**SUPPLEMENTAL MATERIAL**

**Baroreceptor Reflex Function: Impact on Perioperative Outcomes and Chronic Pain**

Heberto Suarez-Roca, MD, PhD1, Rebecca Y. Klinger, MD, MS2, Mihai V. Podgoreanu, MD2, Ru-Rong Ji, PhD1, Martin I. Sigurdsson, MD, PhD2, Nathan Waldron, MD2, Joseph P. Mathew, MD, MHSc, MBA2, and William Maixner, DDS, PhD1

1 Center for Translational Pain Medicine, Department of Anesthesiology, Duke University, Durham, NC

2 Division of Cardiothoracic Anesthesia and Critical Care Medicine, Department of Anesthesiology, Duke University, Durham, NC

**Measurement of baroreflex sensitivity**

One way of assessing baroreceptor function is through the measurement of baroreceptor sensitivity (BRS). BRS is typically defined by the relationship between the change in arterial pressure (AP) and the associated effect on the inter-beat interval.1,2 Most procedures used to evaluate BRS measure the relationship between the change in heart rate, estimated by the R-R interval (RRI) in the electrocardiogram (ECG), as a function of the change in systolic AP (SAP). The gold-standard procedure for assessing BRS is to measure the ratio of change in heart rate to the change in SAP in responses to the intravenous administration of a low dose of a vasopressor agent (e.g., phenylephrine).3-8 Another common technique is to determine the ratio of change in heart rate to change in AP in response to controlled neck suction, which produces a controlled distention of the carotid sinus.7 Techniques used to evaluate cardiopulmonary baroreflexes include measuring heart rate and blood pressure responses to lower body negative or positive pressure, extremity congesting cuffs, controlled hemorrhage, leg elevation, upright tilting, administration of a vasodilator (e.g., nitroglycerine), deep breathing, hand grip stress, and the Valsalva maneuver as reviewed elsewhere.9 Given the evidence that baroreceptors are not only activated by acute changes in arterial pressure but also in response to small natural continuous variations in blood pressure, non-invasive methods have been developed that assess BRS using the spontaneous covariation of heart rate and blood pressure (called ‘spontaneous BRS’).6,10-18

Current methods estimate BRS using either time- or frequency-domain analyses, which attempt to account for the complex physiology of baroreflex regulation, separation of baroreflex signals from background noise under a variety of physiological and pathological conditions have been developed.7,14,19-30In this regard, a delay of usually 1 to 4 beats (or up to 2000-3000 ms) is usually applied to SAP with respect to RRI to obtain the best regression line fit between the SAP and RRI values resulting in a more accurate BRS value, especially at a mean RRI < 800 ms, (i.e., heart rate > 75 bpm).6,31 This is useful for measurements done under pathological conditions, in which the delay in the baroreflex loop is altered (e.g., increased in Type 1 diabetes mellitus) or the baseline heart rate is high, such as hypertension, chronic renal failure, post-myocardial infarction, or heart failure.17

On the other hand, frequency-domain based analyses of SAP and RRI reveal the spectra of specific rhythms that correlate with sympathetic and parasympathetic control of the cardiovascular system. In particular, low-frequency oscillations in the range of < 0.15 Hz within the AP signal have been correlated with sympathetic activity in humans.32,33 In contrast, high-frequency oscillations of > 0.2 Hz and typically seen with heart rate or RRI time series are correlated with parasympathetic activity.34-36 Dynamically assessing or controlling respiratory rate is important, as it influences high frequency oscillations. To assure that the oscillations in the RRI and SAP signals are coherent (i.e., synchronized or correlated with each other), spectral densities obtained from frequency-domain analyses can be integrated over the segment of the low frequency band (LF; 0.04 -0.15 Hz) that exceeds the selected level of coherence (usually 0.5) to exclude oscillations that are not synchronized.4,14,19,37-39 In addition, to ascertaining that SAP and RRI coherent oscillations have an appropriate phase relationship, spectral density is integrated over the segment of the LF band where the phase difference is negative (i.e., SAP changes preceded those expected in the RRI).4 Since a phase difference defines only the relationship between the two signals but not which signal leads to the other (i.e., cause-effect relation between SAP and RRI oscillations), the linear modeling of the SAP as the input signal and the RRI as the output has been developed using different algorithms, such as a transfer function.20-23 Finally, in addition to the expected baroreflex influence of SAP on RRI, the non-baroreflex influence of RRI on SAP can be taken into account for BRS estimation by separately computing the unidirectional causal coherence and gain for SAP on RRI with the bivariate autoregressive model. 24,25 **Table 1S** shows the advantages and limitations of each method. While the results of computing BRS based on either time or frequency domain are not directly comparable to each other a combination of methods should be considered when studying the relationship between BRS and autonomic function. **Figure 1S** shows the estimation of BRS using the time-domain based sequence method.

