Original Article



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Features of Changes in Heart Rate Variability among Medical Students during the Education Process

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Abstract

Background: We investigated the changes of Heart Rate Variability (HRV) parameters, which reflect the state of the regulatory mechanisms of cardiovascular system, among medical students during routine academic period and examination session.

Methods: The study was conducted in Jan-Feb 2024, at the IHSM in Bishkek, Kyrgyz Republic, among the healthy 70 students (mean age 19.71 ± 1.54 years) from India and Pakistan. For each student, two ECG recordings were performed: one 3 weeks before the exam and one 10-15 minutes after exam.

Results: The results of study demonstrated that after the exam, students presented significantly higher HR, AMo, SI, VLF, LF and lower SDNN, TP, HF compared before the exam (P<0.05). Distribution of students showed that on a routine academic period 53% of students belonged to sympathotonic, 14% to vagotonic and 33% to normotonic. During the examination period the number of sympathotonic increased to 74%, but the number of vagotonic and normotonic decreased to 4% and 22% respectively.

Conclusion: Each type revealed an individual reaction to exam stress: in the group of vagotonic there was a slight activation of the SNS; in sympathotonic students there was a tension of regulatory mechanisms, leading to depletion of adaptive mechanisms; in the group of normotonic there was a shift in the vegetative balance towards the sympathetic link of ANS.

Keywords: Heart rate variability; Examination stress; Vegetative regulation; Autonomic nervous system

Introduction

In the last few years, Kyrgyzstan has become more and more attractive country for international students. Medical universities are no exception, where students from India, Pakistan, Bangladesh, Turkey and other countries receive education. During studies at the university, students experience high psycho-emotional loads, which negatively affect the functional state of the body and cause, first of all, changes in the autonomic regulation of the heart rhythm (1). The most val-



Copyright © 2024 Manasovna Satarkulova et al. Published by Tehran University of Medical Sciences. This work is licensed under a Creative Commons Attribution-NonCommercial 4.0 International license. (https://creativecommons.org/licenses/by-nc/4.0/). Non-commercial uses of the work are permitted, provided the original work is properly cited uable and a non-invasive method to define the state of the cardiovascular system and vegetative regulation of the heart from a simple electrocardiogram (ECG) recording is the mathematical analysis of Heart rate variability (HRV), which is a promising tool for assessing various cardiovascular and non-cardiovascular disorders (2,3). One of the reasons of the development of cardiovascular diseases is stress, which leads to reduce human performances, especially dangerous for the medical profession, and changes the balance of the autonomic nervous system (ANS) by the increasing sympathetic activity and thus a decrease in heart rate variability (4).

For this reason, several studies investigated cardiovascular reaction induced by different stress using HRV: physical, psychomotor challenges and social interaction stressors (5-17). Other studies considered the effect of academic stress during the examination time on HRV in healthy university students, as this is a stressful factor in real life (18-22). In examination conditions, students present lower beat-beat variability and higher heart rate compared to normal nonacademic situations (23).

The International Higher School of Medicine (IHSM) is the private university in Kyrgyz Republic and includes 100% of international students, mainly from India and Pakistan. In recent times, that the population of modern students is characterized by unsatisfactory indicators of physical health, high levels of anxiety and low indicators of social optimism (24). In this regard, dynamic control over the functional state of students and their adaptive capabilities is very relevant and important (25). Students at the IHSM generally pass through a stressful period during the examination time; the main goal of this research was to study the changes in HRV parameters, which reflect the state of the regulatory mechanisms of the cardiovascular system among medical students during routine academic period and examination session. Moreover, it was of interest to determine the individual typological features of vegetative regulation response in foreign students to exam stress during the educational process.

Methods

The study was conducted in Jan-Feb 2024, at the IHSM in Bishkek, Kyrgyz Republic, among the healthy 70 male undergraduates (mean age 19.71 \pm 1.54 years) from India and Pakistan. Students had to be healthy and not to take any drugs.

