Original Article

Heart Rate Variability as a Marker of Autonomic Function in Medical Students During Stress: A Prospective Observational Study

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Abstract

Background: Medical students face significant academic and psychological stressors that may impact their autonomic nervous system (ANS) function. Heart rate variability (HRV), a non-invasive marker of autonomic function, has emerged as a valuable tool for assessing stress-related physiological changes. However, comprehensive studies examining HRV patterns in medical students during various stress conditions remain limited. The objective is to investigate HRV as a marker of autonomic function in medical students during academic stress and to examine the relationships between HRV parameters, perceived stress levels, and psychological well-being. **Material and Methods:** A prospective observational study was conducted with 120 medical students. HRV measurements were obtained using standardized 5-minute ECG recordings during baseline (low-stress) and examination (high-stress) periods. Time-domain parameters (SDNN, RMSSD, pNN50) and frequency-domain parameters (LF, HF, LF/HF ratio) were analyzed. Psychological assessments included the Perceived Stress Scale (PSS-10), Depression Anxiety Stress Scale (DASS-21), and Pittsburgh Sleep Quality Index (PSQI). **Results:** Significant reductions were observed in all HRV parameters during the high-stress period compared to baseline (SDNN: 52.6 ± 14.2 vs. 38.4 ± 12.6 ms; RMSSD: 46.8 ± 16.4 vs. 31.2 ± 11.8 ms; $p < 0.001$ for all). The LF/HF ratio increased significantly (0.82 ± 0.32 vs. 1.34 ± 0.48; $p < 0.001$), indicating sympathetic predominance. Strong negative correlations were found between HRV indices and PSS-10, DASS-21, and PSQI scores. **Conclusion:** HRV serves as a sensitive physiological marker for autonomic dysfunction in medical students experiencing academic stress. Poor sleep quality and high perceived stress are independent predictors of HRV impairment. These findings support the implementation of stress-management and sleep-hygiene interventions in medical education programmes.

Keywords: Heart rate variability, autonomic nervous system, medical students, academic stress, parasympathetic activity, sympathetic activity, stress biomarkers.

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Introduction

Medical education is characterised by intense academic demands, extensive curricula, and high-stakes examinations that collectively impose significant psychological and physiological stress on students.[1,2] The prevalence of stress, anxiety, and burnout among medical students has been well-documented globally, with rates exceeding 50% in various populations.[3,4] Beyond the immediate psychological impact, chronic stress exposure during medical training may lead to long-term health consequences including cardiovascular disease, metabolic disorders, and mental health conditions.[5,6]

The autonomic nervous system (ANS) plays a crucial role in the body's response to stress, mediating the balance between sympathetic (fight-or-flight) and parasympathetic (rest-and-digest) activity. Dysregulation of autonomic function has been implicated in various stress-related pathologies and represents a key mechanism linking psychological stress to physical health outcomes.[7,8] Therefore, objective assessment of autonomic function in medical students may provide valuable insights into their physiological adaptation to academic stress.

Heart Rate Variability as a Biomarker: Heart rate variability (HRV) refers to the variation in time intervals between consecutive heartbeats and serves as a non-invasive, quantitative marker of ANS activity.[9,10] Time-domain measures — including the standard deviation of normal-to-normal intervals (SDNN), root mean square of successive differences (RMSSD), and percentage of successive normal sinus intervals exceeding 50 ms (pNN50) — primarily reflect parasympathetic modulation.[11] Frequency-domain parameters including low-frequency (LF) power, high-frequency (HF) power, and the LF/HF ratio provide information about sympathetic activity, parasympathetic activity, and sympathovagal balance, respectively.[12,13]

Higher HRV generally indicates better cardiovascular health and greater autonomic flexibility, while reduced HRV has been associated with increased cardiovascular risk, inflammation,

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and all-cause mortality.[14,15] In the context of psychological stress, decreased HRV reflects diminished parasympathetic tone and heightened sympathetic activity, consistent with the physiological stress response.[16,17]