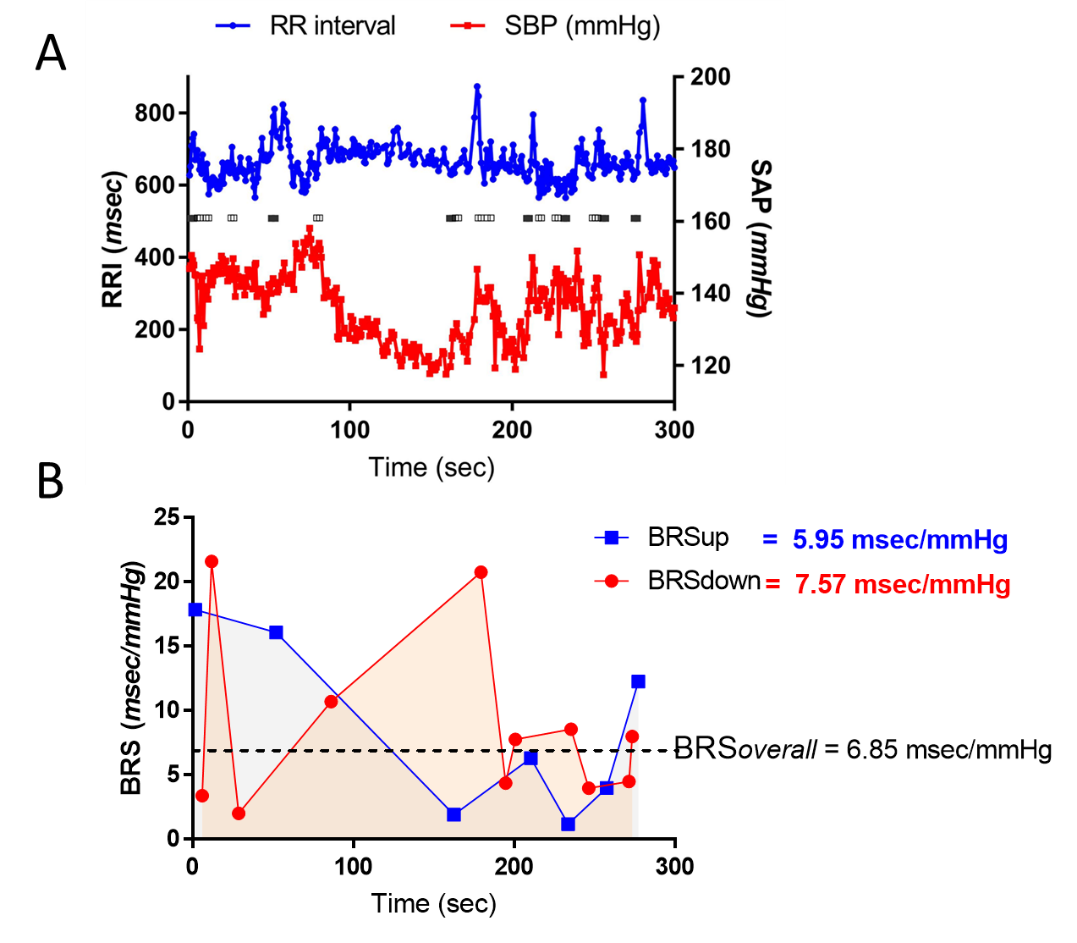
BRS thresholds for defining normal and abnormal BRS have been proposed by the Autonomic Tone and Reflexes After Myocardial Infarction Study.7,40 In general, normal BRS is defined as > 6 ms/mmHg. BRS dysfunction can be classified as moderate (3-6 ms/mmHg) and severe (< 3 ms/mmHg). A recent study of patients with chronic obstructive pulmonary disease verified that the healthy volunteer cohort (average age 67 years) had a spontaneous BRS value of 7.8 ± 4.9 ms/mmHg.41 In contrast, patients with pulmonary disease had a mean BRS of 3.4 ± 2.6 ms/mmHg, and those with a history of myocardial dysfunction had a mean BRS of 3.1 ± 2.1 ms/mmHg. Highly relevant to the perioperative patient population, a technique to assess BRS while accounting for increased arterial stiffness with age has also been established.42

