	<0.001	2 (1)[c]	23 (24)[b]	<0.001	<0.001
Central index, n/h	1 (1)	1 (2)	0.869	0	7 (15)	0.176	0.022
Obstructive index, n/h	4 (8)	24 (22)	0.003	2 (1)[c]	16 (17)[d]	0.003	<0.001
Central sleep apnea n (%)	0	0		0	6 (24)[c,d]		0.025
Total sleep time, min	341 (66)	333 (46)	0.999	325 (83)	295 (48)[a]	0.692	0.057
Stage N1, %	8 (6)	10 (8)	0.999	9 (6)	9 (9)	0.999	0.907
Stage N2, %	60 (9)	61 (11)	0.999	65 (11)	64 (11)	0.999	0.379
Stage N3,%	15 (11)	10 (9)	0.607	13 (9)	13 (11)	0.999	0.432
REM stage, %	17 (6)	18 (6)	0.999	13 (7)	13 (6)[a]	0.999	0.024
Arousal index, n/h	16 (7)	29 (17)	0.047	14 (7)[c]	28 (21)	0.025	0.003
Mean SaO_2, %	96 (2)	95 (2)	0.307	90 (4)[b,c]	90 (4)[b,c]	0.999	<0.001
CT90, %	1 (1)	6 (12)	0.972	45 (39)[b,c]	45 (39)[b,c]	0.999	<0.001
ODI, n/h	3 (2)	25 (21)	<0.001	8 (6)	22 (20)[b]	0.149	<0.001
Supplemental oxygen use n (%)	0	0		2 (12)	2 (8)	0.637	0.170

Abbreviations: C = Control, PH = pulmonary hypertension, NSA = no sleep apnea, SA = sleep apnea, TIA, Transitory ischemic attack, REM = rapid-eye movement, SaO2 = Oxygen saturation, CT90 = cumulative percentage time at SaO2 < 90%, ODI = Oxygen desaturation index.
Results are shown as Mean (SD) or n (%).
[a] p < 0.05 compared to Control-SA.
[b] p < 0.01 compared to Control-No-SA.
[c] p < 0.01 compared to Control-SA.
[d] p < 0.05 compared to Control-No-SA.