The study was approved by the Ethics Committee at the IHSM (Protocol No. 4 of November 9, 2016) and corresponds to the principles outlined in the Helsinki declaration.

Electrocardiogram (ECG) recording and calculation of HRV were carried out by using a psychophysiological testing device of the company "MEDIKOM MTD" (Russia) in siting position for 5 minutes. The participants were monitored 2 times: 3 weeks before the examination session in routine academic conditions and 10-15 minutes after their exam was done. In the process of HRV registration and analysis the recommendations of the Russian scientists (3) and experts of the European Cardiological and North American Electrophysiological Societies was followed (2).

Time domain and frequency domain parameters of HRV were assessed. Time domain analysis include: heart rate (HR, bpm), standard deviation of all RR intervals (SDNN, ms), Mode (Mo, ms), which are determined primarily by the influence of the parasympathetic nervous system (PSNS) and Mode amplitude (AMo, %), which reflect the measure of the mobilizing impact the of sympathetic nervous system (SNS) activity. Besides, the stress index (SI, c.u.) was used in the analysis, which reflects the degree of centralization of heart rhythm management.

Frequency domain analysis consisted in measuring power of high frequency (HF, ms²), low frequency (LF, ms²) and very low frequency (VLF, ms²) waves in the total power of the spectrum (TP, ms²), which reflect sympathetic and parasympathetic activity of the heart. The LF/HF ratio, determines the balance in ANS activity of the heart, and the predominance of the sympathetic or parasympathetic activity acting on it (Table 1).

Variables	Description	Units	Normal values (mean ± SD)
HR	Heart rate	bpm	60-90
SDNN	Standard deviation of all RR intervals	ms	59.5 ± 5
Mo	Mode	ms	900 ± 30
AMo	Mode amplitude	%	35±3
SI	Stress index	c.u.	80-150
ТР	Total power	ms^2	3466±1018
VLF	Very low frequency waves (≤ 0.04 Hz)	ms^2	765 ± 410
LF	Low frequency waves (0.04 to 0.15 Hz)	ms^2	1170 ± 416
HF	High frequency waves (0.15 to 0.4 Hz)	ms^2	975±203
LF/HF	Ratio of sympathetic to parasympathetic ac-	c.u.	1.5-2.0
	tivity		

 Table 1: Definition and normal values of time domain and frequency domain parameters of HRV assessed in this study

The initial type of vegetative regulation was determined based on the SI, AMo and LF/HF: a) SI≤50 c.u.; AMo≤ 30%; LF/HF<1.5 c.u. – predominance of parasympathetic activity (type I,

vagotonic); b) $51 \le SI \le 150$ c.u.; $31 \le AMo \le 50\%$; 1.5 $\le LF/HF \le 2.0$ c.u. – vegetative balance (type II, normotonic); c) SI>150 c.u.; AMo>50%; LF/HF>2.0 c.u. – predominance of sympathetic activity (type III, sympathotonic).

SPSS Statistics 16.0 (Chicago, IL, USA) was used for statistical analysis. Values were displayed as mean \pm standard deviation (M \pm SD). Analysis of continuous variables was performed using 2sample *t*-test and ANOVA was used to compare means among the 3 groups. Pearson's correlation coefficient (r) comparison was used to determine linear associations between numerical variables. *P* values less than or equal to 0.05 were considered statistically significant in paired comparisons. *P* values less than or equal to 0.01 were considered to be statistically significant in multiple comparisons.

Results

Data analysis of time domain parameters of HRV demonstrated that in the period of exam, stu-

dents presented significantly higher HR (94.81 bpm) and lower SDNN (38.29 ms) compared to the pre-exam period (P=0.001), as well as significantly higher Mo (645.65 ms) than during the examination phase (574.93 ms). However, after the exam AMo and SI significantly increased compared to before exam from 41.82% to 48.72% and from 180.53 c.u. to 252.91 c.u. respectively (P= 0.001). These changes denoted an increase activity of SNS and a decrease in PSNS one after exam.