While a recent critical appraisal of the reliability and clinical relevance of spontaneous BRS measurement indicates that it is generally a powerful tool for both prognostication of cardiovascular disease and treatment evaluation for both cardiovascular and non-cardiovascular diseases, several methodologic issues can hamper the reliable measurement of BRS.43 Factors affecting the evaluation of spontaneous BRS include non-sinus rhythm, significant ectopy, and the requirement of recorded time series of R-R interval and arterial blood pressure to satisfy BRS estimation algorithms.43 Concerning reliability, within-subject variability in spontaneous BRS is high44; however, spontaneous BRS remains a more reliable measure of cardiac autonomic control than other non-invasive measures, such as heart rate variability45. The cross-correlation of the baroreflex sensitivity method described by Westerhof et al. significantly increases both the measurability and reliability of BRS estimation.46

**TABLE 1S: Comparison of major methods used to estimate baroreflex sensitivity**

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| --- | --- | --- | --- |
| **Methods** | **Procedure/Computation** | **Applications/Features** | **Caveats/Limitations** |
| **Pharmacological methods***3-7* | The administration of vasoactive drugs elicits acute changes in AP  EKG and SAP waveforms are recorded simultaneously, then beat-to-beat SAP and calculated RRI are plotted.  BRS is defined as the slope of a regression line fitted on a scatter plot of SAP and RRI corresponding pairs of values. | Estimates the vagal component of BRS with vasopressor drugs (e.g., phenylephrine) or the sympathetic component with vasorelaxing drugs (e.g. nitroglycerin, amyl nitrite).7  Is the gold standard for studies in which low BRS measured from the pharmacological method predicts risk of mortality.7,8 | Difficult to select the exact moments at which SAP and RRI change following the administration of the vasoactive drug.4  Phenylephrine method assesses only part of the baroreflex arc: maximal baroreceptor recruitment bylarge changes in SAP (phasic component) but not the minimal activation from natural SAP oscillations(tonic component).6  Vasoactive drugs may directly affect transduction properties of baroreceptors, and besides, central components and SA node, assessing the full capacity of the system – not only baroreceptors – to elicit a vagal response.7  Administration of vasoactive might be a risk for some participants.7 |
| **Functional methods**7 | Acute changes in AP are elicited by physiological maneuvers (Valsalva phase 4 or pressure-vacuum on the neck region).  BRS is estimated by slope measurement similar to the pharmacological methods. | Is an alternative when the administration of vasoactive drugs is not possible. | Valsalva maneuver should not be used in patients with advanced ventricular dysfunction.7 |
| **Sequence method** *6,11-16* | A time-domain based analysis of a time series that identifies sequences of increase in SAP and RRI (*Up* sequence) or decrease in SAP and RRI (*Down* sequence). Sequence linear slopes are then estimated and averaged.  Selection of sequences ≥ 3 beats is based on empirically setting SAP, RRI, and correlation coefficient thresholds.  Breathing is either paced with a metronome during measurement or adjusted for the respiratory rate by estimating it from the peak amplitude of high frequency HRV. | Estimates the spontaneous BRS from naturally occurring fluctuations of SAP. It does not require the use of vasoactive pharmacological agents.  Allows to assess asymmetric impairment of bradycardic and tachycardic baroreflexes as seen in diabetics.17  Its long-term reliability is higher than cross-spectral analysis for some pathological conditions, e.g., chronic heart failure.14  Underestimates and overestimated BRS compared with phenylephrine and nitroprusside slope methods, respectively.18 | Naturally occurring oscillations are frequently caused by breathing patterns. So, derived BRS may depend on breathing depth and frequency. Yet, this influence may not be of significance.16  Number of sequences could be reduced or absent in a 5-minute period in patients with impaired BRS (diabetics, hypertensives).6  Lower sensitivity than phenylephrine method to detect differences if a residual BRS is sufficient to control small SAP oscillations of ± 5 mmHg.8  BRS values are differentially affected by sequence selection criteria in healthy subjects vs. patients (e.g., chronic heart failure).6 |