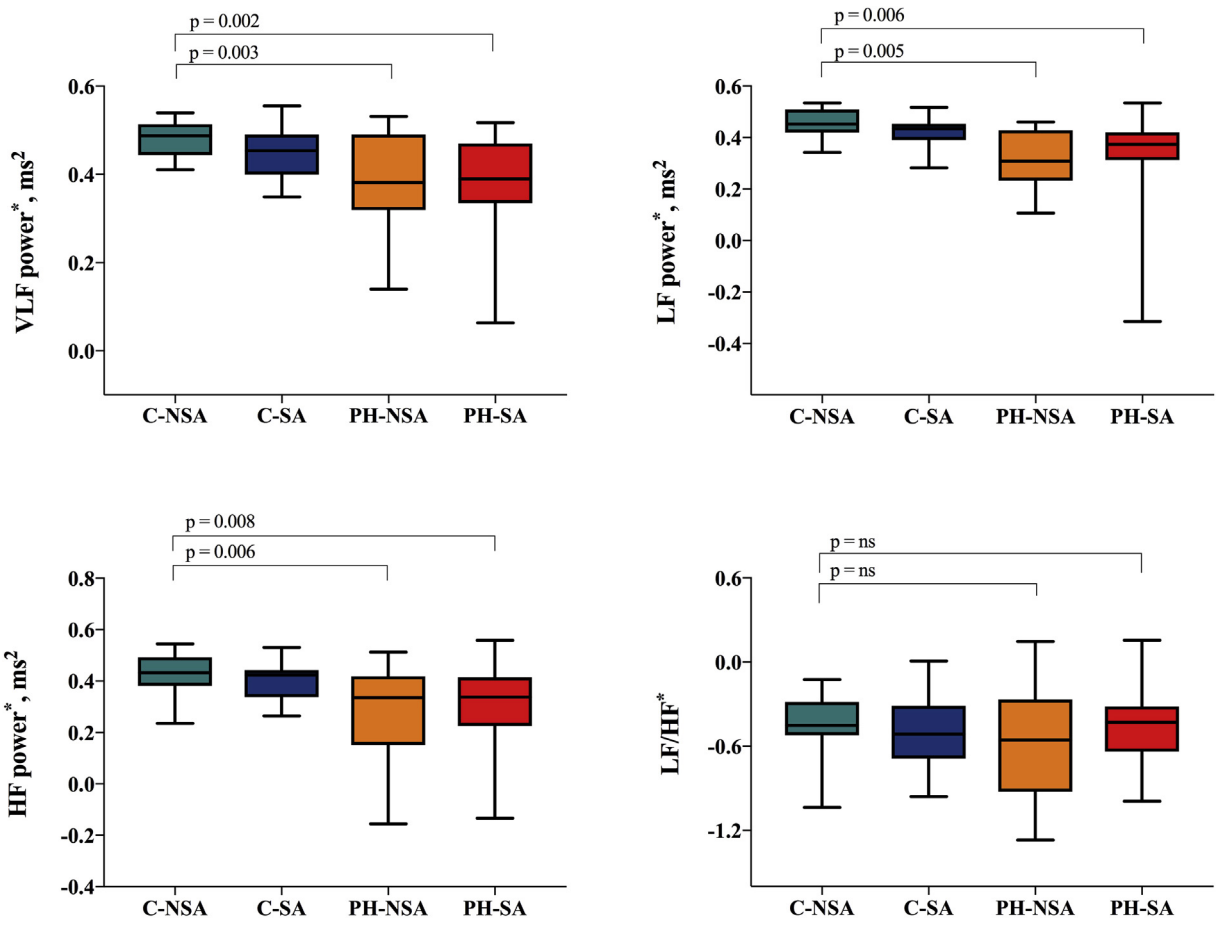

Fig. 2. Frequency domain HRV analysis. Box and whisker plots of HRV in control and PH subjects both with and without SA. Abbreviations: HRV = heart rate variability, C = Control, NSA = no sleep apnea, SA = sleep apnea, PH = pulmonary hypertension, VLF = very low frequency, LF = low frequency, HF = high frequency, LF/HF = low frequency/high frequency ratio. Box and whisker plots presented as median (p5-p95). * Log_{10}.

Table 4
Heart rate variability in control subjects with moderate to severe SA.

	Control NSA (AHI <5) N = 20	Control Moderate-Severe SA (AHI ≥15) N = 10	p-value
Mean R-R, ms	1033 (139)	905 (98)	0.015
SDNN, ms	55 (21)	43 (13)	0.122
Mean heart rate, 1/min	59 (8)	67 (7)	0.014
RMSSD, ms	46 (25)	28 (11)	0.009
pNN50, %	23 (21)	9 (13)	0.044
R-R triangular index	13 (5)	10 (3)	0.111
VLF Power, ms^2	1256 (784)	1086 (1226)	0.267
LF Power, ms^2	1036 (825)	541 (329)	0.155
HF Power, ms^2	893 (895)	326 (326)	0.039
Total Power, ms^2	3185 (2170)	1954 (1347)	0.183
LF/HF ratio	2 (1)	3 (3)	0.475

Abbreviations: NSA = no sleep apnea, SA = sleep apnea, AHI = apnea hypopnea index (events/h), Mean R-R = average of R-R intervals, SDNN = standard deviation of R-R intervals, RMSSD = root mean square of successive R-R interval differences, pNN50 = percentage of normal R-R intervals that differ by > 50 ms, R-R triangular index = number of all R-R intervals divided by the maximum of the density distribution, VLF = very low frequency, LF = low frequency, HF = high frequency.
Results are shown as Mean (SD).

(Somers et al., 1993; Trinder et al., 2001). This delicate balance is disrupted by OSA, which causes intermittent hypoxia, the generation of negative intrathoracic pressure and cortical arousals. In our study, HRV in control subjects with SA did not differ significantly from those without. This unexpected failure to demonstrate an impact of SA on the autonomic nervous system may due to the small cohort and/or the overall mild degree of OSA. The importance of OSA severity was demonstrated by the significant attenuation of parasympathetic outflow in those with moderate-severe OSA compared to those without. Furthermore, in the control subjects a significant negative correlation was shown between indices of hypoxia (CT90) and vagal modulation. This association between the severity of OSA in otherwise healthy subjects and modulation of autonomic activity in HRV is in keeping with other studies (Pan et al., 2016; Wang et al., 2008).

