UNIVERSITY OF PÉCS
FACULTY OF HEALTH SCIENCES
DOCTORAL SCHOOL OF HEALTH SCIENCES

Head of Doctoral School: Prof. István Kiss Ph.D.
Head of programme: Erzsébet Rétsági Ph.D.
Supervisor: Pongrác Ács Ph.D.
Co-supervisor: Miklós Tóth Ph.D.

The Effect of Extreme Physical and Psychological Stress on Autonomic Regulation in Terms of Heart Rate Variability (HRV) Parameters.

Doctoral (Ph.D.) dissertation

Ákos Móra

Pécs, 2024

REVIEW OF THE LITERATURE

In the early 1900s, after the concept of homeostasis was defined, the perspective emerged that the cells of the human body maintain complete constancy, a steady-state condition, in their internal environment. However, in the second half of the century, with the advancement of technology, it became possible to study signal processing, which revealed that continuous variability is a part of biological constancy. The optimal degree of continuous and dynamic variability in the regulation of the human body is necessary for the organism to flexibly adapt to changes in the environment and to maintain the required homeostasis despite adverse conditions. On the other hand, excessively low or high variability is unhealthy and, in addition to improper functioning of the heart and circulatory system, may indicate exhaustion, disease, psychological problems, or intellectual disability. The regulation of homeostasis and circulatory adaptation to environmental stimuli is the responsibility of the autonomic nervous system, which involves both sympathetic and parasympathetic influences.

Healthy heart function, which is capable of adequately responding to environmental stimuli, assumes a continuously varying heart rate. On a healthy resting ECG, the time intervals between consecutive R waves (R-R intervals) are not equal. This phenomenon is referred to as heart rate variability (HRV).

HRV PARAMETERS

Time-domain parameters

Time domain metrics are calculated using one of the most fundamental research methods, examining the changes in appropriate physiological indicators over time.

R-R: The time elapsed between two consecutive R waves, expressed in milliseconds. The sequence of R-R intervals can be measured simply and non-invasively, but due to arrhythmias and various measurement errors, this data cannot be used as a basis for further calculations.

N-N: Normal-to-normal heart cycle. The time elapsed between two consecutive ectopia-free R waves, expressed in milliseconds. This metric serves as the basis for further calculations of time-domain and frequency-domain parameters.

SDNN: The standard deviation of normal heart cycles, measured in milliseconds. The SDNN metric is highly sensitive to the duration of data recording, making it unsuitable for comparing measurements of different lengths.

rMSSD: The root mean square of successive differences in heart cycles. It is the primary indicator of changes in HRV caused by vagal tone. For rMSSD, a higher value indicates greater parasympathetic influence and is, therefore, closely correlated with the HF value.
pNN50: The proportion of consecutive heart cycle pairs in the entire sample where the time difference is at least 50 milliseconds. It shows a strong correlation with parasympathetic activity, and thus with rMSSD and HF metrics.

Frequency domain parameters

The neural background of heart rate variability is that the sympathetic and parasympathetic autonomic tones vary periodically at different frequencies. The goal of analyzing frequency domain metrics is to evaluate this variability.
HF: The HF metric refers to the frequency range between 0.15 and 0.4 Hz, representing parasympathetic dominance and autonomic tone.
LF: The LF metric refers to the frequency range between 0.04 and 0.15 Hz. Although it was previously believed that the LF metric indicated sympathetic tone, it is now proven that both sympathetic and parasympathetic tones influence the LF value.
LF/HF: This was once considered the best indicator of autonomic tone; however, due to the debated physiological significance of the LF metric, the interpretation of the ratio's value is currently unclear.

THE EFFECT OF STRESS ON HRV

According to János Selye, stress is the non-specific response of the body to a stimuli. In defining stress, we distinguish between the triggering stimulus (stressor) and the body's response to it (stress); namely, stress is the body's way of coping with any external or internal threat or sense of danger. In a stressful situation, the sympathetic branch of the autonomic nervous system becomes dominant, which, through the increased secretion of stress hormones (e.g., cortisol, adrenaline), strives to mobilize as much energy as possible in the body to facilitate effective defense.