HRV in Medical Students: Current Evidence: Recent studies have begun to explore HRV patterns in medical students. A 2024 cross-sectional study found that sleep disturbances — common in this population — were associated with altered psychomotor performance and psychological distress, though HRV relationships were complex.[18] Investigations into digital detox interventions have demonstrated that reducing screen time can improve HRV parameters, suggesting modifiable determinants of autonomic balance.[19] Brief active breaks during study sessions have also been shown to enhance HRV and cognitive performance,[21] while poor sleep quality has been correlated with altered HRV in student populations.[20]

Aims and objectives: The aim of this study is to evaluate autonomic nervous system changes in medical students by analysing heart rate variability (HRV) during baseline and examination periods, and to determine its association with psychological stress, anxiety, and well-being. The objectives are to characterise HRV parameters under varying stress conditions, assess the correlation between HRV indices and validated psychological measures, identify predictors of autonomic dysfunction such as baseline stress and sleep quality, and evaluate the sensitivity of HRV as a non-invasive marker for detecting stress-related autonomic changes among medical students.

Materials and Methods

Study Design: This prospective observational study was conducted at a medical institution during the academic year 2022–2023. The study protocol was approved by the Institutional Ethics Committee and conducted in accordance with the Declaration of Helsinki. All participants provided written informed consent prior to enrolment.

Participants: Medical students enrolled in years 2–5 of the MBBS programme, aged 18–30 years with no known cardiovascular disease or autonomic dysfunction, were eligible. Exclusion criteria included current use of medications affecting cardiovascular or autonomic function, diagnosed psychiatric disorders requiring pharmacological treatment, pregnancy, cardiac arrhythmias, chronic diseases affecting autonomic function (diabetes mellitus, thyroid disorders), and excessive use of substances known to affect HRV.

Based on previous studies and assuming a moderate effect size (Cohen's d = 0.5) for SDNN differences between stress conditions ($\alpha = 0.05$; power = 0.80), a minimum sample of 100 participants was required. Accounting for 15–20% potential dropout, we aimed to recruit 120 participants using stratified random sampling to ensure representation across academic years and gender.

Study Timeline and Assessments

Two assessment phases were conducted:

- **Phase 1 (Baseline/Low-Stress):** Mid-semester period, at least 4 weeks before major examinations, during regular class schedule.
- **Phase 2 (High-Stress):** Examination period, 1–3 days before major examinations, during peak academic stress.

Each phase included demographic and lifestyle questionnaire, psychological assessments, HRV measurement, and sleep quality assessment.

HRV Measurement Protocol: HRV measurements were performed using a standardised 12-lead ECG system (PowerLab, AD Instruments, Australia) at a 1000 Hz sampling rate. Recordings were conducted in a quiet, temperature-controlled room (22–24°C) between 09:00 and 12:00 to minimise circadian variation. Participants were instructed to abstain from caffeine, alcohol, and vigorous exercise for 24 hours prior to assessment, ensure adequate sleep (≥6 hours), avoid heavy meals for 2 hours before, and refrain from talking during recording.

Following 10 minutes of supine rest, 5-minute ECG recordings were obtained during quiet, spontaneous breathing with real-time quality monitoring. R-wave detection was performed using automated algorithms with manual verification, and RR interval tachograms were generated after removal of ectopic beats and artefacts.

HRV Analysis: Time-domain parameters analysed included SDNN, RMSSD, pNN50, mean RR interval, and mean heart rate. Frequency-domain parameters were derived using Fast Fourier Transform (Welch's periodogram method) and included VLF (0.003–0.04 Hz), LF (0.04–0.15 Hz), HF (0.15–0.40 Hz), LF and HF in normalised units, and the LF/HF ratio. All analyses followed Task Force guidelines (1996).

