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Cardiovascular reactions to exam situations

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The aim of this study was to examine whether the parameters of cardiac R-R intervals reflect the changes in the emotional and mental components of stress during a difficult and an easy exam. Twelve subjects, 18 to 19 years of age, with no previous experience with exams at university level, took part in the study. The levels of anxiety, high activation and exam apprehension were assessed before a difficult and an easy exam. Subjects' cardiac R-R intervals were continuously registered in the period of five minutes before the exam, during the whole exam, and five minutes after the exam by the Power Lab polygraph. The level of anxiety, high activation and exam apprehension were higher before the difficult exam than before the easy exam. Shorter and more regular R-R intervals were found during the difficult exam as compared to the easy exam. No significant differences in the spectral analyses parameters were found between the difficult and the easy exam, while the differences were significant between the pre-exam, exam and the post-exam periods.

Key words: exam stress, heart rate variability, spectral analysis, exam difficulty

Different investigations have shown the stressfulness of exam periods and exam situations, manifested in increase of anxiety, depressiveness and negative emotional states as the exam approaches (Ellis & Fox, 2004; Gruzelier, Smith, Nagy, & Henderson, 2001; Laidlaw, Naito, Dwivedi, Enzor, & Brincat, 2003; Oaten & Chang, 2005; Ogden & Mtandabari, 1997; Vivian, Koh, & Chia, 2003). Apart from these reactions, it has been shown that immunological and some other physiological functions become impaired during exam periods (Al-Kubati, Fišer, & Siegelova, 1997; Glaser, Pearl, Kiecolt-Glaser, & Malarkey, 1997; Lewis, Nikolova, Chang, & Weekes, 2008; Marsall, Agarwal, Cohen, Henninger, & Morris, 1998). The effects of this impairment were high incidences of respiratory infections (Deinzer & Schuller, 1998) and abdominal pains (Harris & Martin, 1994). Students who reported higher stress before the exam also had higher cortisol level and lower examination scores (Vivian et al., 2003).

Some investigations included continuous follow up of changes in psychophysiological variables which are known indicators of stress level (Huwe, Henning, & Netter, 1998). The results showed an increase in bad mood, lack of energy, bodily discomfort, somatic strain and restlessness during the pre-exam period, with the peak immediately before the exam. The cortisol level and heart rate, which were monitored immediately before and after exams, showed a significant increase after the exam in comparison with the pre exam levels (Huwe et al., 1998).

There is a paucity of studies, however, with recordings of physiological reactions during the exam itself. Two such studies with heart rate monitoring showed that oral exams were more stressful than written exams (Spangler, 1997), and first year medical students having higher heart rates during oral exams than their colleagues in the second year (Carreras & Fernandez-Castro, 1998). These differences in heart rates between the two groups were interpreted as a better adaptation to the exam stress of the second year students. In this study, authors did not control the exam difficulty.

Some studies have related changes in heart rate to emotional reactions or emotional component of stress (Lipman, Grossman, Bridges, Hamner, & Taylor, 2002; Šimić, 2006). Heart rate itself, however, does not clearly reflect changes in mental load, which is one of the major components of stress during exams. Although heart rate is a good indicator of overall stress, which includes reactions to the task difficulty (physical and mental components), as well as emotional reactions, for finer analyses so called *sinus arrhythmia* changes seem more suitable. Sinus arrhythmia changes are easy to monitor *via* changes in cardiac R-R intervals, i.e. the time

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between two successive R peaks of the cardiac cycle. While resting, R-R intervals vary from about 500-1200 ms, while in working or stressful situations, they become shorter and more regular.

Results of some investigations have shown sinus arrhythmia to be a more sensitive indicator of mental load than the heart rate itself (Lee & Park, 1990; Kalsbeek, 1973). It was also shown that sinus arrhythmia decreases, if mental load increases during laboratory tasks of different kind (mental, psychomotor, repetitive) and complexity (Manenica, 1981; Manenica & Krošnjar, 1990; G. Mulder, 1973; L. J. M. Mulder, 1988; Reims, Sevre, Hoieggen, Fossum, & Kjeldsen, 2005; Slavić & Manenica, 2002). A number of authors obtained similar results in real situations, such as office work or on pilots during flights (Backs, Lenneman, & Sicard, 1999; Garde, Laursen, Jorgensen, & Jensen, 2002; Jorna, 1993).