| **Cross-spectral methods7,14,19-25** | A frequency-domain based analysis in which time-localized RRI and SAP signals (times series) are converted to a frequency sequence. Algorithms then compute SAP and RRI power spectra (e.g., fast Fourier transform, transfer function, autoregressive).  Assumes that SAP is mainly associated with the integral of the spectrum over 0.04 -0.15 Hz (LF band).  BRS is generally estimated as either the square root of the ratio between RRI and SAP spectral powers (autoregressive method), or the average of the transfer function modulus (the gain) between SAP and RRI, over the LF band frequencies with coherence ≥0.5.  Non‐baroreflex interactions (influences of RRI on SAP) can be removed by separately computing the unidirectional causal coherence and gain for SAP on RRI (bivariate autoregressive model).24,25 | Estimates the spontaneous BRS from naturally occurring fluctuations of BP.  Ascertains that both SAP and RRI signals have the same frequency components and phase relationships can be taken into consideration.  Using transfer function, allows estimation of BRS with clinical accuracy, under conditions of low signal-to-noise ratio and/or impaired baroreflex gain associated with markedly reduced coherence.  Allows for reducing the influence of non-baroreflex components on BRS estimates.24,26  Reproducibility of results within one-week interval.14 | Reliability of BRS estimates decreases in patients with severe ectopic activity due to the need of ≥ 3-min recordings for acceptable accuracy.7  Difficult to use in pathological conditions with low coherence (e.g., chronic heart failure).*39* Yet, this has been addressed by using the transfer function.7  Some algorithms used to calculate BRS (e.g., autoregressive methods, transfer function) ignore non-baroreflex influences that affect SAP and RRI, such as respiration and central mechanisms, and that as a result modify components in the LF band and BRS estimations.*22,24* Yet, this has been addressed by using the bivariate autoregressive model.24,25 |
| **Autoregressive moving average analysis (ARMA) models**27,28 | A linear model with one output signal: RRI (autoregressive component), two input signals: SAP and instantaneous lung volume (moving average component), and two uncorrelated sources of noise from SAP and respiration.  Breathing guided by a randomly spaced sequence of computer-generated tones since the broadband input is thought to be required for a solution with the model. | Estimates BRS without the influence of respiration and the noise component by enabling a separate evaluation of causal coherence and gains in two directions: from SAP to RRI and from RRI to SAP.  Is highly reproducible within one-week period and significantly correlated with BRS derived from the sequence method and slope method with hypotensive drug (but not with hypertensive drugs).28 | Some individuals are unable to follow  the tones because of cognitive deficit and/or pulmonary disease. Yet, meaningful estimation could be obtained under spontaneous breathing.28  Does not provide the traditional measures of threshold or saturation of the reflex and does not define the exact position of the operating set point on SAP curve.  Is not adequate if the real physiological system is non-linear. |
| **Complex demodulation**29,30 | A non-linear model that provides time-dependent changes in amplitude and phase of a particular frequency component as a function of time.  BRS is estimated by the instantaneous amplitude of complex-demodulated RRI divided by the instantaneous amplitude of complex-demodulated SAP.  Produces a frequency range of 0.04 to 0.14 Hz (LF band) when applied to RRI and SAP signals a center frequency of 0.09 Hz with cutoff frequency of low-pass filter of 0.05 Hz. | Allows modeling the non-lineal features of the arterial baroreflex, which naturally exhibits a sigmoidal stimulus (SAP)–response (RRI) curve.  Has a high temporal resolution of about 15 s, which is better than any seen in spectral method.  Useful for ambulatory, not very clean, recordings since stationary condition for the time series is not required.  BRS values are quite stable.  Results are equivalent to those derived by cross-spectral and sequential analysis. | Has no control over whether the signals are coherent concerning each other or whether a change in the SAP signal precedes a change in the RR interval. |