In the international literature, stress is primarily measured by using validated questionnaires. However, in recent decades, beyond questionnaire surveys, it has become increasingly necessary to apply a physiological indicator or examination method capable of quantifying the extent to which the body has had to cross its variability limits, which represent homeostasis, in coping with external circumstances. The analysis of heart rate variability (HRV) has become the most widely used method, owing to HRV's high sensitivity to changes in the autonomic nervous system.

Physical Stress

All indicators reflecting parasympathetic impacts (rMSSD, pNN50, HF) decrease during exercise compared to pre-exercise baseline values, indicating an increase in sympathetic dominance and a decrease in parasympathetic effects, as well as an increase in heart rate. However, the magnitude of these modifications varies according to individual factors. With physical stress, parasympathetic effects continuously decline until they reach the first ventilatory threshold, however a small degree of vagal control can still be detected even at maximum intensity. The effects of different types of exercise on HRV have been examined and compared multiple times, revealing that exercise intensity above 80% of a person's maximum heart rate influences frequency domain measures, whereas exercise intensity below 80% has no effect.

Psychological Stress

Several researchers have studied the relationship between psychological stress and HRV. These studies commonly observed and detected low parasympathetic activity and elevated sympathetic dominance, resulting in reports of decreased HF values. In the case of psychological stress, experience plays a crucial role; experienced individuals develop psychological models and automatisms for processing information, which gives them a reserve cognitive capacity that can be used to solve unexpected situations. This mental capacity must also be reflected in vagally-regulated HRV indicators, as these show a strong correlation with cognitive performance and prefrontal neural activity.

HYPOTHESES

The goal of the longitudinal study was to demonstrate that HRV is suitable for detecting the effects of stress on the body and for displaying differences between various types of stress.

1. We hypothesized that HRV values would show significant changes even during a 5-day training camp.
2. We assumed that different types of exertion would generate different degrees of changes in HRV metrics among athletes of the same fitness level.

3. We hypothesized that the extent of HRV metric changes would be lower in participants undergoing recovery training compared to those who did not engage in recovery training.

In our comparative study, we attempted to isolate the physical and psychological effects that play a role in the body's regulation simultaneously in everyday life, allowing us to evaluate the effects of physical and psychological stress on the body separately.

4. We hypothesized that we would observe differences in the regulation of the body's response to physical versus psychological stress.
5. We assumed that due to extreme stress, the majority of HRV metrics would show significant changes compared to the resting state.

METHODS

In the dissertation, the evaluation of data from two independent studies is presented. The longitudinal study examined how different types of exertion affect athletes' HRV values during a short, 5-day training camp. The second study aimed to separate the physical and psychological stress experienced by the athletes in order to analyze the effects of each stress type on the body independently.

PARTICIPANTS

In the longitudinal study, 15 healthy, first-division male handball players (age 18.9 ± 0.8 years) participated. The athletes were randomly assigned to three groups: members of Group 1 participated in one intensive training session per day (1; n=5), members of Group 1+1 participated in one intensive and one recovery training session per day (1+1; n=5), and members of Group 2 participated in two intensive training sessions per day (2; n=5) over the course of 5 days. In the comparative study of two different stressors, 63 healthy, second-division male soccer players (age 25.14 ± 5.81 years) participated. The participants completed two independent test protocols a few days apart. The first test involved an extreme physical stress test, while the second test involved an extreme psychological stress trial.

RESEARCH PROTOCOL

In the longitudinal study, the athletes were required to participate in intense training sessions, and depending on their group assignment, in recovery sessions as well. The intense training sessions included a combination of running, strength training, functional exercises, and sport-specific training, all led by the club's strength and conditioning coach. The requirement for the intense sessions was that the athletes rated the session's intensity at an average of at least 15 on the Borg scale, which measures perceived exertion and subjective load intensity. The requirement for the recovery sessions was that their heart rates should not exceed 79% of their maximum heart rate, and that their heart rate should remain between 60% and 79% of their maximum heart rate for 90% of the total training time.