Spectral analysis was used for identification of the effects of various stressors on people, such as paced working (Charnock & Manenica, 1978), and interference of biological rhythms, such as menstrual and circadian body temperature rhythm (Proroković, 1999). Spectral analysis is a mathematical procedure based on autocorrelation functions of the data time series, which separates frequencies of reoccurrence of various composite rhythms within time. The frequencies range in the R-R interval time series normally lies between 0.02-0.50 Hz. Some authors (Aasman, Mulder & Mulder, 1987; Keselbrener & Akselrod, 1998; L. J. M. Mulder, 1988) divided the frequency spectrum into three major bands: low frequency (LF) band between 0.02-0.06 Hz, medium frequency (MF) band between 0.07-0.14 Hz, and high frequency (HF) band between 0.15-0.50 Hz. Different spectral bands were not equally affected by the changes in mental load.

The LF band reflects changes in metabolic processes which regulate body temperature and activity of renin-angiotensin system (L. J. M. Mulder, 1988). The MF band appears to reflect changes in vasomotor system, which seem to be related to short-term regulation of arterial blood pressure, as well as mental load (Althaus, Mulder, Mulder, Roon, & Minderaa, 1998). The HF band reflects respiration pattern and some motor properties of the task, such as repetitiveness. A decrease of power in this band may be caused by changes in respiration rate patterns, or by a task-induced smaller vagal activity, or both (Althaus et al., 1998), as well as a decrease in respiration depth (Sato et al., 1998). Laboratory experiments, where mental arithmetic tasks, Stroop's Task and reaction time tasks were used, showed suppression of the power spectrum in the MF band as the task complexity increased (Aasman et al., 1987; Langewitz, Ruddel, Schumacher, Mulder & Shulte, 1990; L. J. M. Mulder, 1988; Sammer, 1998; Sloan, DeMeersman & Shapiro, 1997; Šiška, 2002) or when the task demands of pilots changed during flights (Veltman, 2002). There were, however, many studies on effects of mental load on parameters of R-R intervals, but very few paying attention to the emotional component in stressful situations.

Various investigations on students have shown that perceived stress increases as the exam approaches (Huwe et al., 1998; Laidlaw et al., 2003; Spangler, 1997; Šimić, 2006) without any references to the exam difficulty. Although the mean of R-R intervals or heart rate may be adequate overall indicators of stress, for finer analyses and possible separation of emotional and mental components of exam stress, variability and spectral indices should be used.

The aim of this study was to examine whether the parameters of R-R intervals reflect the changes in the emotional and mental components of stress during a difficult and an easy exam. Greater exam stress could be expected during the difficult than the easy exam, which would result in differences in the contribution of mental and emotional components to the overall stress. Furthermore, some kind of habituation to the exam situation could be expected, meaning a decrease in the stress during the course of the exam, which would be reflected on the parameters of R-R intervals.

METHOD

Subjects

Twelve female subjects, first year psychology students, 18 to 20 years of age, who had no previous experience with exams at university level, voluntarily took part in the study. All subjects reported to be free of chronic or acute cardiovascular, respiratory or other chronic disease.

Instruments

The level of anxiety was measured using Spielberger's State Anxiety Questionnaire (Spielberger, 2000), which consists of 20 statements. The subjects had to assess each statement on a 4-point scale in relation to how well it described their current state. The reliability coefficient (Cronbach Alpha) obtained on a sample of 531 female students was .93 (Spielberger, 2000). The range of scores was 20-80, higher scores meaning higher anxiety.

The subjects assessed their exam apprehension on a 21-point Borg's scale, where zero meant *not at all*, 10 meant *average* and 20 *extreme apprehension.*

Thayer's Activation-Deactivation Adjective Check List consists of 14 adjectives describing moods. Nine adjectives are related to *high activation*, which is synonymous to stress (Proroković, 2002). The subjects had to mark on a 5-point scale how well the adjectives described their current state. The reliability of the List (Cronbach Alpha) was .90 (Proroković, 2002; Proroković, Manenica & Gregov, 1996). The range of the results was 9-45, higher results meaning higher stress.

For continuous measurements and recordings of cardiac R-R intervals, the Power Lab Polygraph was used together with pre-amplifiers and electrodes.

Procedure

For the purpose of the study, a difficult and an easy exam were chosen out of eight exams of the first year psychology course. In preinvestigation, their difficulty was assessed by 60 older students (who had passed them) using the paired comparison method (Guilford, 1954), and results were expressed on z-scale. The rating of the difficulty in z-values was 1.37 for the difficult, and -2.27 for the easier exam.

