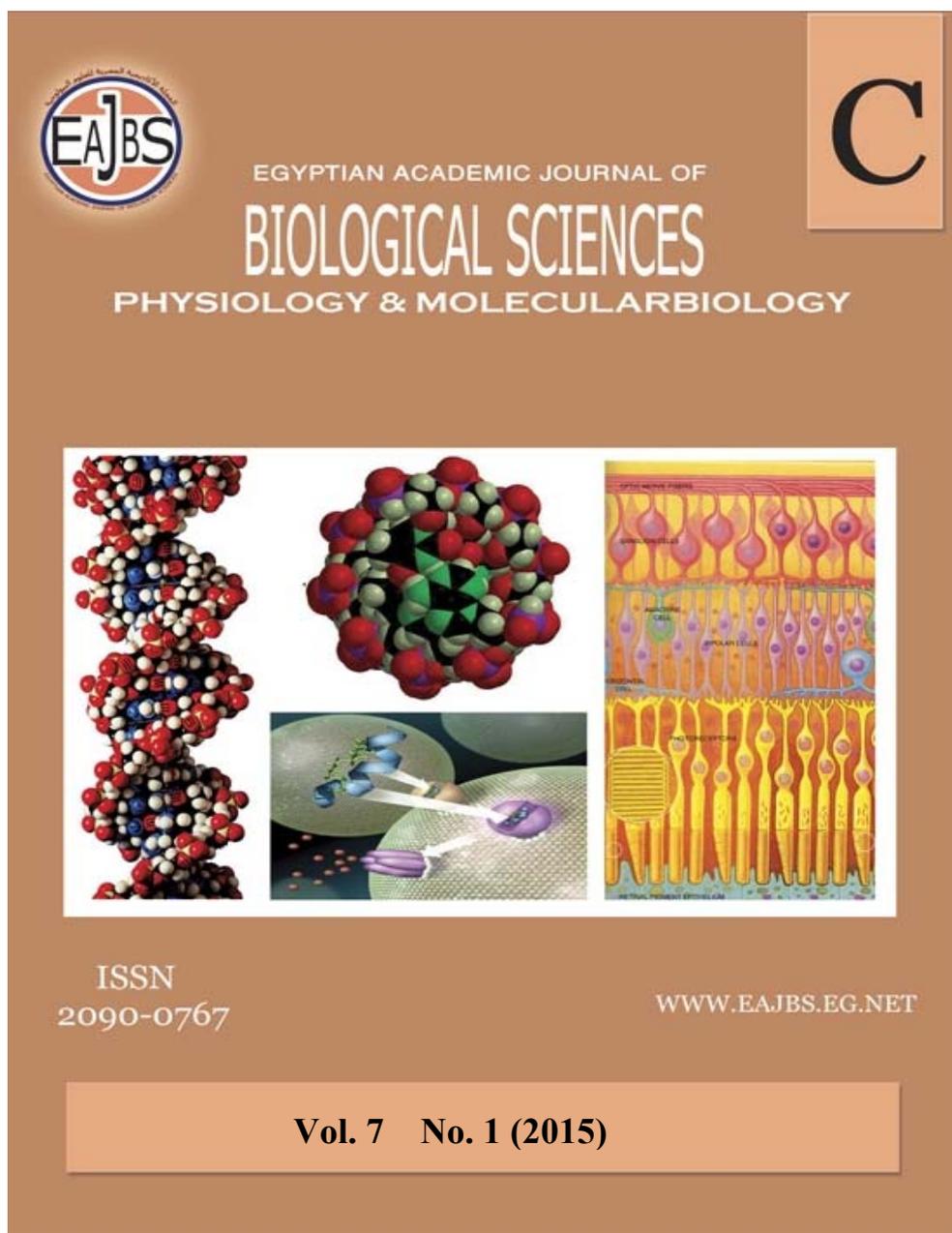


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Effect of Exam Stress on Heart Rate Variability Parameters in Healthy Students

El-Sayed A. M. Shokr

Physiology Department, Faculty of Medicine University of Ha'il, K. S. A.