While autonomic nervous dysfunction has been extensively described among those with left heart failure (Danilowicz-Szymanowicz et al., 2016), less is known about autonomic modulation, in those with RV dysfunction (e.g. PH). Prior experiments in an animal model suggest the existence of a sympathetic reflex mechanism associated with pulmonary artery distention (Juratsch et al., 1977). Additionally, the increased pulmonary vascular resistance generates an increased RV afterload, resulting in RV hypertrophy, subsequent right atrial enlargement (Ciarka et al., 2010; McGowan et al., 2009), RV failure, consequent low systemic cardiac output, which in turn may activate compensatory sympathetic response but further loss of autonomic nervous system modulation, and result in concomitant myocytes injury in both ventricles (Vonk Noordegraaf et al., 2017).

Compared to control our PH subjects had impaired HRV as evidenced by significant reductions in both time (pNN50) and frequency (VLF, LF, HF) domain measures. Prior studies evaluating HRV in PAH patients were performed by assessment of daytime ECG or Holter monitor. These assessments did not take into consideration the potential influence of respiratory frequency (McGowan et al., 2009; Wensel et al., 2009; Yi et al., 2014). However, similar to the previous studies in PAH subjects

Table 5
Clinical characteristics of PH patients according to the presence of SA.

	PH-NSA N = 16	PH-SA N = 25	p-value
Age, years	39 (12)	52 (15)	0.004
Male sex, n (%)	2 (12)	8 (32)	0.265
Body mass index, kg/m^2	27 (8)	32 (6)	0.068
WHO FC I/II/III, %	12/37/50	12/52/36	0.743
PH Risk Score 1/2/3, %	31/69/0	40/56/4	0.576
Etiology of PH			0.458
Idiopathic, n (%)	7 (44)	11 (44)	
Congenital heart disease, n (%)	3 (19)	2 (8)	
Connective tissue disease, n (%)	2 (12)	8 (32)	
CTEPH, n (%)	2 (12)	1 (4)	
Other, n (%)	2 (12)	3 (12)	
On PH treatment, n (%)	5 (31)	10 (40)	0.742
Time on PH treatment, years	0 (0–1)	0 (0–3)	0.331
Epworth Sleepiness Scale	7 (5)	7 (4)	0.753
OSA/CSA (%)	0/0	76/24	
Apnea-hypopnea index, n/h	2 (1)	23 (24)	<0.001
Obstructive Index, n/h	2 (1)	16 (17)	<0.001
Central Index, n/h	0	7 (15)	0.086
Arousal Index, n/h	14 (7)	28 (21)	0.025
Mean SaO_2, %	90 (4)	90 (4)	0.999
CT90, %	45 (39)	45 (39)	0.999
ODI, n/h	8 (6)	22 (20)	0.149
Right heart catheterization			
Mean pulmonary arterial pressure, mmHg	48 (19)	46 (15)	0.671
Pulmonary vascular resistance, WU	10 (6)	8 (4)	0.355
Diastolic pressure gradient, mmHg	18 (11)	18 (11)	0.987
Right atrial pressure, mmHg	7 (3)	8 (4)	0.426
Right ventricle end diastolic pressure, mmHg	8 (5)	11 (6)	0.140
Mixed venous oxygen saturation, %	67 (7)	64 (8)	0.292
Cardiac index, L/min/m^2	2.1 (0.4)	2.1 (0.3)	0.758

Abbreviations: PH = pulmonary hypertension, NSA = no sleep apnea, SA = sleep apnea, WHO FC = World Health Organization functional class, PH Risk Stratification Score 1 = low, 2 = intermediate, 3 = high risk, CTEPH = chronic thromboembolic pulmonary hypertension, OSA = obstructive sleep apnea, CSA = central sleep apnea, SaO_2 = oxygen saturation, CT90 = cumulative percentage time at SaO_2 < 90%, ODI = Oxygen desaturation index, WU = Wood unit. Results are shown as Mean (SD) or n (%).

(Wensel et al., 2009; Yi et al., 2014), we also demonstrated decreased vagal modulation represented by lower pNN50, VLF, and HF compared to healthy subjects, and also an overall reduction in HRV as seen in R-R, R-R triangular index, LF, and total power. Additionally, the severity of RV dysfunction was an independent predictor of lower HF, as shown by multivariable models. These findings are consistent with those observed among PAH with severe RV failure (Fauchier et al., 2004). McGowan *et al*, not only showed lower LF and total power among PAH patients, but also an increase in LF/HF compared to healthy control subjects (McGowan et al., 2009), which was not observed in our study. One possible explanation is that our PH group was relatively less severe compared to the previous study, when higher mean pulmonary artery pressure (63 ± 13 mmHg) and pulmonary vascular resistance (13 ± 7 Wood units) were noted. Another possible reason for the lack of difference in LF/HF may be that our records were performed while patients were sleeping, when it is expected to have a relative blunting of sympathetic tone and a predominance of vagal activity (Bonnet and Arand, 1997).