Concerning frequency domain, TP was significantly higher before the exam (TP: 4034 ms²) than after the examination time (TP: 2569 ms², P=0.001), as well as significantly higher VLF, LF and HF components (VLF: 1449 ms², LF: 1638 ms², HF: 947 ms²) compared to during the exam (VLF: 881 ms², LF: 1145 ms², HF: 541 ms², P <0.05). The LF/HF ratio was 2.02 c.u. in the preexamination period and determined the balance in ANS activity of the heart, while after the exam shifted towards the predominance of SNS activity acting on it (LF/HF: 2.42 c.u., P = 0.05). All these findings together are reflective of the dynamic changes in the physiology of the heart under real life stress conditions (Table 2).

Variables	Before exam (BE) (n=70)	After exam (AE) (n=70)	P-Value
HR, bpm	87.09±10.73	94.81±8.61	0.001*
SDNN, ms	46.75±17.18	38.29±13.45	0.001*
Mo, ms	645.65±136.07	574.93±131.98	0.001*
AMo, %	41.82±12.23	48.72±12.42	0.001*
SI, c.u.	180.53 ± 13.61	252.91±16.07	0.001*
TP, ms ²	4034±342	2569 ± 228	0.001*
VLF, ms ²	1449±137	881±102	0.003*
LF, ms ²	1638±176	1145±107	0.013*
HF, ms ²	947±187	541±145	0.001*
LF/HF, c.u.	2.02±1.14	2.42±1.13	0.025*

Table 2: Time domain and frequency domain parameters of HRV measured before and after the exam

Data are presented as mean \pm standard deviation, *-significance of differences at P < 0.05 in paired comparisons (Before and After exam)

The distribution of students into groups in preexamination period according to SI, AMO and LF/HF values revealed for normotonic students (type II, 33% of participants) the initial values of time domain parameters of HRV: SDNN, Mo, AMo, SI corresponded to the indicators of healthy people and were equal to the norm. In the vagotonic group (type I, 14% of participants) parameters of HRV responsible for the parasympathetic activity, such as SDNN and Mo, had the highest values, while AMo and SI, specify sym-

pathetic activity, were the lowest compared to the group of sympathotonic. However, in the sympathotonic group time domain parameters of HRV had the opposite picture: the lowest values of SDNN and Mo (32.23 ms and 608.83 ms respectively, P = 0.001) and the highest values of AMo and SI (52.42% and 292.58 c.u. respectively, P = 0.001), thereby confirming the predominance of sympathetic activity in the regulation of heart rhythm in this group of students (Table 3).

 Table 3: Time domain parameters of HRV measured before and after the exam based on type of vegetative regulation

Variables		Type I predominance of PSNS (vagotonic) (n=10)	Type II vegetative balance (normotonic) (n=23)	Type III pre- dominance of SNS (sympatho- tonic) (n=37)	P-Value
HR, bpm	BE	73.86±4.06*	79.94±9.70*	92.85±7.79*	0.001**
	AE	90.12±9.54*	90.94±9.70*	96.96±6.75*	0.034
SDNN, ms	BE	82.43±8.62*	54.44±8.22*	32.23±7.16	0.001**
	AE	$50.57 \pm 6.55 *$	44.12±17.12*	30.96 ± 8.43	0.001**
Mo, ms	BE	817.71±87.93*	613.17±14.12*	608.83±10.63	0.001**
	AE	615.81±10.26*	528.04±16.64*	524.06±10.61	0.234
AMo, %	BE	23.29±2.14*	33.69±6.64*	52.42±8.24	0.001**
	AE	36.71±5.06*	45.31±16.25*	53.51±10.65	0.005**
SI, c.u.	BE	39.12±7.11*	94.31±31.77*	292.58±14.75	0.001**
	AE	126.57±55.14*	215.19±16.02*	331.08±17.92	0.007**