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ABSTRACT

Since exam is characterized by stress-induced autonomic cardiac sensations, such as increased heart rate and palpitations, effects of experimental psycho-social stress on heart rate variability are of interest. Medical student usually study under high stress and its reported that most of medical students are more stressful than their colleagues in the other colleges. The time and frequency domain analysis of HRV were performed to assess the changes in sympathovagal balance in a group of 50 healthy students with normal electrocardiogram (ECG) at rest exposed to exam stress. The heart rate variability was recorded before and after exam. The frequency domain variables were computed using PowerLab® acquisition system: very low frequency (VLF) power, low frequency (LF) power, high frequency (HF) power and LF/HF ratio was determined. There was no significant difference in time and frequency parameters in the period before exam compared of the period after exam. This may suggest that adapted our student to exam than that the other student in another college. Because the medical students have many exams per term, so they adapted to exam and not affected by it. Frequent exams in the system-based curriculum help students to manage exam stress.

INTRODUCTION

Heart rate is mainly controlled by autonomic nerve activity to the sinoatrial node. Sympathetic and parasympathetic drive can be non-invasively investigated using Heart Rate Variability (HRV) analysis Altamura G. *et al.*, 1997. A low level of HRV associated with low vagal parasympathetic activity has been identified as a risk marker for all causes of mortality Appel ML, *et al.*, 1989. HRV can be altered by physiological factors, such as aging, gender and physical fitness. The aging process decreases HRV towards a lower parasympathetic modulation. Akselrod S, *et al.*, 1981 and Aubert A, *et al.*, 2001. Concerning gender, parasympathetic modulation of HRV seems to be generally higher in women than in men Barbaro V. *et al.*, 1995 and 1999 and Bigger J., *et al.* 1992. however, aging tends to attenuate this difference Bigger J., *et al.* 1992., the change apparently beginning at the menopause.⁷ Radiofrequency (RF) electromagnetic fields (EMF) of mobile phones are widespread in the living environment. The potential health risk of electromagnetic field emitted by mobile phones is still under debate. Exposition to high-power RF energy may have negative thermal effects on eye, skin and pregnancy Bortkiewicz A, *et al.*, 1996 and 2006, Dewhirst MW, *et al.*, 2003, Elder JA, 2003 and Evans JM, *et al.*, 2001.

Such negative effects have never been demonstrated at the power levels associated with public exposure to RF energy emitted by mobile phones. In this case the produced power is too low to cause the dangerous heating but there are few reports of non-thermal effect exerted by standard Global System for Mobile Communication (GSM). Bortkiewicz A, *et al.*, 1996, Fagard R, *et al.*, 1999 and Foster KR. 2000. Many reports suggest that electromagnetic fields emitted by cellular phones may interfere with work of cardiac pacemakers and other implantable medical devices. Hayes D, *et al.*, 1996 Heynick L, and Merritt J. 2003, Hillert L. *et al.*, 2006, Huikuri H, *et al.*, 1996 and Lerma C, *et al.*, 2006. There are some reports confirming so called non-thermic effect of mobile phones on humans that is not related to heat stress. Huikuri H, *et al.*, 1996. It was shown that occupational exposition to EMF can cause fluctuations in heart rate and heart rate variability (HRV). Huikuri H, *et al.*, 1996, Malliani A, *et al.* 1991 and Ministry of Environment 1998. It is possible that electromagnetic field emitted by cellular telephones may influence the autonomic tone, thus modifying the functioning of circulatory system.

The aim of the present study was to determine the influence of the exam on HRV in healthy young male medical students with a non invasive, widely used method of autonomic function evaluation. Also, System-based curriculum is less stressful to medical students.

METHODS

Subjects: A total 50 healthy male (21.0 ± 1.5 year) participants (from second and third year students, College of Medicine, University of Hail, Hail, KSA) were included in the study. All participants were healthy and none of them were on treatment. The following exclusion criteria were accepted for the

investigation: presence of any serious cardiovascular disease, including arterial hypertension, metabolic and neurological disorders that could influence heart rate variability and serious arrhythmias. All participants had exposed to many times exam per term. The mean numbers of exams are 4 to 5 per month; mean total duration of exam is 10 hours per month. The written informed consent was obtained from all students taking part in the study. The study was approved by the local ethical committee. All subjects got up between 4:30 a.m. and 7:00 a.m. and were asked to abstain from consuming caffeinated beverages and excessive physical activity including gymnastics within 10 hr preceding data collection. They were also requested not to eat and drink on the morning of the experiment. Students were fully habituated to equipment, protocols, and experimenters. Our investigation was performed in a semi-darkened, temperature-controlled quiet laboratory at room temperature (22°C). Before the experiment participants had rested in a laboratory room in a sitting posture for about 5 min. Records were performed between 08:00 and 10:00 in the morning in similar conditions (the same place of the experiment and sitting position) over 5 min periods.

