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Original Article

Does Pulmonary Function Testing Affect Autonomic Nervous System Activity?

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ABSTRACT

OBJECTIVE: The autonomic nervous system (ANS) regulates vital functions such as heart rate (HR) and respiration. Pulmonary function tests (PFTs), which require forced breathing maneuvers, may influence ANS activity, potentially affecting the accuracy of autonomic measurements. This study aimed to investigate the effects of PFT on ANS activity and to assess the reliability of the test order.

MATERIAL AND METHODS: Forty-eight healthy university students (32 women, 16 men; mean age 19±0.92 years) participated. ANS activity was assessed by heart rate variability (HRV) analysis using the Elite HRV Corsense device. HRV was recorded at rest in a seated position (first measurement), was repeated after a 5-minute rest (second measurement), and was recorded again following PFT performed with a Medwelt SP10 spirometer (third measurement).

RESULTS: Comparison of the first and second measurements showed a statistically significant increase only in the root mean square of successive differences (RMSSD) parameter, with no significant changes in other indices. Comparison of the second and third measurements revealed no significant differences in RMSSD or average HR; however, significant changes were observed in the low-frequency (LF) and high-frequency (HF) components and in the LF/HF ratio.

CONCLUSION: Respiratory maneuvers during PFT may temporarily alter ANS activity, particularly affecting parasympathetic–sympathetic balance. The differences between the first and second measurements emphasize the importance of adequate rest periods before HRV assessment. Measurements taken prior to PFT appear to be more reliable for the accurate evaluation of autonomic function.

KEYWORDS: Autonomic nervous system, heart rate variability, pulmonary function test, health economics

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INTRODUCTION

The autonomic nervous system (ANS) is a component of the peripheral nervous system that helps regulate arterial blood pressure, sweating, body temperature, gastrointestinal motility and secretions, bladder emptying, and various other visceral functions.[1] The ANS is primarily activated by several centers located in the hypothalamus, brainstem, and spinal cord. Additionally, signals from the cerebral cortex, particularly the limbic cortex, are transmitted to lower centers, thereby influencing autonomic control. Autonomic efferent signals are transmitted to different regions of the body through two main subdivisions: the sympathetic and parasympathetic nervous systems. Many body regions and visceral functions are predominantly regulated by these two systems.[2]

Heart rate variability (HRV) represents the variation in time intervals between consecutive heartbeats and is one of the most significant cardiovascular and autonomic health indices. HRV, a biomarker of cardiac vagal control is a non-invasive

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electrocardiographic (ECG) measure that can be obtained from long-term Holter monitoring or short-term ECG recordings. Studies suggest that HRV is a reliable test for confirming the presence of dysautonomia.[3,4]

HRV is measured using two main parameters. In time-domain measurements, calculated from the intervals between successive beats in ECG recordings, the intervals between consecutive regular beats originating from the sinoatrial node [referred to as normal-to-normal (NN) intervals] are assessed. The most commonly used indices in a 24-hour HRV recording include the mean heart rate (HR), the percentage of successive R–R intervals (RR) differing by more than 50 ms (pNN50), and the root mean square of successive differences (RMSSD). While mean HR reflects average HR over an hour, RMSSD and pNN50 represent the parasympathetic component of autonomic tone. The other primary parameters, frequency-domain measurements, involve classifying HR signals based on their frequency and power. Evaluated parameters include high-frequency (HF), low-frequency (LF), medium-frequency, ultra-LF, and very-LF.