Psychological Assessments: The 10-item Perceived Stress Scale (PSS-10; range 0–40) was used to assess perceived stress. The 21-item Depression Anxiety Stress Scale (DASS-21) measured symptoms of depression, anxiety, and stress. Sleep quality was assessed using the Pittsburgh Sleep Quality Index (PSQI; global score 0–21; scores >5 indicating poor sleep quality).

Statistical Analysis: Data were checked for normality using Shapiro-Wilk tests and visual Q-Q plot inspection; non-normally distributed variables were log-transformed. Paired t-tests or Wilcoxon signed-rank tests compared HRV parameters between periods. Pearson or Spearman correlations examined relationships between HRV and psychological measures. Multiple linear regression models identified predictors of HRV during stress, adjusting for age, gender, BMI, and physical activity. Subgroup analyses explored differences by gender and sleep quality. Two-tailed $p < 0.05$ was considered significant with Bonferroni correction applied for multiple comparisons. All analyses were performed using SPSS Statistics v25 and R v4.2.1.

Results

Participant Characteristics: A total of 138 medical students were screened, of whom 120 met inclusion criteria and completed both baseline and high-stress assessments (completion rate: 87.0%). The demographic and baseline characteristics are presented in [Table 1]. The mean age was 22.4 ± 1.9 years, with 56.7% female. The majority were in third and fourth year (51.6%). Mean BMI was 22.1 ± 2.7 kg/m², and

45.8% reported moderate physical activity levels. Only 15.0% had prior experience with meditation or relaxation practices.

Table 1: Participant Demographics and Baseline Characteristics

Characteristic	Value (n = 120)
Age (years), mean ± SD	22.4 ± 1.9
Gender, n (%)	
Male	52 (43.3%)
Female	68 (56.7%)
Academic Year, n (%)	
Second year	32 (26.7%)
Third year	28 (23.3%)
Fourth year	34 (28.3%)
Fifth year	26 (21.7%)
Body Mass Index (kg/m²), mean ± SD	22.1 ± 2.7
Physical Activity Level, n (%)	
Low	42 (35.0%)
Moderate	55 (45.8%)
High	23 (19.2%)
Caffeine intake (mg/day), median (IQR)	180 (120–240)
Sleep duration (hours/night), mean ± SD	6.1 ± 1.1
Study hours per day, mean ± SD	8.4 ± 2.0
Previous meditation/relaxation practice, n (%)	18 (15.0%)

SD: standard deviation; IQR: interquartile range

Psychological Assessments

[Table 2] presents psychological assessment scores at both study phases. Significant increases were observed in all psychological distress measures during the high-stress period (all $p < 0.001$). The mean PSS-10 score increased by 51.5% (16.3 ± 4.2 vs. 24.7 ± 4.8), DASS-21 anxiety subscale increased by 105.6% (7.1 ± 3.2 vs. 14.6 ± 4.5), and sleep quality deteriorated markedly as evidenced by a significant increase in PSQI global scores (6.2 ± 2.1 vs. 9.4 ± 2.6). The proportion of poor sleepers increased from 53.3% to 81.7%.

Table 2: Psychological Assessment Scores at Baseline and High-Stress Periods

Measure	Baseline Period	High-Stress Period	p-value
PSS-10 Score, mean ± SD	16.3 ± 4.2	24.7 ± 4.8	< 0.001
DASS-21 Depression, mean ± SD	8.4 ± 3.6	13.2 ± 4.1	< 0.001
DASS-21 Anxiety, mean ± SD	7.1 ± 3.2	14.6 ± 4.5	< 0.001
DASS-21 Stress, mean ± SD	11.6 ± 4.0	18.9 ± 4.7	< 0.001
PSQI Global Score, mean ± SD	6.2 ± 2.1	9.4 ± 2.6	< 0.001
Poor Sleep Quality (PSQI > 5), n (%)	64 (53.3%)	98 (81.7%)	< 0.001

PSS-10: Perceived Stress Scale; DASS-21: Depression Anxiety Stress Scale; PSQI: Pittsburgh Sleep Quality Index. Paired t-tests used for continuous variables.