Our hypothesis was that the presence of SA would negatively impact autonomic function as assessed by HRV by further adversely impacting RV structure and function. The possible mechanisms by which SA may impact RV structure and function include (Minic et al., 2014): 1) The exaggerated swings in the negative intrathoracic pressure cause an increase in RV transmural pressure, amplified RV venous return, increased RV preload and a leftward shift of the ventricular septum during the apneic phase in OSA and the hyperpneic phase in CSA. 2) The intermittent hypoxia elicits pulmonary vasoconstriction, oxidative stress and the generation of reactive oxygen species predisposing the vasculature to endothelial injury. 3) Elevated sympathetic activation with detrimental effects on cardio-renal hemodynamics during the day.

However, we neither observed any further modulation of HRV in PH patients with co-existent SA compared to those without, nor any correlation with markers of increasing SA severity. This suggests that PH rather than SA is the predominant driver for autonomic modulation and that SA does not exert a significant additional impact. It has previously been proposed that under extreme sympathetic nervous activity stimulation there is a plateau response as a result of receptor saturation or blockade (Malik and Camm, 1993).

Table 6
Univariate analysis of heart rate variability predictors among pulmonary hypertension patients.

Parameter	Mean difference in HF power ms^2 * (%, 95% CI)	p-value	Mean difference in LF/HF* (%, 95% CI)	p-value
Age, per year	0.001 (−0.002 to 0.005)	0.491	−0.001 (−0.008 to 0.006)	0.793
Male sex	−0.044 (−0.170 to 0.082)	0.487	0.167 (−0.081 to 0.414)	0.181
Body mass index, per kg/m^2	−0.001 (−0.009 to 0.007)	0.801	0.002 (−0.013 to 0.017)	0.803
Smoking	−0.071 (−0.424 to 0.281)	0.684	0.576 (−0.104 to 1.256)	0.095
Hypertension	0.008 (−0.130 to 0.145)	0.909	−0.026 (−0.300 to 0.249)	0.851
Epworth Sleepiness Scale, per unit	−0.1 (−0.024 to 0.005)	0.194	0.022 (−0.008 to 0.052)	0.139
Apnea-hypopnea index, per event/h	<0.001 (−0.002 to 0.003)	0.883	0.001 (−0.004 to 0.006)	0.630
Arousal index, per n/h	<0.001 (−0.003 to 0.003)	0.762	0.007 (0.002–0.012)	0.014[c]
CT90, per %	<0.001 (−0.001 to 0.001)	0.956	<0.001 (−0.003 to 0.003)	0.895
mPAP, per mmHg	−0.001 (−0.004 to 0.003)	0.700	0.002 (−0.004 to 0.009)	0.495
PVR, per Wood unit	−0.011 (−0.023 to 0.001)	0.059	0.017 (−0.016 to 0.050)	0.293
Cardiac index, per L/min/m^2	0.113 (−0.130 to 0.356)	0.343	0.183 (−0.221 to 0.588)	0.354
DPG, per mmHg	−0.006 (−0.011 to 0.001)	0.034[a]	0.008 (−0.002 to 0.018)	0.113
SvO2, %	0.002 (−0.006 to 0.011)	0.592	0.001 (−0.014 to 0.017)	0.850
Right atrial pressure, mmHg	−0.009 (−0.025 to 0.008)	0.271	0.008 (−0.032 to 0.048)	0.693
Moderate to severe RV dysfunction	−0.156 (−0.260 to − 0.051)	0.005[b]	0.121 (−0.093 to 0.335)	0.261
Pericardial effusion	− 0.145 (−0.316 to 0.027)	0.095	0.387 (0.083–0.691)	0.014[d]
B-type natriuretic peptide ≥50 ng/L	0.004 (−0.105 to 0.113)	0.942	−0.237 (−0.466 to −0.009)	0.042[e]

Abbreviations: HF = high frequency, LF/HF = low frequency/high frequency ratio, CT90 = cumulative percentage time at Oxygen saturation <90%, mPAP = mean pulmonary arterial pressure, PVR = pulmonary vascular resistance, DPG = diastolic pressure gradient, SvO2 = mixed venous Oxygen saturation.