Heart rate variability indices includes, (mean HR, mean R-R intervals, RMSSD and SDRR) as well as power spectral analysis (VLF, LF, HF, TP (total power), and LF/HF ratio) were obtained from short term (5 minutes) recording of ECG using Power Lab ®acquisition system.

ECG data analysis:

The ECG was sampled at 1000 Hz with the Power Lab ®acquisition system (AD Instruments Pty Ltd, Castle Hill, Australia) installed on IBM computer. Thus the accuracy of the measurements was 1 ms. The first minute of each ECG recording was disregarded to allow for

stabilization of the data prior to analysis. The detection of the QRS complex was conducted using the Gritzali's algorithm. RR interval sequence was defined by the duration between two consecutive R-peaks. These data were edited to eliminate any glitches, due to premature cardiac contraction. Each RR interval was visually validated by two experts before temporal and spectral analysis. Definitions and abbreviations for time domain analysis are shown in Table 1. For each RR sequence, three classical temporal parameters were then extracted : the mean RR, which represents mean HR; Standard deviation of RR intervals (SDRR), which reflects all the cyclic components responsible for variability in the period of recording, and RMSSD (Root mean square of successive RR intervals difference) between adjacent RR intervals, which is considered as an index of parasympathetic modulation of

HR. Prior to power spectrum density estimation, the RR sequence, which is intrinsically non-evenly spaced data, was linearly interpolated in order to obtain a series of uniformly sampled data. An interpretation of frequency contents of HRV was therefore possible independently of the mean RR value. The retained sampling rate was then set to 2 Hz. Using a sliding window of 64s duration, time-varying auto-regressive (TVAR) modeling of the interpolated RR sequence was performed to estimate its power spectrum (ms²) in order to eliminate the slight non-stationarities of the sequence. On the basis of the well-known Akaike information criteria, the order of the TVAR model was set to 12. The spectrum is divided into three bands as the following: very low frequency (0 – 0.05 Hz), low frequency (0.05 – 0.15 Hz), high frequency (0.15 – 0.5 Hz) and total power (0 – 0.5 Hz).

Table 1: Definitions and abbreviations for time domain analysis of heart rate variability.

Variable	Unites	Definition
SDRR	Ms	Standard deviation of normal RR intervals
MHR	Beats/min	Mean Heart rate
SNN50	Ms	The number of time that the difference between adjacent normal RR intervals greater than 50 ms, computed over the entire 24-hour recording
RMSSD	Ms	Root mean square of successive RR intervals difference: the square root of the mean of the sum of the squares of the differences between adjacent normal RR intervals over the entire 24-hour ECG recording

Statistical analysis

The values are expressed as mean \pm SD. The statistical comparisons were performed by one way analysis of variance (ANOVA) followed by Duncan's multiple range test (DMRT), using SPSS version 15.0 for windows (SPSS Inc. Chicago; <http://www.spss.com>). The values are considered statistically significant if the p value was less than 0.05.

RESULTS

The present study revealed that the mean heart rate did not change

significantly over 15 min period before the exam (period) than that after the exam (period II) (Table 2). No arrhythmias were noted in the analyzed records before and after the exam setting. The analysis of the time domain HRV parameters in for the period I and II showed that SDNN was no significantly increase where the HRV before exam in comparison with period II after exam. The rest of the parameters of the time analysis measured within the particular periods of the investigation did not differ significantly from each other (Table 2).

Table 2: Time domain heart rate variability (HRV) parameters in 15-min intervals before exam (period I) and after exam (period II).

Parameters	Before exam (period I)	After exam (period II)	<i>p</i> values
MHR	136.80 ± 2.63	130.21 ± 3.32	<i>p</i> < 0.05
SDNN	255.72 ± 1.12	245.98 ± 4.77	<i>p</i> < 0.05
RMSSD	278.81 ± 1.09	266.54 ± 1.51	<i>p</i> < 0.05
NN50	54.54 ± 0.32	51.48 ± 0.40	<i>p</i> < 0.05

Values are expressed as mean ± SD for 25 young healthy subjects in each period. Different superscript denotes for significant differences between periods as analyzed by one-way ANOVA followed by DMRT (*p* < 0.05)

The analysis of the frequency domain HRV parameters demonstrated that VLF, and HF parameters were no significantly increased over the 15 minute period before exam in comparison with after exam (Table 3). LF was no significantly decreased during 15 min period after the exam compared with period I before exam. LF/HF ratio was also no significantly lower after exam in comparison with the period before exam (Table 3).