**Note:** ECG: electrocardiogram, AP: arterial blood pressure, BRS: baroreflex sensitivity, SAP: systolic arterial pressure, RRI: R-R interval, LF: low-frequency, HRV: heart rate variability



**FIGURE 1S:** Estimation of BRS using the sequence method.EKG and SAP waveforms are recorded simultaneously, then beat-to-beat SAP and the calculated RRI are plotted against time course.A.SAP and RR interval signals and the baroreflex sequences acquired using the sequence method.Closed squares indicate UP sequences. Open squares indicate DOWN sequences. Sequence selection criteria: SAP > 0.5 mmHg, RRI > 1 ms, sequence > 3, a significant correlation coefficient (r > 0.9). Note that the significant sequences cluster in segments where SAP and RRI signals apparently oscillate more coherently (in this case, at the start and the end of this recording.). B. Within-subject variability of the BRS. Mean UP, DOWN, and overall BRS calculated from the sequences shown in the previous figure.

**REFERENCES**

1. Parati G, Di Rienzo M, Mancia G: How to measure baroreflex sensitivity: from the cardiovascular laboratory to daily life. J Hypertens 2000; 18: 7-19

2. Reyes Del Paso GA, Gonzales MI, Hernandez JA: Comparison of baroreceptor cardiac reflex sensivity estimates from intersystolic and ECG R-R intervals. Psychophysiology 2010; 47: 1102-1108

3. Smyth HS, Sleight P, Pickering GW: Reflex regulation of arterial pressure during sleep in man. A quantitative method of assessing baroreflex sensitivity. Circ Res 1969; 24: 109-21

4. Kuusela TA: Methodological Aspects of Baroreflex Sensitivity Analysis, Heart Rate Variability (HRV) Signal Analysis: Clinical Applications

Edited by Kamath MW, M.; Upton A., CRC Press 2012, pp 43–58

5. Borovikova LV, Ivanova S, Zhang M, Yang H, Botchkina GI, Watkins LR, Wang H, Abumrad N, Eaton JW, Tracey KJ: Vagus nerve stimulation attenuates the systemic inflammatory response to endotoxin. Nature 2000; 405: 458-62

6. Davies LC, Francis DP, Scott AC, Ponikowski P, Piepoli M, Coats AJ: Effect of altering conditions of the sequence method on baroreflex sensitivity. J Hypertens 2001; 19: 1279-87

7. La Rovere MT, Pinna GD, Raczak G: Baroreflex sensitivity: measurement and clinical implications. Ann Noninvasive Electrocardiol 2008; 13: 191-207

8. Watkins LL, Fainman C, Dimsdale J, Ziegler MG: Assessment of baroreflex control from beat-to-beat blood pressure and heart rate changes: a validation study. Psychophysiology 1995; 32: 411-4

9. Mark AL, Mancia G: Cardiopulmonary baroreflexes in humans. Compr Physiol 2011; Supl 8: 795-813

10. Duschek S, Dietel A, Schandry R, Reyes Del Paso GA: Increased baroreflex sensitivity and reduced cardiovascular reactivity in individuals with chronic low blood pressure. Hypertens Res 2008; 31: 1873-8

11. Parati G, Di Rienzo M, Bertinieri G, Pomidossi G, Casadei R, Groppelli A, Pedotti A, Zanchetti A, Mancia G: Evaluation of the baroreceptor-heart rate reflex by 24-hour intra-arterial blood pressure monitoring in humans. Hypertension 1988; 12: 214-22