The difficult exam was the course subject (*Biological Psychology*), which included 120 hours of teaching during the academic year, and it was obligatory for the enrolment in the second year. The easy exam was on an introductory subject (*Introduction to genetics*), which included 30 hours of teaching, and it was not obligatory to pass to enroll in the second year. The students took their exams during the regular exam terms with the in-between time interval not less than 15 days. The sequence of the exams was according to the subjects' choice, where seven subjects took the difficult exam first followed by the easy exam, and five subjects did it in the opposite order.

Since we dealt with naïve subjects, they underwent a process of habituation to the laboratory conditions, which started one month before the exam. It consisted of five measurements and recordings of R-R intervals during resting and during a psychomotor task performance, lasting for a half an hour. These measurements were done by the same experimenter with the same equipment and methodology, as it was used during the oral exam situations.

This study included measurement of anxiety levels, high activation and exam apprehension one hour before the exam. The recordings of the R-R intervals began five minutes before the exam, continued through the exam and finished five minutes after the exam. Procedures were the same for the difficult and the easy exam. During the exam itself, the subject was with the examiner in one room, while the equipment and the experimenter were in the other. The examiner was asked before the exam to press the *event marker button* to mark the beginning and the end of the exam. The recording session continued for five minutes after the exam. The durations of the exams were not the same, because the difficult exam lasted twenty minutes on average, and the easy exam ten minutes on average. For purpose of this study, first eight minutes period was analysed for every subject. Before starting, this study was approved by the University Research Ethics Committee.

Statistical analysis

The time series of R-R intervals were first analysed on a minute-to-minute basis before, during and after the exam, where the mean R-R intervals, standard deviation (SD) and DM index (DM) were calculated. DM index is the mean of absolute differences between successive R-R intervals. Both standard deviation and DM index are variability indices, where the first shows the variability of results around the mean, and the latter reflects the internal consistency (homogeneity) of the results. Standard deviation may be significantly affected by the drift of results in the course of time, while such drift does not affect the DM index. Since the two variabilities are important in this kind of data, it is recommendable to use both indices.

The time series of R-R intervals were also analyzed using spectral analysis, which gave individual spectrum frequencies before, during and after the difficult and the easy exam, over five minute periods. A fast Fourier transform was used to compute the power spectrum for LF (0.02-0.06 Hz), MF (0.07-0.14 Hz) and HF (0.15-0.50 Hz) bands. For the two exams, a spectral power parameter, squared modulation index (SMI index), was calculated for pre-exam, exam and post-exam periods, as well as for the three frequency bands. SMI indices are the sums of squared spectral densities, divided by the corresponding mean square of R-R intervals. They were also transformed to $\log_{10}$ values to reduce the skewness of distributions.

One-way ANOVA was performed to test the effects of exam difficulty on state anxiety, high activation and exam apprehension (Table 1). Two-way ANOVA was used for the analysis of effects of exam difficulty and minute-to-minute changes in the cardiac R-R interval parameters during the pre-exam, exam and post-exam periods (Table 2). The same procedure was used for the analysis of the effects of exam difficulty and periods (pre-exam, exam and post-exam) on spectral analyses parameters, i.e. the SMI indices (Table 3). Pearson's correlation coefficients were also calculated, where value of $p < .05$ was considered as statistically significant. All data were analyzed using the Statistica 7.0 software package.

RESULTS

The results showed significant differences in state anxiety, high activation and exam apprehension due to the exam difficulty (Table 1). They were at a higher level before the difficult than before the easy exam.

High correlations were found between general level of stress (high activation) and state anxiety before both exams ($r = .94$, $p < .001$, the difficult exam; $r = .92$, $p < .001$, the easy exam). There were also significant correlations between state anxiety and exam apprehension ($r = .70$, $p < .01$, the difficult exam; $r = .72$, $p < .01$, the easy exam).