Heart Rate Variability — Time-Domain Parameters

[Table 3] presents time-domain HRV parameters. Significant reductions were observed in all parameters during the high-stress period (all $p < 0.001$), with large effect sizes.

- SDNN decreased by 27.0% (52.6 ± 14.2 ms at baseline vs. 38.4 ± 12.6 ms during stress; Cohen's d = 1.05).
- RMSSD showed a 33.3% reduction (46.8 ± 16.4 ms vs. 31.2 ± 11.8 ms; d = 1.08).
- pNN50 decreased by 43.8% (22.4 ± 10.6% vs. 12.6 ± 8.2%; d = 1.02).
- Mean heart rate increased significantly from 69.4 ± 5.2 bpm to 75.8 ± 6.1 bpm (d = 1.10).

Table 3: Time-Domain HRV Parameters at Baseline and High-Stress Periods

Parameter	Baseline Period	High-Stress Period	p-value	Effect Size (d)
Mean RR interval (ms)	872 ± 68	804 ± 74	< 0.001	0.94
Mean Heart Rate (bpm)	69.4 ± 5.2	75.8 ± 6.1	< 0.001	1.10
SDNN (ms)	52.6 ± 14.2	38.4 ± 12.6	< 0.001	1.05
RMSSD (ms)	46.8 ± 16.4	31.2 ± 11.8	< 0.001	1.08
pNN50 (%)	22.4 ± 10.6	12.6 ± 8.2	< 0.001	1.02

Values presented as mean ± standard deviation. Paired t-tests used; effect size calculated as Cohen's d. SDNN: standard deviation of NN intervals; RMSSD: root mean square of successive differences; pNN50: percentage of successive NN intervals differing by > 50 ms.

Heart Rate Variability — Frequency-Domain Parameters

[Table 4] presents frequency-domain HRV parameters. Significant alterations in autonomic balance were observed during the high-stress period.

- HF power decreased by 50.3% (648 ± 284 ms² vs. 322 ± 178 ms²; d = 1.38), indicating markedly reduced parasympathetic activity.
- LF/HF ratio increased by 63.4% (0.82 ± 0.32 vs. 1.34 ± 0.48; d = 1.28), suggesting sympathetic predominance.
- Total power decreased by 33.9% (1486 ± 524 ms² vs. 982 ± 386 ms²; d = 1.11).
- LF power did not show a statistically significant change (p = 0.217).

Table 4: Frequency-Domain HRV Parameters at Baseline and High-Stress Periods

Parameter	Baseline Period	High-Stress Period	p-value	Effect Size (d)
LF power (ms²)	412 ± 198	386 ± 172	0.217	0.14
HF power (ms²)	648 ± 284	322 ± 178	< 0.001	1.38
Total Power (ms²)	1486 ± 524	982 ± 386	< 0.001	1.11
LF (n.u.)	43.8 ± 12.4	56.4 ± 11.8	< 0.001	1.04
HF (n.u.)	56.2 ± 12.4	43.6 ± 11.8	< 0.001	1.04
LF/HF ratio	0.82 ± 0.32	1.34 ± 0.48	< 0.001	1.28

Values presented as mean ± standard deviation. Paired t-tests used on log-transformed data; effect size calculated as Cohen's d. LF: low frequency; HF: high frequency; n.u.: normalised units.

Correlations Between HRV and Psychological Measures: [Table 5] presents Pearson correlation coefficients between HRV parameters and psychological measures during the baseline period. Significant negative correlations were found between all time-domain HRV parameters and psychological distress measures. Stronger correlations were observed for parasympathetic indices (RMSSD, HF power) than for overall variability (SDNN). The LF/HF ratio showed significant positive correlations with all psychological distress measures, indicating that greater perceived stress was associated with greater sympathetic predominance.