In the comparative study, the extreme physical stress test involved participants completing a voluntary, gradually intensifying running program on a treadmill until exhaustion, based on the modified Bruce protocol. The extreme psychological stress protocol was completed by the participants a few days later. After entering the examination room, their task was to walk across the room, but they were warned that unexpected situations might occur, and they would need to decide whether the situation posed a danger to them or not. After some time, a specialist entered the room through a side door and fired two blank shots in the direction of the participant, intentionally aiming beside them. Although the intervention avoided injury, it achieved the intended stress effect of the study.

HRV MEASUREMENT

In the longitudinal study, we used a Polar V800 heart rate monitor with a Polar H7 heart rate sensor. The athletes conducted the measurement each morning before the morning training session, in a supine position. The measurement lasted 9 minutes, during which we analyzed 265 ectopic-free heart cycles following stabilization of heart rate. In the comparative study, we had the opportunity to measure HRV metrics under laboratory conditions, so we used a 12-lead ECG immediately before the test (pre) and 30 minutes after the test (post). The measurements were taken in a supine position, and from the data, we selected 265 ectopic-free heart cycles for analysis.

RESULTS

LONGITUDINAL STUDY

The training load values of athletes in Group 2 were significantly higher during the first 3 days compared to the values of the other two groups (2 vs. 1+1 p=0.0005; 2 vs. 1 p=0.0001), which resulted from the fact that the sum of the TL (training load) indicators from the two intense training sessions was significantly higher than the TL values from a single session in the other two groups. On the fourth and fifth days, we found significant differences only between the members of Group 2 and Group 1 (p=0.0191). Due to the substantial decrease in the training load indicator, there was also a significant difference between the TL values of Group 2 in the first three days and those on the fourth and fifth days (p=0.0078). The substantial decrease in the training load indicator in Group 2 can be attributed to increased fatigue. Parallel to the changes in TL values, the amount of time the athletes spent training above 80% of their maximum heart rate also changed.

We did not find a correlation between the athletes' rating of perceived exertion (Borg scale) and the training load values of a given session (r=0.091), nor were we able to demonstrate significant correlations within the groups (r=0.238 in Group 2, r=-0.335 in Group 1+1, r=0.369 in Group 1). After separating the data from the first 3 days based on the training load results, we still did not find significant correlations (r=0.267 in Group 2, r=-0.189 in Group 1+1, r=0.300 in Group 1). We also did not find a relationship between the player's position and the Borg scale ratings (r=0.022), nor between the position and training load values (r=-0.370).

It is clearly visible that the SDNN and average R-R interval values of Group 2, which were elevated compared to the baseline measurement, decreased considerably until the fourth and fifth days, then started to rise again. The results are consistent with the changes in the training load indicator, as the high TL values achieved in the first three days were accompanied by a decrease in the athletes' time-domain measurements, while the decrease in TL due to fatigue allowed these measurements to rise. This observation is further supported by the changes in pNN50 data, which, after a significant increase measured on the morning of the second day, reached its lowest value on the fourth day, then reversed with the decrease in the TL indicator, showing an increase that was in line with the changes in other HRV parameters.

In addition to the time-domain indicators, the frequency-domain parameters also showed the previously described pattern. Both the high-frequency HF and the low-frequency LF indicators, after being elevated compared to the baseline measurement, gradually decreased until the fourth day, then increased in the second half of the week, contrary to the training load indicator. The dynamic changes in the LF/HF ratio did not follow the pattern of the aforementioned indicators, showing significant individual variability. No consistent trend could be observed within the groups. The study confirmed the relationship described in international literature between rMSSD and the HF component. The dynamics of these two parameters, which indicate the level of parasympathetic activity, are consistent across all three groups.

COMPARATIVE STUDY

After the extreme physical stress test, all HRV parameters increased significantly compared to the resting values. Following the extreme psychological stress test, most of the variables also showed significant changes, although some indicators decreased. During both test protocols, we observed significant changes in the NNmax, NNmean, pNN50, rMSSD, HF, and LF/HF parameters.