Further analysis of results included a minute-to-minute analysis of the mean R-R intervals and the two variability parameters, for the periods before, during and after exams

Table 1
Pre-exam level of state anxiety, exam apprehension and high activation

Variables	Difficult exam ($M \pm SD$)	Easy exam ($M \pm SD$)	F(1,11)
State anxiety	48.25 ± 13.56	30.08 ± 9.14	24.15***
Exam apprehension	12.67 ± 4.31	5.08 ± 2.60	26.09***
High activation	27.58 ± 8.28	15.08 ± 5.70	35.02***

***p < .001

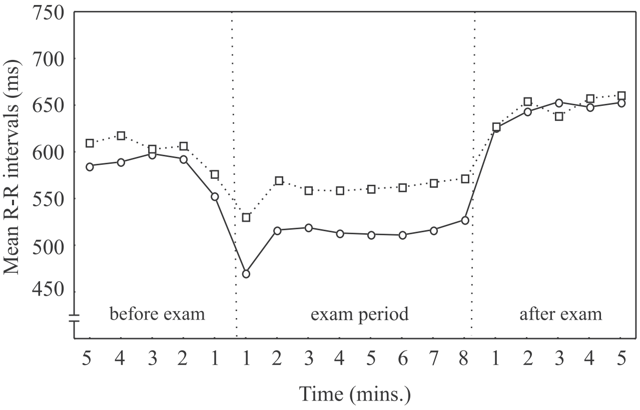

Figure 1. Changes in means of R-R intervals before, during and after the difficult exam (full lines) and easy exam (broken lines). Vertical lines divide pre-exam, exam and post-exam periods.

(Figure 1). The reason for this analysis was to see changes in the parameters due to habituation during the exam.

ANOVA showed significant differences in the means of R-R intervals between the difficult and the easy exam during the examination period, but not during pre-exam and post-exam periods (Table 2). R-R intervals were significantly shorter during the difficult exam (Figure 1).

There were no differences in variability parameters between the difficult and the easy exam, during pre-exam, as well as post-exam periods (Table 2). However, these differences were found during the examination period (Table 2).

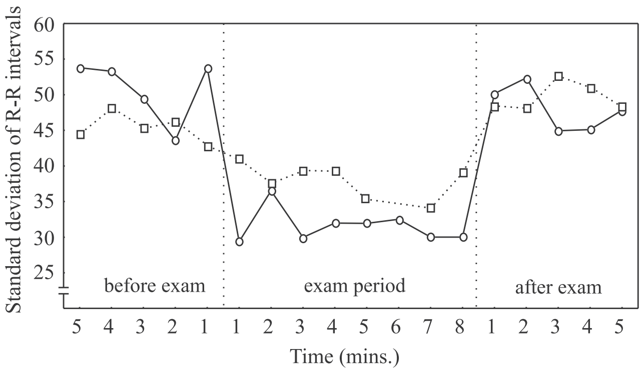

Figure 2. Changes in standard deviations of R-R intervals before, during and after the difficult exam (full lines) and easy exam (broken lines). Vertical lines divide pre-exam, exam and post-exam periods.

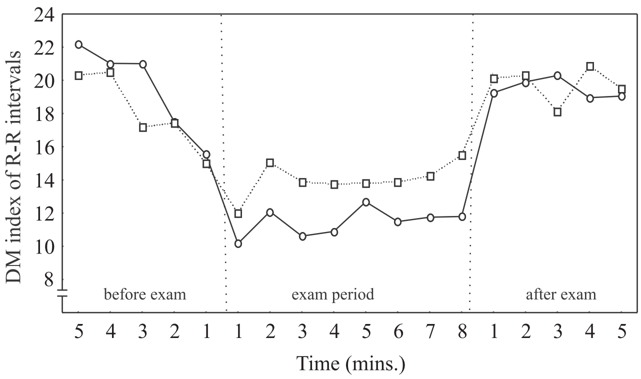

Figure 3. Changes in DM indices of R-R intervals before, during and after the difficult exam (full lines) and easy exam (broken lines). Vertical lines divide pre-exam, exam and post-exam periods.

Table 2
ANOVAs of minute-to-minute changes in cardiac parameters, done separately for each of the pre-exam, exam and post-exam periods

		Pre-exam period		Exam period		Post-exam period	
Variable	Source of variation	*df*	*F*	*df*	*F*	*df*	*F*
M of R-R intervals	Exam difficulty	1/11	0.57	1/11	5.03*	1/11	0.01
	Course of time (mins)	4/44	11.01**	7/77	5.52**	4/44	6.09**
SD of R-R intervals	Exam difficulty	1/11	0.93	1/11	4.75*	1/11	0.09
	Course of time (mins)	4/44	0.94	7/77	2.60*	4/44	0.15
DM-index of R-R intervals	Exam difficulty	1/11	0.28	1/11	4.61*	1/11	0.01
	Course of time (mins)	4/44	7.99**	7/77	2.82**	4/44	0.42

*p < .05. **p < .01.