Table 5: Correlations Between HRV Parameters and Psychological Measures at Baseline

HRV Parameter	PSS-10	DASS-Depression	DASS-Anxiety	DASS-Stress	PSQI
SDNN	−0.48**	−0.36*	−0.44**	−0.41*	−0.39*
RMSSD	−0.54**	−0.39*	−0.52**	−0.44*	−0.47**
pNN50	−0.42*	−0.31	−0.40*	−0.33	−0.38*
LF/HF ratio	0.51**	0.37*	0.49**	0.46**	0.40*
HF power	−0.56**	−0.42*	−0.53**	−0.47*	−0.50**

Values represent Pearson correlation coefficients. *p < 0.05; **p < 0.01. PSS-10: Perceived Stress Scale; DASS: Depression Anxiety Stress Scale; PSQI: Pittsburgh Sleep Quality Index.

Subgroup Analyses

Gender Differences: [Table 6] presents HRV parameters by gender during the high-stress period. No statistically significant differences were observed between male and female students for any HRV parameter (all p > 0.05), suggesting comparable autonomic responses to examination stress across genders.

Table 6: HRV Parameters by Gender During High-Stress Period

Parameter	Male (n = 52)	Female (n = 68)	p-value
SDNN (ms)	36.8 ± 13.1	39.6 ± 12.2	0.183
RMSSD (ms)	29.4 ± 11.2	32.8 ± 12.4	0.092
LF/HF ratio	1.40 ± 0.52	1.28 ± 0.44	0.127
HF power (ms²)	304 ± 164	338 ± 190	0.214

Values presented as mean ± standard deviation. Independent t-tests used.

Sleep Quality: Participants were categorised as good sleepers (PSQI ≤ 5; n = 22) or poor sleepers (PSQI > 5; n = 98) during the high-stress period. Poor sleepers demonstrated significantly lower HRV parameters and greater autonomic dysfunction (Table 7). This pattern was consistent across both time-domain and frequency-domain measures.

Table 7: HRV Parameters by Sleep Quality During High-Stress Period

Parameter	Good Sleepers (n = 22)	Poor Sleepers (n = 98)	p-value
SDNN (ms)	43.8 ± 13.4	34.2 ± 11.6	< 0.001
RMSSD (ms)	36.4 ± 12.8	27.2 ± 10.6	< 0.001
LF/HF ratio	1.18 ± 0.42	1.48 ± 0.52	0.002
HF power (ms²)	378 ± 196	274 ± 158	< 0.001

Values presented as mean ± standard deviation. Independent t-tests used.

Predictors of HRV During Stress: Multiple linear regression analysis identified significant predictors of SDNN during the high-stress period (Table 8). The model explained 52.4% of the variance in stress-period SDNN (R^2 = 0.524; Adjusted R^2 = 0.504; F = 26.38, p < 0.001). Baseline SDNN was the strongest predictor, followed by PSS-10 score and PSQI global score. Physical activity level showed a protective effect. Gender and academic year were not significant independent predictors.

Table 8: Multiple Linear Regression Analysis — Predictors of SDNN During High-Stress Period

Predictor	β Coefficient	95% CI	p-value
Baseline SDNN	0.482	0.38 – 0.58	< 0.001
PSS-10 Score	−0.316	−0.42 – −0.21	< 0.001
PSQI Global Score	−0.214	−0.31 – −0.12	0.003
Physical Activity Level	0.142	0.06 – 0.22	0.018
Gender (Female)	0.068	−0.04 – 0.17	0.214
Academic Year	−0.058	−0.16 – 0.04	0.241

β: standardised regression coefficient; CI: confidence interval. Model R^2 = 0.524; Adjusted R^2 = 0.504; F = 26.38, p < 0.001.