Data are presented as mean \pm standard deviation, * - significance of differences at *P*<0.05 in paired comparisons (Before and After exam); ** -significance of differences at *P*<0.01 in multiple comparisons (between groups: type I, type II and type III)

After the exam, significant changes occurred in groups of vagotonic and normotonic students: decreased SDNN (50.57 ms and 44.12 ms respectively, P < 0.05) and Mo (615.81 ms and 528.04 ms respectively, P < 0.05), increased AMo (36.71% and 45.31% respectively, P < 0.05) and SI (126.57 c.u. and 215.19 c.u. respectively, P < 0.05)

HR, bpm SDNN, ms 📕 Before exam 📕 After exam 📕 Before exam 📕 After exam 120 120 100 100 80 80 60 60 40 40 20 20 0 0 Type I Туре П Type III Type I Type II Type III Mo, ms SI, c.u. Before exam After exam 📕 Before exam 📕 After exam Ξ 1200 700 1000 600 500 800 • 400 600 300 400 200 200 100 0 0 Type I Туре П Type III Type II Type III Type I



Box plot represented changes of time domain parameters of HRV among the healthy 70 male undergraduates before and after exam, clustered based on type of vegetative regulation with represent outliers and extreme outliers respectively in the displayed data.

0.05), indicating a shift in vegetative balance to-

wards the predominance of the sympathetic ac-

tivity. In the sympathotonic group of students we

did not find any significant change in their time

domain parameters of HRV, other than HR,

which significantly increased in all groups of stu-

dents and became above 90 bpm (Fig. 1).

The analysis of frequency domain parameters of HRV showed the reliability of differences before exam in all three groups of students (Table 4).

Variables		Type I predominance of PSNS (vagoton- ic) (n=10)	Type II vegetative balance (normotonic) (n=23)	Type III pre- dominance of SNS (sympatho- tonic) (n=37)	P-Value
TP, ms ²	BE	11403±265*	4869±210	1565±688	0.000**
	AE	4267±107*	3723±321	1529 ± 87	0.001**
VLF, ms ²	BE	3934±149*	1581±713	576 ± 32	0.000**
	AE	1185±313*	1276±132	541±43	0.016
LF, ms ²	BE	4144±116*	2010 ± 121	649±30	0.000**
	AE	1876±715*	1785±174	665 ± 38	0.002**
HF, ms ²	BE	3325±784*	1278±774*	339±19	0.000**
	AE	1206±620*	663±56*	324±22	0.000**
LF/HF,	BE	1.32 ± 0.46	$1.78 \pm 1.04 *$	2.21 ± 0.98	0.068
c.u.	AE	2.08 ± 1.45	$2.60 \pm 0.93 *$	2.52 ± 1.04	0.543

 Table 4: Frequency domain parameters of HRV measured before and after the exam based on type of vegetative regulation

Data are presented as mean \pm standard deviation, * - significance of differences at P < 0.05 in paired comparisons (Before and After exam); ** -significance of between groups: type I, type II and type III). Differences at P < 0.01 in in multiple comparisons

The table 4 demonstrated that before the exam the TP of vagotonic was highest (TP: 11403 ms²) compared with normotonic (TP: 4869 ms²) and sympathotonic (TP: 1565 ms²). The group of vagotonic students also had the highest values of VLF, LF and HF components, while sympathotonic had the lowest values of VLF, LF and HF. In the group of normotonic all frequency domain parameters of HRV were within the limits of the normal values. The LF/HF corresponding ratio, which interpret to mean of sympathetic to parasympathetic activity in vagotonic was 1.32 c.u. and determined the predominance of parasympathetic activity, in normotonic was 1.78 c.u. and correlated with vegetative balance, while in sympathotonic it was 2.21 c.u. and indicated the predominance of sympathetic activity. After the exam, frequency domain parameters of HRV were not significantly changed in the groups of normotonic and sympathotonic students. However, in the vagotonic group frequency domain parameters, such as TP, VLF, LF and HF significantly decreased but did not go out of normative corridor, because of their highest initial values. The ratio of LF/HF during the exam did not significantly change, but shifted in the vagotonic group towards vegetative balance (LF/HF: 2.08 c.u.) and in the normotonic group towards the sympathetic link of regulation (LF/HF: 2.52 c.u.) (Fig. 2).