Table 3: Frequency domain heart rate variability (HRV) parameters in 15-min intervals before exam (period I) and after exam (period II).

Parameters	Before exam (period I)	After exam (period II)	<i>p</i> values
TP	4344.50 ± 248.71	4241.33 ± 75.69	<i>p</i> < 0.05
VLF	24411.00 ± 39.43	23540.17 ± 68.86	<i>p</i> < 0.05
LF	15365.17 ± 122.67	14683.67 ± 74.67	<i>p</i> < 0.05
HF	8366.33 ± 61.19	7722.83 ± 120.66	<i>p</i> < 0.05
LF/HF	2.25 ± 0.03	1.90 ± .03	<i>p</i> < 0.05

Values are expressed as mean ± SD for 25 young healthy subjects in each period. Different superscript denotes for significant differences between periods as analyzed by one-way ANOVA followed by DMRT (*p* < 0.05)

DISCUSSION

The present study revealed that the exam may cause the increase in parasympathetic tone (the highest values of HF parameter were noted before the exam) and the decrease in sympathetic tone (the lowest values of LF/HF ratio after the exam). It is known that the efferent vagal activity is a major contributor to the HF component. Pomeranz M, *et al.*, 1985, Szmigielski S. *et al.*, 1998, Task Force, 1996, Taylor J, *et al.*, 1998 and Tropea and Lee 1992. On the other hand, LF is a marker reflecting both sympathetic and vagal activity^{26,27} and the LF/HF ratio is considered to mirror sympathovagal balance or reflect the sympathetic modulations. SDNN parameters were the

highest before the exam. SDNN represents joint sympathetic and parasympathetic modulation of heart rate Pickard W. and Moros E. 2001. These results were in agreements with previous published data.¹⁹

In our study, VLF increased before the exam. The increase in very low frequency in the exposed subjects could be related to parasympathetic activation as VLF is very much dependent on parasympathetic tone. Pomeranz M, *et al.* 1998, suggested that parasympathetic nervous system is the dominant determinant of VLF Vikman S, *et al.*, 2003. In studies on atrial fibrillation (AF) the increase in VLF component together with other parasympathetic markers predicted the

early recurrence of AF after cardioversion Wranicz J, *et al.*, 2004. However, the physiologic interpretation of VLF oscillations is still a subject of debate. Reduction in VLF is associated with increased risk for sudden cardiac death.³⁰ Different physiological mechanisms for VLF have been proposed: physical activity, thermoregulation, renin-angiotensin-aldosterone system, slow respiratory patterns and parasympathetic mechanisms Vikman S, *et al.*, 2003.

Regarding widespread use of exam to all students in all stages of their age for a long time. On the other hand, it is important to evaluate whether the extensive use of exam in various types of jobs could exert influence on heart that is not only related to mental stress Taylor J, *et al.*, 1998. Results of our investigation suggest that repeating of exam may exert a noticeable effect on autonomic balance, though the pattern it represents is not typical for the deteriorious effect on HRV, i.e. lack of a typical decrease in parasympathetic activity with the domination of sympathetic system. The increase in vagal activity can be beneficial in cardiovascular diseases. However, it has not been elucidated yet how much vagal activity or its markers have to increase in order to provide adequate protection or how the proper balance between parasympathetic and sympathetic tone should be expressed Tropea and Lee 1992. Thus, results of our investigation show that the repeating of exam can have a positive health effect. This is a preliminary study to demonstrate that the exam may cause changes in autonomic balance probably related to a non-thermal bioeffect. Further study may be needed to investigate the how much increase in HRV can be beneficial in cardiovascular diseases.

CONCLUSIONS

In conclusion, the above results showed that the exam may influence heart rate variability and change the autonomic balance. The increases in the parasympathetic tone concomitant with the decrease in the sympathetic tone measured indirectly by analysis of heart rate variability were observed before and after exam. Changes in heart rate variability observed before and after exam could be affected by panic of the students from the exam. The aim of the present study was to determine the influence of the exam on HRV in healthy young male medical students with a non-invasive, widely used method of autonomic function evaluation. Also, System-based curriculum is less stressful to medical students. Frequent exams in the system-based curriculum help students to manage exam stress. So, they adapted to the exam and the results show that.