Regarding autonomic tone, LF reflects sympathetic activity, whereas HF represents the parasympathetic component; therefore, the LF/HF ratio indicates sympathetic/parasympathetic balance.[5,6] Respiration is essential for the survival of all living organisms. The respiratory system comprises organs, tissues, and supporting structures involved in breathing. Spirometric assessment is one of the most common methods for measuring lung capacities and volumes. Although spirometry alone is insufficient for diagnosis, it provides measurements of inhaled and exhaled air volumes over time, expressed in liters. The volume of air generated by the lungs is expressed in liters per second.[7,8]

The respiratory and ANSs are interconnected. Respiration, a physiological process, is regulated both voluntarily and involuntarily; the ANS mediates its involuntary control. The vagus nerve and sympathetic nerve fibers innervate the lungs. Vagus nerve fibers stimulate the muscles and glands of the bronchi, causing bronchoconstriction and increased secretion. Efferent sympathetic nerve fibers relax bronchial smooth muscle, producing bronchodilation.[9]

The ANS and the respiratory system are two essential physiological systems that interact. In this context, pulmonary function tests (PFT) may affect an individual's ANS activity. This study investigates whether PFTs should be administered before or after tests assessing ANS activity. The findings will contribute to a better understanding of the interaction between these two systems and to establishing a scientific foundation regarding the accuracy and reliability of the sequence in which the tests are applied.

Main Points

- Pulmonary function testing (PFT) can cause temporary changes in autonomic nervous system (ANS) activity, as measured by heart rate variability (HRV).
- Both parasympathetic and sympathetic balance are affected following PFT maneuvers.
- A sufficient rest period before HRV measurement is crucial for reliable assessment of autonomic function.
- The findings highlight the importance of considering respiratory testing effects in studies evaluating ANS activity.

MATERIAL AND METHODS

This study was conducted between October 10 and November 30, 2024, and involved students enrolled in the Department of Physiotherapy and Rehabilitation, Faculty of Health Sciences, Mudanya University. The Ethics Committee for Human Research at Mudanya University approved the research on October 4, 2024 (reference number E-40839601-50.04-44).

In the referenced study, a power analysis based on the HF parameter of HRV conducted with a 95% confidence level, 80% power, a P = 0.05 statistical significance level, and an effect size of 0.44 calculated using the G*Power 3.1 program, determined that 33 cases were required.[10]

Forty-eight university students (aged 18–25 years) who volunteered and who had no systemic diseases, no active disease diagnoses, and no cooperation issues were included. Exclusion criteria for the study included pregnancy, malignancy, presence of a pacemaker, current use of medications, smoking within 2 hours of measurements, and alcohol consumption within 24 hours of measurements. All students included in the study were informed and signed informed consent forms. Evaluations were conducted by the same physiotherapist through face-to-face interviews.

Participants' demographic data (age, height, body weight, education level, exercise habits, and smoking habits) were recorded on the demographic data form. PFTs were performed using the Medwelt SP10 spirometer. A separate mouthpiece was used for each individual, and three measurements were taken, with the highest values recorded. All spirometric assessments were conducted in accordance with the American Thoracic Society/European Respiratory Society (ATS/ERS) guidelines. Participants were evaluated in a seated position with a nose clip in place, and an experienced physiotherapist provided standardized verbal instructions and encouragement throughout the procedure.

Each maneuver was initiated following a maximal inspiration and continued with a rapid, forceful, and sustained expiration lasting at least 6 seconds. At least three acceptable maneuvers were obtained for each participant; acceptability and reproducibility were assessed according to ATS/ERS criteria. Reproducibility was confirmed when the difference between the two highest FEV1 and forced vital capacity (FVC) values was ≤150 mL; otherwise, additional trials were performed.

The measured FEV1, FVC, and FEV1/FVC values were documented in the participant follow-up form.[8] In accordance with guideline recommendations, the highest FEV1 and FVC values (not necessarily from the same maneuver) were used in the analysis, and the FEV1/FVC ratio was calculated accordingly.

FEV1: This value represents the maximum volume of air exhaled during the first second of forced expiration following maximal inspiration.

FVC: This refers to the total volume of air forcefully exhaled after a long, deep inspiration.

FEV1/FVC: This ratio is used to diagnose airway obstruction and in the differential diagnosis of restrictive diseases. In young adults, this ratio is typically above 75%; however, it decreases with age. In individuals with airway obstruction, this ratio is below 70%, whereas in restrictive diseases, this ratio is usually normal or increased.