Fig. 2: Changes of frequency domain parameters of HRV measured before and after the exam based on type of vegetative regulation

Box plot represented changes of frequency domain parameters of HRV among the healthy 70 male undergraduates before and after exam, clustered based on type of vegetative regulation with represent outliers and extreme outliers respectively in the displayed data.

Correlation analysis between time and frequency domain parameters of HRV allowed us to determine the existence of negative HF links with SI (r= -0.661) and AMo (r= -0.762) specify the activity of SNS, and a positive correlation with SDNN (r=0.845) reflecting the activity of the PSNS. Correlations were also found between VLF and SI (r= -0.579), VLF and AMo (r= -0.676), VLF and SDNN (r=0.792). The calculation of the correlation coefficients of LF with SI (r=0.594) and AMo (r=0.702) demonstrated the presence of positive associations and negative correlation with SDNN (r=-0.881).

Distribution of students before and after the exam based on type of vegetative regulation showed that on a routine academic period the majority of students belonged to sympathotonic (53%), vagotonic and normotonic were 14% and 33% respectively, during the examination period the number of sympathotonic increased to 74%, however the number of vagotonic and normotonic decreased to 4% and 22% respectively (Fig. 3).



Fig. 3: Distribution of students before and after the exam based on type of vegetative regulation

Thus, the predominance of SNS was clearly manifested in 2/3 of the students. That is, mental stress was expressed in a more "active" form, targeting the students to overcome the difficulties.

Discussion

The objective of this study was to define the changes of time and frequency domain parameters of HRV before and after exam among foreign students, where examination is considered a normal source of stress in the academic life at IHSM. Moreover, it was of interest to determine the individual typological features of vegetative regulation response in medical students to exam stress during the educational process.

To achieve these targets, participants were monitored 2 times: 3 weeks before the examination session in routine academic conditions and 10-15 minutes after their exam was done.

Generally, after the exam, students had lower HRV compared to before the exam, with a decrease in PSNS activity, reflected by the depression in SDNN and Mo, at the same time an increase in SNS activity, as evidenced by an elevation in HR, AMo and SI (18,19,27). It should be

noted, that during the exams, SDNN was 36% lower than the normal values, according to A.N. Fleishman, at SDNN<50 ms, there is a significant tension of the regulatory mechanisms and high risk of developing cardiovascular disorders when central regulatory mechanisms are involved in the adaptation process (3,27). After the exam, the decrease in Mo and increase in AMo was due to the "mobilizing" effect of sympathetic influences on the heart rate (2,3,27). On a routine academic day, SI was by 20% higher than the normal level and on examination time increased by another 40%. Spectral analysis of HRV, which determine the modulatory effects of neural mechanisms on the sinus node (2), in our study demonstrated, that in non-exam period among the medical students presented higher frequency domain parameters TP, VLF, LF and HF, however in examination period, these indicators were significantly lower, which were same to that revealed in another studies (20,28,29). So, TP, as a characteristic of the adaptive potential of the body (30), decreased by 36% during the exams and was below the normal values. While the lowfrequency components (VLF and LF), which are considered as a marker of sympathetic modulation (31), decreased by 40% and 30% respectively, but these shifts did not go beyond the normative corridor. The HF component reflecting vagal activity, significantly decreased by 42% and was below the normal values, showing the suppression of parasympathetic effects on the heart rate (32). The value of LF/HF was significantly increased by 22% after the exam, indicating a higher sympathetic input compared to parasympathetic input of the ANS (33).