* Log_{10}.

[a] Model $R^2 = 0.125$, Model R^2 adjusted = 0.099 (p = 0.034).

[b] Model $R^2 = 0.196$, Model R^2 adjusted = 0.175 (p = 0.005).

[c] Model $R^2 = 0.146$, Model R^2 adjusted = 0.124 (p = 0.014).

[d] Model $R^2 = 0.165$, Model R^2 adjusted = 0.140 (p = 0.014).

[e] Model $R^2 = 0.130$, Model R^2 adjusted = 0.101 (p = 0.042).

Table 7
Multivariable analysis of HF and LF/HF ratio predictors among PH patients.

HF predictors[a]	Mean difference in HF Power ms^{2c} (%, 95% CI)	p-value
Moderate to Severe RV dysfunction	−0.167 (−0.279 to −0.055)	0.005
Age, years	0.002 (−0.002 to 0.006)	0.380
LVEF, %	−0.001 (−0.007 to 0.006)	0.884
LF/HF predictors[b]	**Mean difference in LF/HF Power ms^{2c} (%, 95% CI)**	**p-value**
Arousal index, events/h	0.004 (−0.001 to 0.010)	0.136
Pericardial effusion	0.364 (−0.091 to 0.820)	0.112
BNP $\geq$ 50 ng/L	−0.136 (−0.368 to 0.097)	0.239

Abbreviations: HF = high frequency, LF = low frequency, LF/HF = low frequency/high frequency ratio, CI = confidence interval, RV = right ventricular, LVEF = left ventricular ejection fraction, BNP = B-type natriuretic peptide.
[a] Model R^2 (Model R^2 adjusted): 0.246 (0.178), p = 0.024.
[b] Model R^2 (Model R^2 adjusted): 0.304 (0.217), p = 0.031.
[c] Log_{10}.

There are multiple strengths of our study which include the relatively large sample size, the clearly defined group of PH subjects diagnosed by right heart catheterization, matched for potential confounders including age, sex, BMI and AHI to medication-free control subjects. Furthermore, HRV was confined to periods free of apnea and hypopneic events which can impinge upon HRV rhythmical oscillations (Tobaldini et al., 2013). By analyzing all data from a single sleep state we avoided potential effects of sleep state on HRV. Additionally, the restriction of analysis to stage N2 avoided the possible influence of behavioral influences on HRV, due to the relative stability and predominantly metabolic regulation of cardiovascular and respiratory systems, during this sleep stage (Tobaldini et al., 2013). Finally, high coherence between respiration and HRV ensured that respiratory frequency was not inadvertently impacting HRV parameters (Notarius and Floras, 2001; Task Force of ESC/NASPE; Tobaldini et al., 2013).

Potential limitations of our study include the small number of subjects in the SA and NSA subgroups; the few subjects with severe OSA in PH; the exclusion of patients with excessive ectopic beats, which on one hand lead to more reliable ECG segments, but on the other hand may have ruled out those with more severe PH; and the indirect measurement of autonomic modulation through HRV. However, the performance of micropolyneuropathic assessment of sympathetic nervous activity is technically difficult, and we wanted to use a feasible non-invasive marker that could be easily incorporated into clinical practice. Furthermore, neither the presence nor absence of a correlation can definitely establish causation or lack thereof.

5. Conclusions

In conclusion, our study confirmed previous findings of impaired HRV in PH compared to control subjects. We also demonstrated an impact of severe OSA on HRV in control subjects but not in PH. Therefore, we did not demonstrate greater autonomic dysregulation in those with both SA and PH. Further, larger studies should be conducted to confirm these findings and an assessment of the effect of treatment of SA in PH may identify a benefit in PH patients.

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Declarations

Author contribution statement

Carolina Gonzaga Carvalho: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Wrote the paper.

Richard Bresler: Performed the experiments; Analyzed and interpreted the data: Wrote the paper.

Ying Xuan Zhi: Analyzed and interpreted the data; Wrote the paper.

Hisham Alshaer, John T. Granton: Conceived and designed the experiments; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Clodagh M Ryan: Conceived and designed the experiments; Performed the experiments; contributed reagents, materials, analysis tools or data; Wrote the paper.

Funding statement

This work was supported by Ontario Thoracic Society - Grant in Aid.