Assessment of the ANS cannot be performed directly using physiological tests. With advances in technology, new methods have been developed, and many of these are used in scientific research. The most commonly used method for measuring ANS activity is HRV analysis. HRV analysis is based on the observation of RR waves present at rest, and in our study this method was applied with the Elite HRV Corsense device, which operates on this principle. The values measured by the device are as follows:[11]

- RMSSD is used as an instantaneous index of the parasympathetic branch of the ANS and forms the basis for the HRV score.

- LF power refers to power in the frequency range between 0.04 and 0.15 Hz. It is directly proportional to sympathetic nervous system activity and thus represents sympathetic activity.

HF power refers to the spectral power in the frequency range between 0.15 and 0.40 Hz. The HF band reflects parasympathetic activity and is highly correlated with time-domain measures such as PNN50 and RMSSD.

- The LF/HF ratio is the ratio of LF power to HF power and is commonly used to indicate the sympathovagal balance. It represents the balance between the opposing branches of the ANS.

- Mean HR.

Study Plan

After participants signed the voluntary consent form, ANS activity was measured while participants were seated. Following this, a 5-minute rest period was allowed for the body to relax, after which ANS activity was measured again. The difference between the two measurements will help to understand the balance between the sympathetic and parasympathetic branches of the nervous system and to indicate whether any change has occurred in the participants' ANS activity. Subsequently, while still seated, participants underwent PFT. Participants were asked to take a maximal inspiration and then exhale as forcefully and rapidly as possible for the measurement. This measurement was repeated three times at one-minute intervals, and the highest value was recorded. This approach aimed to prevent issues such as incorrect respiratory patterns, insufficient motivation, or technical errors during the test. One minute after completion of the PFT, ANS activity was measured again. In this way, statistical answers were sought to determine whether ANS activity remained at the same level before and after the PFT or was affected, and to identify the correct order of measurements.

Statistical Analysis

The data were analyzed using the SPSS 25.0 software package. Continuous variables were presented as mean ± standard deviation, and categorical variables were presented as frequency and percentage. The normality of the data was tested using the Kolmogorov-Smirnov and Shapiro-Wilk tests. If the data were normally distributed, the parametric Paired samples test was used; otherwise, the non-parametric Wilcoxon signed-ranks test was applied. In our study, the mean HR was usually distributed in the initial measurements, whereas RMSSD, LF, HF, and LF/HF were not. The mean HR and RMSSD were approximately normally distributed in the second and third measurements, but LF, HF, and LF/HF did not follow a normal distribution.

RESULTS

Sociodemographic Data

The study included 48 participants, of whom 32 were female and 16 were male. The average age of the individuals was 19±0.92 years, their mean height was 1.70±0.11 m, and their average body weight was 65.22±14.87 kg. The average body mass index of the individuals was 22.09±3.19 kg/m^2. Data related to the demographic characteristics of the cases are presented in Table 1.

Table 2 presents the ANS activity values—RMSSD, HF, LF, LF/HF, and mean HR— measured three times, and the participants' pulmonary function test (PFT) scores.

Table 3 shows the results of the Wilcoxon signed-rank test and the paired-samples test applied to determine the relationship between the first and second measurements of ANS activity. When examining the results of autonomic activity markers, no statistically significant change was observed in any autonomic activity measure except for the RMSSD parameter.

Table 4 shows the results of the Paired samples test and the Wilcoxon signed ranks test, which were applied to determine the relationship between the second and third measurements of the ANS, the latter taken after the PFT. The results indicate that RMSSD and mean HR did not change, whereas LF, HF, and LF/HF showed statistically significant changes.