As can be seen in Figures 2 and 3, R-R intervals were more regular during the difficult exam in comparison to the easy one. However, higher exam apprehension before the easy exam was associated with shorter R-R intervals in the first minute of exam situation ($r = -.62$, $p < .05$), while for the rest of the exam they were not significant.

For both exams, R-R intervals were significantly shorter and more regular during the exam period in comparison with pre-exam and post-exam periods (Figures 1-3). As shown in Figures 1-3, significant increase in R-R intervals and their variability parameters was shown after the first minute of the exam (Table 2). Since spectral analysis would not be appropriate to apply on a minute-to minute basis, because of a small number of data, first five-minute periods were analysed, whose $\log_{10}$ of SMI indices for the three frequency bands are shown in Figure 4.

Two-way ANOVA did not show significant differences in SMI indices between the difficult and the easy exam, for any of the frequency bands. The differences were significant between pre-exam, exam and post-exam periods, for all three frequency bands (Table 3). As shown in Figure 4, SMI indices were significantly smaller for both exams during the exam period, in comparison with pre- and post-exam periods. As it can be seen in Figure 4, a comparison of the SMI indices between the LF and MF bands showed significantly smaller SMI indices in the MF band for all the three periods of the difficult and the easy exam ($p < .01$).

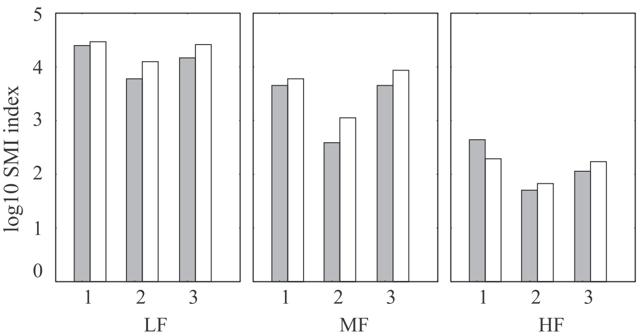

Figure 4. Changes in SMI indices for LF, MF and HF bands before (1), during (2) and after (3) difficult exam (gray columns) and easy exam (white columns).

Table 3
ANOVA of changes in spectral parameters during pre-exam, exam and post-exam period (periods)

Variable	Source of variation	*df*	*F*
SMI index for LF band	Exam difficulty	1/11	0.90
	Periods	2/22	6.09**
SMI index for MF band	Exam difficulty	1/11	0.76
	Periods	2/22	15.48**
SMI index for HF band	Exam difficulty	1/11	0.03
	Periods	2/22	16.40**

**p < .01.

DISCUSSION

Some earlier studies found an increase in state anxiety during pre-exam period, but with no reference given to the exam difficulty (Dimitriev, Dimitriev, Karpenko & Saperova, 2008; Marazziti et al., 2007). This finding was more prominent in female subjects (Bhansali & Trivedi, 2008; Kosmala-Anderson & Wallace, 2007). The results of this study showed that the perceived exam difficulty had effects on state anxiety, high activation, and exam apprehension during pre-exam period, indicating generally higher stress before the more difficult exam in comparison with the easier exam.

Cardiac R-R intervals are often used as indicators of stress level. In this study, continuous minute-to-minute recording of cardiac R-R intervals during the real examination situation was used. It was expected that measurement which started five minutes before the exam and finished five minutes after the exam, could give more insight into changes in stress level during the exams of different difficulty. Shorter and more regular R-R intervals were found during the exam situations in comparison to the pre- and post-exam periods, which indicated a higher stress level and mental load during the exam itself. The R-R parameters also indicated higher mental load and stress levels during the more difficult exam, which could have been expected due to the pre-perceived differences in the exam difficulty. Although the levels of anxiety, high activation, and exam apprehension were higher during the pre-exam period for the difficult exam, no differences were found in the cardiac parameters during this period between the easy and the difficult exam. The same parameters, however, showed the first minute of examination to be the most stressful followed by an initial recovery towards more or less stable level for the rest of the exam (Figures 1-3). It seems that stress in the first minute was induced, at least partly, by uncertainty of the exam questions. After the subject started answering the questions, the stress seemed to have decreased, which partly may be due to the situational, i.e. exam habituation.