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REFERENCES

- Altamura G, Toscano S, Gentilucci G, Ammirati F. (1997). Influence of digital and analogue cellular telephones on implanted pacemakers. *Eur Heart*,18: 1632–1641.
- Appel ML, Berger RD, Saul JP, Smith JM, Cohen RJ. (1989). Beat to beat variability in cardiovascular variables: noise or music? *J Am Coll Cardiol*, 14:1139–48.
- Askelrod Akselrod S, Gordon D, Ubel FA, Shannon DC, Barger AC, Cohen RJ. (1981). Power spectrum analysis of heart rate fluctuation: a quantitative probe of beat to beat cardiovascular control. *Science*, 213:215–2.

- Aubert A, Beckers F, Ramaekers D. (2001). Short-term heart rate variability in young athletes. *J Cardiol* 37 Suppl, 1:85–8.
- Barbaro V, Bartolini P, Donato A, Militello C. (1999). Electromagnetic interference of analog cellular telephones with pacemakers: in vitro and in vivo studies. *Pace*, 22:626–634.
- Bigger J, Fleiss J, Steinman R, Rolnitzky L, Kleiger R. and Rottman J. (1992). Frequency domain measures of heart period variability and mortality after myocardial infarction. *Circulation* 8:164–71.
- Bigger J, Fleiss J, Steinman R, Rolnitzky L, Schneider W, Stein K. (1995). RR variability in healthy, middleaged persons compared with patients with chronic coronary heart disease or recent acute myocardial infarction. *Circulation* 91:1936–43.
- Bortkiewicz A, Gadzicka E, Zmys'lonny M, Szymczak W. (2006). Neurovegetative disturbances in workers exposed to 50 Hz electromagnetic fields. *Int J Occup Med Environ Health* 19:53–60.
- Bortkiewicz A, Gadzicka E, Zmyslonny M. Heart rate variability in workers exposed to medium-frequency electromagnetic fields. *J Auton Nerv Syst* 1996; 59:91–97.
- Dewhirst MW, Viglianti BL, Lora-Michiels M, Hanson M, Hoopes PJ. Basic principles of thermal dosimetry and thermal thresholds for tissue damage from hyperthermia. *Int J Hypertherm* 2003; 19:267–294.
- Elder JA. Ocular effects of radiofrequency energy. *Bioelectromagnetics* 6 (Suppl.) 2003;148–161.
- Evans JM, Ziegler MG, Patwardham AR. Gender differences in autonomic cardiovascular regulation: spectral, hormonal, and hemodynamic indexes. *Journal of Applied Physiology* 2001; 91:2611–2618.
- Fagard R, Pardaens K. and Staessen J. (1999). Influence of demographic, anthropometric and lifestyle characteristics on heart rate and its variability in the population. *J Hypertens* 11:1589–99.
- Foster KR. (2000). The mechanism paradox: constraints on interactions between radiofrequency fields and biological systems. In: Moriarty M, Mothersill C, Seymour C, Edington CM, Ward JF, Fry RJM (Eds.) 222–6, Allen Press, Inc, Lawrence.
- Hayes D, Carrillo R, Findlay G, Embrey M. (1996). State of the science: pacemaker and defibrillator interference from wireless communication devices. *Pace* 19: 1419–1430.
- Heynick L, Merritt J. (2003). Radiofrequency fields and teratogenesis. *Bioelectromag* 6 (Suppl.) 174–186.
- Hillert L, Ahlbom A, Neasham D, Feychting M, Jarup L, Navin R, Elliott P. Call-related factors influencing output power from mobile phones. *J Expo Sci Environ Epidemiol* 2006; 16, 507–14.
- Huikuri H, Pikkujamsa S, Airaksinen K, Ikaheimo M, Rantala A, Kauma H, Lilja M, Kesaniemi Y. Sex related differences in autonomic modulation of heart rate in middle-aged subjects. *Circulation* 1996; 94:122–5.
- Lerma C, Vallejo M, Urias K, Hermosillo AG, Cardenas M. Differences in cardiac autonomic modulation between women and men. *Arch Cardiol Mex* 2006; 76:277–82.
- Malliani A, Pagani M, Lombardi F, Cerutti S. Cardiovascular neural regulation explored in the frequency domain. *Circulation* 1991; 84:1482–92.
- Ministry of Environment, Poland Dz.U. nr 107 poz. 676. Protection against electromagnetic fields 1998, Poland (in Polish).