DISCUSSION

This study investigated the effect of the PFT on ANS activity. To ensure methodological accuracy, particular attention was

Table 1. Sociodemographic data

	n (%)	X ± SD	Min–max
Gender			
Female	32 (66.7)	-	-
Male	16 (33.3)	-	-
Age		19±0.92	18–21
Height (m)		1.70±0.11	1.56–1.99
Weight (kg)		65.22±14.87	42–105
BMI		22.09±3.19	16–29.32

n: number of cases, X: mean, SD: standard deviation, Min: minimum, max: maximum, BMI: body mass index given to the timing of autonomic measurements and resting conditions. Participants were allowed to rest for five minutes after the first measurement, after which the same autonomic assessment was repeated.

When the results were examined, most HRV parameters did not show statistically significant differences between the two measurements. However, a statistically significant increase in RMSSD, an indicator of parasympathetic activity, was observed. This finding suggests that a short resting period following the initial measurement may be necessary to obtain a more stable and reliable assessment of autonomic activity. The increase in parasympathetic modulation after rest indicates that the first measurement, taken without adequate recovery, may reflect transient autonomic responses rather than baseline autonomic function.

From a clinical perspective, this finding highlights the importance of standardizing rest intervals prior to autonomic measurements, particularly in protocols involving respiratory maneuvers or physical effort. The observed difference between the two measurements contributes to understanding the balance between sympathetic (stress-related) and parasympathetic (relaxation-related) activity. An increase in parasympathetic activity after rest may also indicate adequate autonomic adaptability in healthy individuals.

PFTs are widely used tools for the evaluation and monitoring of respiratory system function. These tests require participants to perform standardized respiratory maneuvers involving maximal inspiration and forceful expiration to assess lung volumes and airflow.[12] According to current guidelines, at least three acceptable and reproducible maneuvers are required to ensure measurement reliability. Because these maneuvers demand maximal effort, they may act as an acute physiological stressor capable of altering ANS activity.

For this reason, autonomic activity was measured both before and after the PFT in the present study. When pre- and post-test autonomic parameters were compared, statistically significant changes were observed. Specifically, a significant increase in LF and LF/HF ratio—indices associated with sympathetic modulation—and a significant decrease in HF, reflecting parasympathetic activity, were detected. These findings indicate that PFT-related respiratory maneuvers transiently shift autonomic balance toward sympathetic dominance. Consequently, autonomic measurements intended to reflect baseline autonomic function should be performed before PFT administration. This recommendation may be particularly relevant in clinical or research settings in which HRV is used as an outcome measure.

HRV provides a quantitative assessment of beat-to-beat fluctuations in HR and reflects complex interactions between the cardiovascular and ANSs. HRV is considered a sensitive marker of neurocardiac regulation and physiological adaptability.[13-15] In the present study, HRV analysis using the Elite HRV device enabled an objective and non-invasive evaluation of autonomic responses to respiratory maneuvers. The observed changes support the notion that respiratory effort can significantly modulate cardiac autonomic function.

Previous studies have also demonstrated the influence of respiratory maneuvers on autonomic activity. Ajudia et al.[16] investigated the effects of intensive spirometry on cardiac autonomic function in healthy young adults and reported

Table 2. Autonomic nervous system activity (1st, 2nd, and 3rd measurements) and PFT scores

		X ± SD	Min–max
First measurement	RMSSD	64.39±32.51	9.11–187.32
	LF	5935.05±4342.30	42.12–18730.44
	HF	1950.37±2419.57	143.2–14953.42
	LF/HF	5.43±5.53	0.06–31.60
	Mean heart rate	93.89±13.00	72.02–137.04
Second measurement	RMSSD	69.37±29.38	8.90–132.00
	LF	5472.70±4161.81	92.39–18485.00
	HF	1867.72±1808.34	56.90–7301.00
	LF/HF	4.91±5.10	0.10–29.90
	Mean heart rate	92.35±10.93	71.03–129.08
Third measurement	RMSSD	67.77±27.04	10.00–138.90
	LF	6511.81±3974.23	80.10–14944.30
	HF	2117.79±2217.09	46.64–10467.78
	LF/HF	5.41±5.07	0.54–21.99
	Mean heart rate	92.97±12.32	72.07–129.09
Pulmonary function test measurement	FEV1 (L)	2.67±1.026	0.60–5.20
	FVC (L)	3.79±0.95	2.43–6.37
	FEV1/FVC (%)	0.73±0.24	0.15–1.34