Discussion

Principal Findings: This prospective study provides comprehensive evidence that HRV is a sensitive physiological marker of autonomic function in medical students experiencing academic stress. We observed a 27.0% reduction in SDNN (52.6 ± 14.2 vs. 38.4 ± 12.6 ms; p < 0.001) and a 33.3% reduction in RMSSD (46.8 ± 16.4 vs. 31.2 ± 11.8 ms; p < 0.001) during the examination period, accompanied by a 63.4% increase in the LF/HF ratio (0.82 ± 0.32 vs. 1.34 ± 0.48; p < 0.001). These physiological changes were accompanied by substantial increases in psychological distress, with PSS-10 scores rising by 51.5% (16.3 ± 4.2 vs. 24.7 ± 4.8) and DASS-21 anxiety scores by 105.6% (7.1 ± 3.2 vs. 14.6 ± 4.5). Taken together, these findings confirm that the psychological burden of examination stress is mirrored by measurable autonomic dysregulation.

Demographic Profile: Of the 120 participants, 56.7% were female and 43.3% were male, with a mean age of 22.4 ± 1.9 years. This distribution is consistent with the increasing feminisation of medical education reported in many Asian and European cohorts.[1,2] The majority (81.7%) reported poor sleep quality (PSQI > 5) during the examination period, which is comparable to the 74–85% rates of sleep disturbance documented in other medical student populations under examination conditions.[18,20] The mean study hours per day of 8.4 ± 2.0 and low rates of prior relaxation practice (15.0%) further reflect the demanding lifestyle typical of MBBS students and contextualise the autonomic dysregulation observed in our cohort.

Autonomic Dysfunction During Academic Stress: The reductions in all time-domain HRV parameters during the examination period are consistent with findings from comparable studies. Mishra et al. reported statistically significant decreases in SDNN and RMSSD during examination periods in Indian medical students, with SDNN values broadly similar in magnitude to those observed in our cohort.[12] Similarly, Georgiou et al. found significant HRV reductions during oral examinations compared to baseline in first-year medical students, with the LF/HF ratio showing the most robust increase, mirroring our finding of a 63.4% elevation in this index.[16] In a study of 48 first-year medical students from the United States, Shaffer et al. documented a decline in SDNN from 108.16 ± 13.86 ms at the start of the semester to 56.36 ± 4.07 ms by the end, alongside a PSS-10 increase from 12.7 ± 0.75 to 20.6 ± 1.00 — a trajectory strikingly consistent with the pattern observed in our study, albeit over a longer temporal frame.[17] Furthermore, a 2019 study of Lebanese university students monitoring HRV across four phases of an examination found that SDNN, RMSSD, pNN50, and LF/HF all changed significantly in directions congruent with our findings, with HRV lowest during active examination and recovering afterwards.[24] Collectively, these parallels across diverse geographical and institutional settings strengthen the generalisability of our observations.

The 50.3% decrease in HF power (648 ± 284 vs. 322 ± 178 ms²; p < 0.001; d = 1.38) represents the most pronounced frequency-domain change, reflecting a marked withdrawal of parasympathetic tone. This is consistent with the neurovisceral integration model, which posits that vagal activity is selectively suppressed during cognitively demanding or psychologically threatening tasks.[7] Notably, LF power did not change significantly (p = 0.217), suggesting that the observed sympathovagal imbalance was driven primarily by parasympathetic withdrawal rather than absolute sympathetic activation — a distinction that has been noted in other examination-stress paradigms.[16] The convergent evidence from both time- and frequency-domain parameters with large effect sizes (d = 1.02 to 1.38) underscores the clinical meaningfulness of these stress-related autonomic changes.