This research results demonstrated that during the exams there was significant change in the parasympathetic to sympathetic activity, which leads to a pronounced tension of regulatory mechanisms aimed to successfully pass exams. The conducted study on the assessment of the effect of exam stress on HRV indicated that the results are similar to those revealed by previous investigations (19,22,34-37). However, in the course of the research, the international students clearly showed individual body reactions to exam stress, which differed from the average values of the general sample. Averaged data "lubricate" important patterns of response (38) and features of adaptive rearrangements in the process of educational activity. In this regard, we had differentiated the medical students into groups with different levels of vegetative regulation based on the initial tone and evaluated the changes of the main time domain and frequency domain parameters of HRV to the examination situation. The stress index (SI), which characterizes the tension of regulatory systems, and ratio LF/HF, which reflects the balance or predominance of the sympathetic or parasympathetic link of the ANS, was chosen as a criterion of vegetative balance.

Distribution of students based on type of vegetative regulation showed that on a routine academic period the majority of students belonged to sympathotonic (53%), vagotonic and normotonic were 14% and 33% respectively. During the examination period the number of sympathotonic increased to 74%, however the number of vagotonic and normotonic decreased to 4% and 22% respectively. Analysis of the average HRV parameters by groups before the exam revealed significant differences in the main features used to assess the sympathetic (AMo, SI) and parasympathetic nervous systems (SDNN, Mo). Thus, for normotonic students the initial values of time domain parameters of HRV (SDNN, Mo, AMo, SI) corresponded to the values of normal healthy individuals in relative rest condition. Such individuals are characterized by a balanced influence of both the sympathetic and parasympathetic links of the ANS, and the state of satisfactory adaptation (39).

SDNN in the group of vagotonics exceeded by 34% and 60% respectively compared with normotonics and sympathotonics. The highest SDNN values indicated an increased the role of ANS, especially the parasympathetic division, in the regulation of heart rhythm (2,30,39). While the lowest SDNN indicators provided prognostic information in the identification of individuals at risk for various cardiovascular disorders and indicated a significant stress on regulatory systems (2,27,30,39). The pronounced tension of the regulatory mechanisms in sympathotonic students can also be judged by the SI parameters, the value of which was higher by 62% and 84% respectively compared with normotonic and vagotonic. Several studies had reported that the excessive predominance of sympathetic influences in the regulation of heart rate reflected the state of chronic stress in the body and indicated the state of unsatisfactory adaptation (28,39).

After the exam, there was significant difference in time domain parameters of HRV among normotonic and vagotonic students, but in the group of sympathotonic no significant change in HRV. Once the exam was over, SDNN significantly decreased by 20% and 39% respectively in normotonic and vagotonic groups, indicating a reducing of the parasympathetic activity (2,39), but in same groups of students AMo were increased by 11% and 13% respectively. In addition, similar to AMo, SI increased 2 and 3 times respectively from the initial values in the group of normotonics and vagotonics. These results indicated a shift in vegetative balance towards the predominance of the sympathetic activity, similar to results revealed by previous investigations (25,28), where in healthy young people, the reaction to exam stress, especially with intense mental stress, is accompanied by a significant increase in the SI.

In the group of sympathotonics, there was no significant increase in SI, this may be attributed perhaps to the fact of pronounced tension of regulatory systems at the pre-examination period. However, the spectral analysis of HRV showed the significant differences in the group of vagotonic, who had the highest values of TP and its components (VLF, LF and HF) in the preexamination period, which is typical for people with high adaptive capabilities of the body and resistant to adverse environmental factors (28,30,39,40). In the group of sympathotonic, where there was no significant difference in the values of frequency domain parameters of HRV compared before and after the exam, which may be due to excessive functional stress of the body even before the exams.

Conclusion

Dynamic monitoring of the functional state of students by the recording short term ECG with the determination of types vegetative regulation is very important for the prognostic value to identify high risk persons.

Journalism Ethics considerations

Ethical issues (Including plagiarism, informed consent, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc.) have been completely observed by the authors.

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Conflict of interest

The authors declare that there is no conflict of interest.

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