12. Hughson RL, Quintin L, Annat G, Yamamoto Y, Gharib C: Spontaneous baroreflex by sequence and power spectral methods in humans. Clin Physiol 1993; 13: 663-76

13. Parlow J, Viale JP, Annat G, Hughson R, Quintin L: Spontaneous cardiac baroreflex in humans. Comparison with drug-induced responses. Hypertension 1995; 25: 1058-68

14. Herpin D, Ragot S: Mid- and long-term reproducibility of noninvasive measurements of spontaneous arterial baroreflex sensitivity in healthy volunteers. Am J Hypertens 1997; 10: 790-7

15. Parati G, Di Rienzo M, Mancia G: How to measure baroreflex sensitivity: from the cardiovascular laboratory to daily life. J Hypertens 2000; 18: 7-19

16. Parodi G, Bellandi B, Valenti R, Memisha G, Giuliani G, Velluzzi S, Migliorini A, Carrabba N, Antoniucci D: Heart rate as an independent prognostic risk factor in patients with acute myocardial infarction undergoing primary percutaneous coronary intervention. Atherosclerosis 2010; 211: 255-9

17. Javorka M, Lazarova Z, Tonhajzerova I, Turianikova Z, Honzikova N, Fiser B, Javorka K, Baumert M: Baroreflex analysis in diabetes mellitus: linear and nonlinear approaches. Med Biol Eng Comput 2011; 49: 279-88

18. Johnson P, Shore A, Potter J, Panerai R, James M: Baroreflex sensitivity measured by spectral and sequence analysis in cerebrovascular disease : methodological considerations. Clin Auton Res 2006; 16: 270-5

19. Robbe HW, Mulder LJ, Ruddel H, Langewitz WA, Veldman JB, Mulder G: Assessment of baroreceptor reflex sensitivity by means of spectral analysis. Hypertension 1987; 10: 538-43

20. Porta A, Baselli G, Rimoldi O, Malliani A, Pagani M: Assessing baroreflex gain from spontaneous variability in conscious dogs: role of causality and respiration. Am J Physiol Heart Circ Physiol 2000; 279: H2558-67

21. Pinna GD, Maestri R: New criteria for estimating baroreflex sensitivity using the transfer function method. Med Biol Eng Comput 2002; 40: 79-84

22. Pinna GD, Maestri R, Raczak G, La Rovere MT: Measuring baroreflex sensitivity from the gain function between arterial pressure and heart period. Clin Sci (Lond) 2002; 103: 81-8

23. Porta A, Bari V, Bassani T, Marchi A, Pistuddi V, Ranucci M: Model-based causal closed-loop approach to the estimate of baroreflex sensitivity during propofol anesthesia in patients undergoing coronary artery bypass graft. J Appl Physiol (1985) 2013; 115: 1032-42

24. Ondrusova K, Svacinova J, Javorka M, Novak J, Novakova M, Novakova Z: Impaired Baroreflex Function during Orthostatic Challenge in Patients after Spinal Cord Injury. J Neurotrauma 2017

25. Svacinova J, Javorka M, Novakova Z, Zavodna E, Czippelova B, Honzikova N: Development of causal interactions between systolic blood pressure and inter-beat intervals in adolescents. Physiol Res 2015; 64: 821-9

26. Porta A, Furlan R, Rimoldi O, Pagani M, Malliani A, van de Borne P: Quantifying the strength of the linear causal coupling in closed loop interacting cardiovascular variability signals. Biol Cybern 2002; 86: 241-51

27. Patton DJ, Triedman JK, Perrott MH, Vidian AA, Saul JP: Baroreflex gain: characterization using autoregressive moving average analysis. Am J Physiol 1996; 270: H1240-9

28. O'Leary DD, Lin DC, Hughson RL: Determination of baroreflex gain using auto-regressive moving-average analysis during spontaneous breathing. Clin Physiol 1999; 19: 369-77

29. Hayano J, Taylor JA, Yamada A, Mukai S, Hori R, Asakawa T, Yokoyama K, Watanabe Y, Takata K, Fujinami T: Continuous assessment of hemodynamic control by complex demodulation of cardiovascular variability. Am J Physiol 1993; 264: H1229-38