Similar results were obtained by Zeller, Handschin, Gyr, Benedict, and Batteagay (2004) when they used heart rate as an indicator of exam stress. As in the present study, they found the peak of heart rate at the beginning of the exam, followed by its decline during the rest of the exam. Heart rate also proved to be a good indicator of students' stress in classroom and laboratory activities, such as oral reports, exams, and laboratory exercises (Elwess & Vogt, 2005). In another study, which included speech presentations, heart rate increased as the presentation progressed (Grossman, Wilhelm, Kawachi & Sparrow, 2001). Although different in nature, the results of the present study are in agreement with those reported by Spangler, Pekrun, Kramer, and Hofmann (2002), who found that negative emotions, such as anxiety and hopelessness (but not anger) peak at the beginning of the exam, while joy and hope increases during the exam and reaches the peak at its end.

Since the exam stress consists of two major components, i.e. mental and emotional, their relative contributions to overall stress was assessed by the use of spectral analysis of R-R intervals. Its parameters were compared for the two exam situations (difficult and easy exam), where lower SMI indices were found for all the frequency bands during exams, as compared to pre- and post-exam periods. Furthermore, lower SMI indices for the LF and MF bands during the exams could be interpreted as an increase in emotional and mental component of stress, respectively. The other R-R parameters singled out the exam situations as the most stressful in comparison with the other two periods. These interpretations seem acceptable, because exams are usually highly emotionally charged and mentally demanding situations.

Some authors (Sloan et al., 1996) have related changes in the LF band of R-R intervals to the effects of emotional component of stress. Other studies, however, have related changes in the MF band to changes in mental load (Aasman et al., 1987; Langewitz et al., 1990; L. J. M. Mulder, 1988; Sammer, 1998; Veltman, 2002). This seems plausible, since changes in the emotional state depend on relatively slow changes in the limbic and autonomic nervous systems, while mental activity depends on comparatively faster processes of the cortex.

In this study, significantly smaller SMI indices in the MF band were found than in the LF band during the exams suggesting thus that mental component of the stress was more prominent during the exam itself.

The changes in the HF band which, according to previous studies (L. J. M. Mulder, 1988), reflects breathing and speech motorics, also showed a decrease in SMI indices during the exams in comparison with pre-and post-exam periods. This difference was not necessarily due to the changes in respiration per se, but most probably due to the effects of speech on respiration patterns since the exams were oral. Using multiple regression with an index of spectral power in HF band as a criterion measure and respiration rate and tidal volume as predictors, almost 60% of the variance spectral power in this band were explained by respiratory parameters (Grossman & Taylor, 2007). On the other hand, some authors (Althaus et al., 1998; Sloan et al., 1996) point out that stress could be the cause of spectrum power suppression in HF band, which they explained by the reduced vagal activity on the cardiac atrial node. Furthermore, no significant differences in the SMI indices were found between the difficult and the easy exams, in the three frequency bands, which was not expected according to the obtained variability parameters in this study. This apparent discrepancy in the results suggests that, although the two exams implied different levels of stress, as seen via cardiac parameters, they had, nevertheless, similar frequency compositions. This means that the factors influencing cardiovascular dynamics were more or less the same in both exam situations.

CONCLUSIONS AND RECOMMENDATIONS FOR FURTHER STUDIES

In conclusion, it could be said that the levels of anxiety, high activation and exam apprehension indicated higher stress before the difficult exam. Standard parameters of R-R intervals, such as the mean and the variability indices, showed good discriminative characteristics in differentiation of stress levels between the exam periods of the difficult and the easy exam. As expected, a higher stress level was found during the difficult exam. The stress level, as indicated by the means and DM indices of R-R intervals, was the highest during the first minute of the exam period. After this, it seems that the exam habituation took place in both exams. Spectral parameters also showed differences in stress level between the exams, on one side, and pre-exam and post-exam situations, on the other, but there was no difference between the exam periods in the mental or emotional components of stress. Lower SMI indices for LF and MF bands during the exam situations may be interpreted as an increase in mental and emotional components of stress, respectively, compared with pre- and post-exam periods.

Further use of spectral analysis in similar studies is needed to find out more about its applicability in time series analyses of this kind of data. Future studies on larger samples of both genders are also required for quantification and comparison of mental and emotional components of stress via the changes in SMI indices in LF and MF bands. In the present study, subjects of the same gender were chosen to eliminate effects of the gender variable. One should be aware, though, that the obtained results and conclusions based on them may not be extended to male subjects.

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