PFT: pulmonary function test, X: mean, SD: standard deviation, Min: minimum, max: maximum, RMSSD: root mean square of successive differences, LF: low-frequency, HF: high-frequency, LF/HF: ratio of low-frequency to high-frequency, L: liter, FVC: forced vital capacity changes in HR parameters following prolonged respiratory training. Although the magnitude and direction of autonomic responses differed between their study and ours, both studies confirm that respiratory maneuvers are capable of altering autonomic regulation. Methodological differences, including intervention duration, breathing frequency, and resting conditions, may explain the variability in outcomes.

Memarian et al.[17] used deep breathing tests to assess cardiovagal function and demonstrated associations between respiratory sinus arrhythmia (RSA) and HRV indices. However, because RSA was not directly measured in the present study, only the HRV-derived parameters included in our results were emphasized in the discussion to avoid interpretative ambiguity. This distinction is important to ensure consistency between reported results and literature comparisons. Nonetheless, both studies underscore the broader concept that controlled respiratory maneuvers have a measurable impact on autonomic regulation.

Ali et al.[18] evaluated parasympathetic reactivity during deep breathing and reported discrepancies between RMSSD and RSA responses. Consistent with their findings, our study observed that RMSSD did not always increase following respiratory maneuvers, suggesting that RMSSD alone may not fully capture parasympathetic reactivity in such contexts. These results indicate that incorporating multiple autonomic indices may provide a more comprehensive assessment of autonomic responses to respiratory interventions.

Malhotra et al.[19] examined the acute effects of slow and deep breathing on HRV and found increased parasympathetic modulation following controlled breathing exercises. Although our study did not involve respiratory training, the shared finding is that respiratory patterns and effort can acutely influence autonomic balance. This supports the hypothesis that even short-term respiratory interventions or tests can transiently modify autonomic function.

Despite these findings, the present study has limitations regarding the temporal dynamics of autonomic recovery. Autonomic activity was assessed only at a single post-test time point, and different rest durations were not compared. Future studies evaluating autonomic responses at multiple intervals following PFT (e.g., immediately after, at 5 and 10 minutes, and during longer recovery periods) may provide deeper insight into autonomic recovery patterns.

Clinically, such information could contribute to optimizing PFT protocols and improving the interpretation of autonomic measurements obtained in conjunction with respiratory testing. Comparing different rest durations may also help establish standardized guidelines for autonomic assessment following PFTs, thereby enhancing the clinical and research utility of these measurements.

Study Limitations

This study has several limitations. Firstly, the limited sample size may restrict the generalizability of the findings. The inclusion of only healthy adults, while strengthening internal validity and allowing for a clearer interpretation of autonomic responses, limits the applicability of the results to clinical populations and different age groups. Therefore, the effects of PFTs on ANS activity should be interpreted with caution when extrapolating to individuals with cardiopulmonary or neurological conditions. Another important limitation is the lack of a detailed evaluation of autonomic responses at multiple time points following the PFT. Autonomic activity was assessed at a single post-test time interval, and the effects of different recovery durations were not

Table 3. The relationship between autonomic nervous system activity in the first and second measurements

	1. measurement	2. measurement	P
RMSSD			
Mean ± SD	64.39±32.51	69.37±29.38	0.036[1]
Min–max	5–187	8.90–132.00	
LF			
Mean ± SD	5935.05±4342.30	5472.70±4161.81	0.644[1]
Min–max	42–18730	92.39–18485.00	
HF			
Mean ± SD	1950.37±2419.57	1867.72±1808.34	0.300[1]
Min–max	14–14953	56.90–7301.00	
LF/HF			
Mean ± SD	5.43±5.53	4.91±5.10	0.460[1]
Min–max	0.06–31.60	0.10–29.90	
Mean heart rate			
Mean ± SD	93.89±13.00	92.35±10.93	0.228[2]
Min–max	72.0–137.0	71.0–129.0	