- Pickard W, Moros E. Energy deposition processes in biological tissue: nonthermal biohazards seem unlikely in the ultra-high frequency range. *Bioelectromag* 2001; 22: 97–105.
- Pomeranz M, Macaulay RJB, Caudill MA, Kutz I, Adam D, Gordon D, Kilborn KM, Barger AC, Shannon DC, Cohen RJ, Benson M. (1985). Assessment of autonomic function in humans by heart rate spectral analysis. *Am J Physiol*, 248: 151–163.
- Szmigielski S, Bortkiewicz A, Gadzicka E, Zmyslony M, Kubacki R. Alteration of diurnal rhythms of blood pressure and heart rate to workers exposed to radiofrequency electromagnetic fields. *Blood Press Monit* 1998; 3:323–330.
- Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology. Heart rate variability: standards of measurement, physiological interpretation, and clinical use. *Circulation* 1996; 93:1043–65.
- Taylor J, Carr D, Myers C, Eckberg D. Mechanisms underlying very low frequency RR-interval oscillations in humans. *Circulation* 1998; 98:547–55.
- Tropea B, Lee R. Thermal injury kinetics in electrical trauma. *J Biomech Eng* 1992; 114:241–250.
- Vikman S, Mäkikallio TH, Yli-Mäyry S, Nurmi M, Airaksinen KE, Huikuri HV. Heart rate variability and recurrence of atrial fibrillation after electrical cardioversion. *Ann Med* 2003; 35: 36–42.
- Wranicz J, Rosiak M, Cygankiewicz I, Kula P, Kula K, Zareba W. Sex steroids and heart rate variability in patients after myocardial infarction. *Ann Noninvasive Electrocardiol* 2004; 9:156–61.

ARABIC SUMMERY

تأثير ضغط الامتحان قياس التغيرات في ضربات القلب للطلاب الاصحاء

السيد احمد محمد شكر

قسم الفسيولوجي- كلية الطب- جامعة حائل

تتميز الامتحانات من خلال الأحاسيس التي تسببها من الإجهاد القلبي اللاإرادي، مثل زيادة معدل ضربات القلب والخفقان، وآثار الإجهاد النفسي والاجتماعي الذي يؤدي الى تقلب معدل ضربات القلب وخصوصا لطالب الطب الذي يدرس عادة تحت الضغوط العالية والتي بلغت أن معظم طلاب الطب هي أكثر إرهاقا من زملائهم في الكليات الأخرى. أجريت التحاليل الوقت والتردد مجال الهريفي لتقييم التغيرات في التوازن العصبي في مجموعة من الطلاب ذو صحة جيدة وتوصيلهم بجهاز قياس التغيرات في ضربات القلب تم تسجيل تقلب معدل ضربات القلب قبل وبعد الامتحان. حسب التغيرات في مجال التردد باستخدام نظام الحصول PowerLab®: تم تحديد الترددات المنخفضة جدا (VLF) ، التردد المنخفض (LF) ، عالية التردد الطاقة (HF) ونسبة HF / LF. تم تسجيل ECG في ظروف موحدة من ٨ حتي ٩ صباحا في وضعية الجلوس، في غضون ١٥ دقيقة. لم يكن هناك فرق كبير في التغيرات لضربات قلب الطلاب قبل و بعد الامتحانات. وهذا قد يشير إلى أن طلاب الطب قد تكيفوا لاداء الامتحانات عن الطلاب في الكليات الأخرى. لأن طلاب الطب لديهم العديد من الامتحانات لكل فصل دراسي لذاحدث لهم نوع من التكيف على اداء الامتحانات و لم يعد لديهم الرهبة و الخوف الموجود لدى الطلاب الاخرين الموجودين بباقي كليات الجامعة ذو المناهج التقليدية حيث انهم يؤدو امتحان واحد او امتحانين لكل ترم.

بينت النتائج ان الامتحانات المتكررة لدى طلاب كلية الطب ادت الى تكيفهم و عدم خوفهم من الامتحانات مختلفين عن باقي كليات الجامعة ذو المناهج التقليدية حيث انهم يؤدو امتحان واحد او امتحانين لكل ترم. الاعداد الجيد للمناهج يمكن أن تساعد الطلاب على التحكم في التوتر لديهم.