Competing interest statement

The authors declare the following conflict of interests: Carolina Gonzaga Carvalho was supported by a grant from the CNPq – National Council for Scientific and Technological Development, Brazil. John Granton's institution has received funds to support research from Actelion Pharmaceuticals and Bayer. John Granton has served as a member of steering, data and safety monitoring, and adjudication committees for Bellerophon, Actelion, and United Therapeutics, respectively. These agencies had no role in the design, collection, analysis of data, or writing of the manuscript. Richard Bresler, Ying Xuan Zhi, Hisham Alshaer, Clodagh Ryan have nothing to disclose.

Additional information

No additional information is available for this paper.

Acknowledgements

The authors want to thank Richard Hummel for his assistance in data processing.

References

Berry, R.B., Budhiraja, R., Gottlieb, D.J., Gozal, D., Iber, C., Kapur, V.K., Marcus, C.L., Mehra, R., Parthasarathy, S., Quan, S.F., Redline, S., Strohl, K.P., Davidson Ward, S.L., Tangredi, M.M., 2012. Rules for scoring respiratory events in sleep: update of the 2007 AASM manual for the scoring of sleep and associated events. Deliberations of the sleep apnea definitions Task Force of the American academy of sleep medicine. J. Clin. Sleep Med.: JCSM : Off. Publ. Am. Acad. Sleep Med. 8, 597–619.

Bonnet, M.H., Arand, D.L., 1997. Heart rate variability: sleep stage, time of night, and arousal influences. Electroencephalogr. Clin. Neurophysiol. 102, 390–396.

Bradley, T.D., Floras, J.S., 2003. Sleep apnea and heart failure: Part I: obstructive sleep apnea. Circulation 107, 1671–1678.

Ciarka, A., Doan, V., Velez-Roa, S., Naeije, R., van de Borne, P., 2010. Prognostic significance of sympathetic nervous system activation in pulmonary arterial hypertension. Am. J. Respir. Crit. Care Med. 181, 1269–1275.

D'Alonzo, G.E., Barst, R.J., Ayres, S.M., Bergofsky, E.H., Brundage, B.H., Detre, K.M., Fishman, A.P., Goldring, R.M., Groves, B.M., Kernis, J.T., et al., 1991. Survival in patients with primary pulmonary hypertension. Results from a national prospective registry. Ann. Intern. Med. 115, 343–349.

Danilowicz-Szymanowicz, L., Suchecka, J., Niemirycz-Makurat, A., Rozwadowska, K., Raczak, G., 2016. Autonomic predictors of hospitalization due to heart failure decompensation in patients with left ventricular systolic dysfunction. PLoS One 11, e0152372.

Fauchier, L., Babuty, D., Melin, A., Bonnet, P., Cosnay, P., Paul Fauchier, J., 2004. Heart rate variability in severe right or left heart failure: the role of pulmonary hypertension and resistances. Eur. J. Heart Fail. 6, 181–185.

Galie, N., Humbert, M., Vachiery, J.L., Gibbs, S., Lang, I., Torbicki, A., Simonneau, G., Peacock, A., Vonk Noordegraaf, A., Beghetti, M., Ghofrani, A., Gomez Sanchez, M.A., Hansmann, G., Klepetko, W., Lancellotti, P., Matucci, M., McDonagh, T., Pierard, L.A., Trindade, P.T., Zompatori, M., Hoeper, M., Aboyans, V., Vaz Carneiro, A., Achenbach, S., Agewall, S., Allanore, Y., Asteggiano, R., Paolo Badano, L., Albert Barbera, J., Bouvaist, H., Bueno, H., Byrne, R.A., Carerj, S., Castro, G., Erol, C., Falk, V., Funck-Brentano, C., Gorenflo, M., Granton, J., Iung, B., Kiely, D.G., Kirchhof, P., Kjellstrom, B., Landmesser, U., Lekakis, J., Lionis, C., Lip, G.Y., Orfanos, S.E., Park, M.H., Piepoli, M.F., Ponikowski, P., Revel, M.P., Rigau, D., Rosenkranz, S., Voller, H., Luis Zamorano, J., 2016. 2015 ESC/ERS guidelines for the diagnosis and treatment of pulmonary hypertension: the joint Task Force for the

diagnosis and treatment of pulmonary hypertension of the european society of cardiology (ESC) and the european respiratory society (ERS): endorsed by: association for european paediatric and congenital cardiology (AEPC), international society for heart and lung transplantation (ISHLT). Eur. Heart J. 37, 67–119.