[1]Wilcoxon signed-ranks test, [2]Paired samples test

SD: standard deviation, Min: minimum, max: maximum, RMSSD: root mean square of successive differences, LF: low-frequency, HF: high-frequency, LF/HF: ratio of low-frequency to high-frequency

Table 4. The relationship between autonomic nervous system activity in the second and third measurements

	1. measurement	2. measurement	P
RMSSD			
Mean ± SD	69.37±29.38	67.77±27.04	0.588[2]
Min–max	8.90–132.00	10.00–138.00	
LF			
Mean ± SD	5472.70±4161.81	6511.81±3974.23	**0.041[1]**
Min–max	92.39–18485.00	80.00–14944.00	
HF			
Mean ± SD	1867.72±1808.34	2117.79±2217.09	**0.052[1]**
Min–max	56.90–7301.00	46.00–10467.00	
LF/HF			
Mean ± SD	4.91±5.10	5.41±5.07	**0.049[1]**
Min–max	0.10–29.90	0.50–21.99	
Mean heart rate			
Mean ± SD	92.35±10.93	92.97±12.32	0.614[2]
Min–max	71.0–129.0	72.0–129.0	

[1]Wilcoxon signed-ranks test, [2]Paired samples test

SD: standard deviation, Min: minimum, max: maximum, RMSSD: root mean square of successive differences, LF: low-frequency, HF: high-frequency, LF/HF: ratio of low-frequency to high-frequency systematically compared. Because autonomic responses may vary with the duration of the rest period following respiratory maneuvers, this limitation may have reduced the depth of interpretation and the study's overall contribution to the literature.

Future studies comparing ANS responses at different recovery intervals after PFT (e.g., immediate, short-term, and longer rest periods) may provide more comprehensive insights and contribute to the development of optimized and standardized testing protocols. Additionally, comparing different test types and autonomic measurement parameters may further enhance the consistency and clinical relevance of future findings.

CONCLUSION

This study investigated the effects of PFTs on ANS activity. The findings suggest that respiratory maneuvers can influence autonomic activity, particularly by altering parasympathetic reactivity. The rest period provided after the initial measurement was important in explaining the increase in parasympathetic activity. The observed increase in RMSSD highlights the importance of rest periods in accurately measuring autonomic activity.

Measurements taken before and after the PFT showed an increase in sympathetic nervous system activity, indicated by LF and the LF/HF ratio, and a decrease in parasympathetic nervous system activity, reflected by HF. These results suggest that PFTs temporarily affect ANS activity, and this effect can be validated using different measurement methods.

The HRV analysis method used in our study proved to be an effective tool for objectively assessing the dynamics of the autonomic system. However, further research is needed to understand how different respiratory maneuvers and measurement parameters affect autonomic activity. Despite methodological differences across studies, respiratory maneuvers significantly influence autonomic activity, and selecting appropriate measurement methods is crucial for accurately evaluating this interaction. These findings may provide valuable insights into the potential effects of PFTs on the ANS and serve as an important reference for future studies.

Ethics

Ethics Committee Approval: The Ethics Committee for Human Research at Mudanya University approved the research on October 4, 2024 (reference number E-40839601-50.04-44).

Informed Consent: Written informed consent was obtained from all participants prior to their inclusion in the study.

Footnotes

Authorship Contributions

Surgical and Medical Practices: G.Y., S.E.G., E.K., Concept: G.Y., S.E.G., E.K., Design: G.Y., S.E.G., E.K., Data Collection or Processing: G.Y., S.E.G., Analysis or Interpretation: G.Y., S.E.G., Literature Search: G.Y., S.E.G., E.K., Writing: G.Y., S.E.G.

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