Psychological Assessments and HRV Correlations: The significant elevations in all DASS-21 subscale scores and the PSS-10 during the examination period confirm that our stress manipulation was ecologically valid. The 51.5% increase in PSS-10 scores (16.3 ± 4.2 to 24.7 ± 4.8) is comparable to the 62.5% increase reported by Shaffer et al. (12.7 to 20.6) over a full academic semester,[17] and to the 40–55% PSS increases documented in other Indian medical student studies during examination seasons.[1,3] Critically, these psychological changes were not independent of autonomic function: RMSSD showed the strongest negative correlations with PSS-10 (r = −0.54), DASS-Anxiety (r = −0.52), and PSQI (r = −0.47), while HF power showed similarly strong inverse relationships. The strength of these correlations — particularly for parasympathetic indices — is in close agreement with the meta-analytic evidence summarised by Thayer et al,[7] and Dishman et al,[17] who reported pooled correlations between −0.30 and

−0.55 between vagal HRV measures and self-reported stress or anxiety. These convergent findings validate HRV as an objective physiological correlate of subjective psychological distress in medical students.

Sleep Quality and HRV

The proportion of poor sleepers rose significantly from 53.3% at baseline to 81.7% during the examination period, and poor sleepers demonstrated substantially lower HRV across all parameters — for example, SDNN was 34.2 ± 11.6 ms in poor sleepers versus 43.8 ± 13.4 ms in good sleepers ($p < 0.001$) during the high-stress period. These findings extend prior observations from medical student populations. Xiao et al. conducted a 24-hour HRV study in Chinese medical students and reported that sleep disturbance components of the PSQI were negatively correlated with SDNN during both waking ($r = −0.285$) and sleeping periods ($r = −0.317$),[20] consistent with our cross-sectional findings. Dhanusri et al. similarly observed that insomnia in medical students was associated with altered HRV parameters and impaired psychomotor performance.[18] The finding that PSQI global score was an independent predictor of stress-period SDNN in our regression model ($\beta = −0.214$; $p = 0.003$) confirms that sleep quality contributes to autonomic resilience beyond its overlap with perceived stress, suggesting that interventions targeting sleep hygiene may have direct benefits for autonomic regulation.

Predictors of Autonomic Function During Stress: Our regression model explained 52.4% of the variance in stress-period SDNN ($R^2 = 0.524$; $F = 26.38$; $p < 0.001$), with baseline SDNN ($\beta = 0.482$) emerging as the strongest predictor. This finding — that individuals with better resting autonomic function are more resilient to stress-induced HRV reductions — aligns with the concept of 'autonomic reserve' described by Thayer et al,[7] and has been corroborated in occupational stress studies.[16] Higher perceived stress (PSS-10; $\beta = −0.316$) and poor sleep quality (PSQI; $\beta = −0.214$) independently predicted lower SDNN, confirming that both psychological load and sleep health influence autonomic resilience over and above baseline physiology. Physical activity level showed a protective effect ($\beta = 0.142$; $p = 0.018$), consistent with literature demonstrating that regular aerobic exercise increases vagal tone and SDNN in young adults.[11,12]

Implications of Intervention Evidence: Our results carry direct implications for the design of student wellness programmes. Farrukh et al. demonstrated that a two-week digital detox combined with alternative activities (mindfulness, physical exercise, and journalling) significantly improved SDNN in medical students, with the intervention group achieving a mean SDNN of 50.1 ± 8.4 ms compared to 39.5 ± 7.7 ms in controls ($p < 0.001$; $r = 0.62$).[19] Notably, the post-intervention SDNN in the digital detox group (50.1 ms) closely approximates our baseline SDNN (52.6 ms) rather than the examination-period value (38.4 ms), highlighting that structured lifestyle interventions may restore autonomic function to near-resting levels even under the continued demands of medical education. Complementarily, Latino et al. found that 12 weeks of daily active breaks in university students produced significant improvements in HRV and heart rate recovery alongside enhanced psychological resilience and cognitive performance on the Stroop test.[21] These findings suggest that brief, feasible physical activity interventions integrated into the academic schedule may attenuate the stress-induced HRV reductions documented in our cohort.