Hillebrand, S., Gast, K.B., de Mutsert, R., Swenne, C.A., Jukema, J.W., Middeldorp, S., Rosendaal, F.R., Dekkers, O.M., 2013. Heart rate variability and first cardiovascular event in populations without known cardiovascular disease: meta-analysis and dose-response meta-regression. Europace : Eur. Pacing, Arrhythm, Cardiac Electrophysiol.: J. Working Groups Cardiac Pacing, Arrhythm., Cardiac Cell. Electrophysiol. Eur. Soc. Cardiol. 15, 742–749.

Iber, C., Ancoli-Israel, S., Chesson, A., 2007. The AASM Manual for the Scoring of Sleep and Associated Events. American Academy of Sleep Medicine.

Johns, M.W., 1991. A new method for measuring daytime sleepiness: the Epworth sleepiness scale. Sleep 14, 540–545.

Juratsch, C.E., Jengo, J.A., Laks, M.M., 1977. Role of the autonomic nervous system and pulmonary artery receptors in production of experimental pulmonary hypertension. Chest 71, 265–269.

Klein, A.L., Abbara, S., Agler, D.A., Appleton, C.P., Asher, C.R., Hoit, B., Hung, J., Garcia, M.J., Kronzon, I., Oh, J.K., Rodriguez, E.R., Schaff, H.V., Schoenhagen, P., Tan, C.D., White, R.D., 2013. American society of echocardiography clinical recommendations for multimodality cardiovascular imaging of patients with pericardial disease: endorsed by the society for cardiovascular magnetic resonance and society of cardiovascular computed tomography. J. Am. Soc. Echocardiogr. : Off. Publ. Am. Soc. Echocardiogr. 26, 965-1012 e1015.

Lahiri, M.K., Kannankeril, P.J., Goldberger, J.J., 2008. Assessment of autonomic function in cardiovascular disease: physiological basis and prognostic implications. J. Am. Coll. Cardiol. 51, 1725–1733.

Leuchte, H.H., Ten Freyhaus, H., Gall, H., Halank, M., Hoeper, M.M., Kaemmerer, H., Kahler, C., Riemekasten, G., Ulrich, S., Schwaiblmair, M., Ewert, R., 2018. Risk stratification strategy and assessment of disease progression in patients with pulmonary arterial hypertension: updated recommendations from the Cologne Consensus Conference 2018. Int. J. Cardiol. 272S, 20–29.

Maheshwari, A., Norby, F.L., Soliman, E.Z., Adabag, S., Whitsel, E.A., Alonso, A., Chen, L.Y., 2016. Low heart rate variability in a 2-minute electrocardiogram recording is associated with an increased risk of sudden cardiac death in the general population: the atherosclerosis risk in communities study. PLoS One 11, e0161648.

Malik, M., Camm, A.J., 1993. Components of heart rate variability–what they really mean and what we really measure. Am. J. Cardiol. 72, 821–822.

McGowan, C.L., Swiston, J.S., Notarius, C.F., Mak, S., Morris, B.L., Picton, P.E., Granton, J.T., Floras, J.S., 2009. Discordance between microneurographic and heart-rate spectral indices of sympathetic activity in pulmonary arterial hypertension. Heart (British Cardiac Soc.) 95, 754–758.

Minic, M., Granton, J.T., Ryan, C.M., 2014. Sleep disordered breathing in group 1 pulmonary arterial hypertension. J. Clin. Sleep Med.: JCSM : Off. Publ. Am. Acad. Sleep Med. 10, 277–283.

Narkiewicz, K., Montano, N., Cogliati, C., van de Borne, P.J., Dyken, M.E., Somers, V.K., 1998. Altered cardiovascular variability in obstructive sleep apnea. Circulation 98, 1071–1077.

Notarius, C.F., Floras, J.S., 2001. Limitations of the use of spectral analysis of heart rate variability for the estimation of cardiac sympathetic activity in heart failure. Europace : Eur. Pacing, Arrhythm, Cardiac Electrophysiol.: J. Working Groups Cardiac Pacing, Arrhythm., Cardiac Cell. Electrophysiol. Eur. Soc. Cardiol. 3, 29–38.

Pagani, M., Lombardi, F., Guzzetti, S., Rimoldi, O., Furlan, R., Pizzinelli, P., Sandrone, G., Malfatto, G., Dell'Orto, S., Piccaluga, E., et al., 1986. Power spectral analysis of heart rate and arterial pressure variabilities as a marker of sympatho-vagal interaction in man and conscious dog. Circ. Res. 59, 178–193.