30. Kim SY, Euler DE: Baroreflex sensitivity assessed by complex demodulation of cardiovascular variability. Hypertension 1997; 29: 1119-25

31. Martinez-Garcia P, Lerma C, Infante O: Baroreflex sensitivity estimation by the sequence method with delayed signals. Clin Auton Res 2012; 22: 289-97

32. Grasso R, Schena F, Gulli G, Cevese A: Does low-frequency variability of heart period reflect a specific parasympathetic mechanism? J Auton Nerv Syst 1997; 63: 30-8

33. Jardine DL, Ikram H, Frampton CM, Frethey R, Bennett SI, Crozier IG: Autonomic control of vasovagal syncope. Am J Physiol 1998; 274: H2110-5

34. Eckberg DL: Physiological basis for human autonomic rhythms. Ann Med 2000; 32: 341-9

35. Eckberg DL: Human sinus arrhythmia as an index of vagal cardiac outflow. J Appl Physiol Respir Environ Exerc Physiol 1983; 54: 961-6

36. Saul JP, Berger RD, Albrecht P, Stein SP, Chen MH, Cohen RJ: Transfer function analysis of the circulation: unique insights into cardiovascular regulation. Am J Physiol 1991; 261: H1231-45

37. de Boer RW, Karemaker JM, Strackee J: Relationships between short-term blood-pressure fluctuations and heart-rate variability in resting subjects. I: A spectral analysis approach. Med Biol Eng Comput 1985; 23: 352-8

38. Airaksinen KE, Tahvanainen KU, Kuusela TA, Huikuri HV, Niemela MJ, Karjalainen P, Eckberg DL: Cross spectral analysis in assessment of baroreflex gain in patients with coronary artery disease. Ann Noninvasive Electrocardiol 1997; 2: 229-35

39. Colombo R, Mazzuero G, Spinatonda G, Lanfranchi P, Giannuzzi P, Ponikowski P, Coats AJ, Minuco G: Comparison between spectral analysis and the phenylephrine method for the assessment of baroreflex sensitivity in chronic heart failure. Clin Sci (Lond) 1999; 97: 503-13

40. La Rovere MT, Pinna GD, Hohnloser SH, Marcus FI, Mortara A, Nohara R, Bigger JT, Jr., Camm AJ, Schwartz PJ, Tone AIA, Reflexes After Myocardial I: Baroreflex sensitivity and heart rate variability in the identification of patients at risk for life-threatening arrhythmias: implications for clinical trials. Circulation 2001; 103: 2072-7

41. Costes F, Roche F, Pichot V, Vergnon JM, Garet M, Barthelemy JC: Influence of exercise training on cardiac baroreflex sensitivity in patients with COPD. Eur Respir J 2004; 23: 396-401

42. Kornet L, Hoeks AP, Janssen BJ, Houben AJ, De Leeuw PW, Reneman RS: Neural activity of the cardiac baroreflex decreases with age in normotensive and hypertensive subjects. J Hypertens 2005; 23: 815-23

43. Pinna GD, Maestri R, La Rovere MT: Assessment of baroreflex sensitivity from spontaneous oscillations of blood pressure and heart rate: proven clinical value? Physiol Meas 2015; 36: 741-53

44. Maestri R, Raczak G, Torunski A, Sukiennik A, Kozlowski D, La Rovere MT, Pinna GD: Day-by-day variability of spontaneous baroreflex sensitivity measurements: implications for their reliability in clinical and research applications. J Hypertens 2009; 27: 806-12

45. Pinna GD, Maestri R, Torunski A, Danilowicz-Szymanowicz L, Szwoch M, La Rovere MT, Raczak G: Heart rate variability measures: a fresh look at reliability. Clin Sci (Lond) 2007; 113: 131-40

46. Westerhof BE, Gisolf J, Stok WJ, Wesseling KH, Karemaker JM: Time-domain cross-correlation baroreflex sensitivity: performance on the EUROBAVAR data set. J Hypertens 2004; 22: 1371-80