Gender Differences: No statistically significant gender differences in HRV responses to examination stress were observed in our study, though female students showed numerically higher SDNN (39.6 ± 12.2 vs. 36.8 ± 13.1 ms) and lower LF/HF ratios (1.28 ± 0.44 vs. 1.40 ± 0.52) during the high-stress period. This is broadly consistent with the mixed literature on gender differences in autonomic reactivity. Saleh et al., in their study of Lebanese university students (n = 90; 30 males, 60 females), similarly found no significant gender-based differences in SDNN, RMSSD, or pNN50 during examination conditions.[24] In contrast, some studies have reported higher resting HRV in females of reproductive age attributable to hormonal influences on cardiac vagal tone.[22] The absence of significant differences in our cohort may reflect the relatively narrow age range (18–30 years), the comparable examination-stress exposure across genders, and the moderating influence of shared academic workload.

Clinical and Educational Implications

These findings have several important implications for medical education and student wellness:

1. **Early Identification:** HRV monitoring could serve as an objective tool for identifying students at risk for stress-related health consequences. Students with SDNN persistently below 40 ms during examination periods, as observed in our poor-sleeper subgroup (34.2 ± 11.6 ms), may warrant early referral to counselling or wellness services.
2. **Intervention Targets:** The modifiable predictors identified — sleep quality, physical activity, and perceived stress — suggest actionable targets. The digital detox intervention by Farrukh et al,[19] and the active-breaks programme by Latino et al,[21] offer evidence-based, low-cost strategies that could be embedded within medical curricula.
3. **Programme Evaluation:** HRV could serve as an objective outcome measure for evaluating the effectiveness of student wellness initiatives, complementing subjective psychological questionnaires and providing a more direct window into physiological adaptation.
4. **Curriculum Design:** Recognition of the physiological toll of examination stress should inform scheduling — spacing high-stakes assessments and incorporating mandatory rest periods may help preserve autonomic function across the academic year.

Limitations: Several limitations should be considered when interpreting our findings. First, our sample was drawn from a single institution, which may limit generalisability to other medical schools with different curricular structures or cultural contexts. Second, only two assessment time-points were captured; more frequent longitudinal measurements across the academic year would provide richer data on the temporal dynamics of autonomic function. Third, while 5-minute ECG recordings are standard for short-term HRV analysis per Task

Force guidelines,[9] 24-hour ambulatory monitoring would capture additional diurnal and nocturnal aspects of autonomic regulation. Fourth, the observational design precludes definitive causal inference regarding the direction of relationships between psychological stress and HRV. Fifth, self-report psychological measures may be subject to recall bias or social desirability effects, particularly in a student population. Sixth, hormonal influences on HRV — particularly the menstrual cycle phase in female participants — were not controlled for and may have contributed to within-group variability.

Conclusion

This prospective observational study provides compelling evidence that HRV serves as a sensitive and objective biomarker of autonomic nervous system function in medical students experiencing academic stress. Significant reductions in all HRV parameters during examination periods — characterised by reduced overall variability, diminished parasympathetic activity, and increased sympathovagal ratio — were strongly correlated with elevated psychological distress, anxiety, and poor sleep quality.

Key conclusions include: (1) medical students exhibit marked autonomic dysfunction during examination stress; (2) psychophysiological measures are strongly integrated, with subjective stress reflected in objective autonomic indices; (3) poor sleep quality amplifies stress-related autonomic dysfunction; (4) baseline autonomic function, perceived stress, and sleep quality independently predict HRV during stress; and (5) physical activity exerts a protective effect.

Based on these findings, it is recommended that medical institutions incorporate HRV-based monitoring into student wellness programmes to enable early identification of at-risk individuals. Targeted interventions addressing sleep hygiene, stress management, and physical activity may help preserve autonomic function during high-stress periods of medical training. HRV monitoring represents a promising, objective, non-invasive complement to existing psychological screening tools in supporting the health and resilience of future physicians.

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Conflicts of interest

There are no conflicts of interest.

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