Pan, W.Y., Su, M.C., Wu, H.T., Su, T.J., Lin, M.C., Sun, C.K., 2016. Multiscale entropic assessment of autonomic dysfunction in patients with obstructive sleep apnea and therapeutic impact of continuous positive airway pressure treatment. Sleep Med. 20, 12–17.

Shaffer, F., Ginsberg, J.P., 2017. An overview of heart rate variability metrics and norms. Front. Public Health 5, 258.

Sitbon, O., Sattler, C., Bertoletti, L., Savale, L., Cottin, V., Jais, X., De Groote, P., Chaouat, A., Chabannes, C., Bergot, E., Bouvaist, H., Dauphin, C., Bourdin, A., Bauer, F., Montani, D., Humbert, M., Simonneau, G., 2016. Initial dual oral combination therapy in pulmonary arterial hypertension. Eur. Respir. J. 47, 1727–1736.

Somers, V.K., Dyken, M.E., Mark, A.L., Abboud, F.M., 1993. Sympathetic-nerve activity during sleep in normal subjects. N. Engl. J. Med. 328, 303–307.

Somers, V.K., Dyken, M.E., Clary, M.P., Abboud, F.M., 1995. Sympathetic neural mechanisms in obstructive sleep apnea. J. Clin. Investig. 96, 1897–1904.

Stein, P.K., Pu, Y., 2012. Heart rate variability, sleep and sleep disorders. Sleep Med. Rev. 16, 47–66.

Tarvainen, M.P., Niskanen, J.P., Lipponen, J.A., Ranta-Aho, P.O., Karjalainen, P.A., 2014. Kubios HRV–heart rate variability analysis software. Comput. Methods Progr. Biomed. 113, 210–220.

Task Force of ESC/NASPE, 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. Circulation 93, 1043–1065.

Tobaldini, E., Nobili, L., Strada, S., Casali, K.R., Braghiroli, A., Montano, N., 2013. Heart rate variability in normal and pathological sleep. Front. Physiol. 4, 294.

Trinder, J., Kleiman, J., Carrington, M., Smith, S., Breen, S., Tan, N., Kim, Y., 2001. Autonomic activity during human sleep as a function of time and sleep stage. Journal of Sleep Res 10, 253–264.

Tulppo, M.P., Kiviniemi, A.M., Hautala, A.J., Kallio, M., Seppanen, T., Makikallio, T.H., Huikuri, H.V., 2005. Physiological background of the loss of fractal heart rate dynamics. Circulation 112, 314–319.

Velez-Roa, S., Ciarka, A., Najem, B., Vachiery, J.L., Naeije, R., van de Borne, P., 2004. Increased sympathetic nerve activity in pulmonary artery hypertension. Circulation 110, 1308–1312.

Vonk Noordegraaf, A., Westerhof, B.E., Westerhof, N., 2017. The relationship between the right ventricle and its load in pulmonary hypertension. J. Am. Coll. Cardiol. 69, 236–243.

Wang, W., Tretriluxana, S., Redline, S., Surovec, S., Gottlieb, D.J., Khoo, M.C., 2008. Association of cardiac autonomic function measures with severity of sleep-disordered breathing in a community-based sample. Journal of sleep research 17, 251–262.

Wensel, R., Jilek, C., Dorr, M., Francis, D.P., Stadler, H., Lange, T., Blumberg, F., Opitz, C., Pfeifer, M., Ewert, R., 2009. Impaired cardiac autonomic control relates to disease severity in pulmonary hypertension. Eur. Respir. J. 34, 895–901.

Wiklund, U., Olofsson, B.O., Franklin, K., Blom, H., Bjerle, P., Niklasson, U., 2000. Autonomic cardiovascular regulation in patients with obstructive sleep apnoea: a study based on spectral analysis of heart rate variability. Clin. Physiol. 20, 234–241.

Yi, H.T., Hsieh, Y.C., Wu, T.J., Huang, J.L., Lin, W.W., Liang, K.W., Su, C.S., Tsai, W.J., Wang, K.Y., 2014. Heart rate variability parameters and ventricular arrhythmia correlate with pulmonary arterial pressure in adult patients with idiopathic pulmonary arterial hypertension. Heart Lung: J. Crit. Care 43, 534–540.