All HRV values, except for the LF indicator, changed in opposite directions during the physical and psychological stress tests. The changes in the NNmax (ms), NNmean (ms), SDNN (ms), pNN50 (%), rMSSD (ms), VLF (%), HF (%), and LF/HF ratio parameters showed highly significant ($p<0.001$) differences between the test protocols. After grouping based on the average relative VO_2peak values, we found significant differences in the change of the LF/HF ratio during the physical stress test, as well as in the change of the VLF parameter during the psychological stress test. Pearson's correlation matrix revealed strong correlations between the extent of changes in certain HRV indicators for both the physical and psychological stress protocols.

DISCUSSION

LONGITUDINAL STUDY

Based on the athletes' TL values, it can be seen that the training load value of the athletes in Group 2, significantly differed from that of the other two groups during the first 3 days, due to the fact that they spent approximately twice as much time on intense training compared to the other two groups. However, this significant difference disappeared during the training sessions on the 4th and 5th days, as the continuous accumulation of fatigue without proper recovery training, disabled the athletes maintaining the same level of intensity as in the trainings before. This is further supported by the fact that the time spent on training above 80% of the athletes' personal maximum heart rate significantly decreased after the third day. On average, during the first three days, they spent 60% of the total training time above the 80% of their maximum heart rate value, but this number dropped to 15% and 18% on the fourth and fifth days, respectively. The changes of various HRV parameters also support this argument. In case of the members of Group 2, the average R-R interval, pNN50, and HF indicators showed a continuous decrease until the morning of the 4th day, followed by a gradual increase during the morning measurements on the 5th and 6th days. A decrease in the average R-R intervals (an increase in heart rate) indicates sympathetic dominance, while an increase of this parameter indicates elevated parasympathetic tone. The change in the pNN50 parameter follows the same pattern, where its incessive decrease up until the morning of the 4th day signifies an increasing dominance of sympathetic tone, but as fatigue set in, the body enhanced vagus activity to return to homeostasis, leading to an increase in the pNN50 parameter.

The similar dynamic change can be seen in the HF component, which indicates parasympathetic effects, is self-explanatory and is further corroborated by the dynamic changes in the rMSSD parameter, which strongly correlates with the HF parameter. The literature review mentioned that the physiological basis of the LF component is still debated, and the previously popular view that it reflects sympathetic effects has been debunked. If it were indicative of sympathetic effects, its trend should have changed contrary to the above mentioned tendency, but the pattern recorded during the training camp matched that of the parameters mentioned above. The analysis of the LF/HF ratio did not reveal any clear, definitive trend regarding the development of autonomic tone, as the values measured for the participants and the individual variability of the data showed considerable variability and strong heterogeneity within the groups. This observation confirms that the LF indicator is not, or not exclusively, an indicator of sympathetic tone, thus proving that the LF/HF ratio is also not suitable for demonstrating autonomic regulatory processes.

For the 1+1 group, we were curious about the significance of recovery training during longitudinal loading. The TL values for the participants in this group did not show a difference between the first three and the last two days of the training camp, and the amount of time spent above 80% of their individual maximum heart rate only slightly decreased compared to the athletes in Group 2. When evaluating the HRV indicators, particularly the average RR interval and the LF and HF parameters, it can be seen that the results for this group remained close to the baseline values, with no significant changes observed.

COMPARATIVE STUDY

According to the findings of Clemente-Suárez and Robles-Pérez (2013), participants in a combat situation exhibited increased sympathetic tone, indicated by the decrease in both average R-R intervals and the SDNN metrics, signifying the activation of the “fight-or-flight” mechanism. Our study, however, did not confirm these results. We measured increasing SDNN and average R-R interval values in our participants. We believe the different results can be attributed to the choosen sampling methodology of participants, as the research of Clemente-Suárez and Robles-Pérez involved experienced soldiers, whereas we examined second-division football players who experienced increased stress levels even before the psychological test began, triggered by the mere handover of a weapon.

In the aforementioned study, the soldiers had to perform a complex mental task during the test (a protocol specifically developed for soldiers), whereas, in our study, participants were given a very simple task (they had to walk from one end of the room to the other), which was necessary to avoid the onset of long-term psychological symptoms. Based on the results of the psychological stress protocol, we could not confirm the findings of studies that focused on measuring pre-competition anxiety. These studies reported an increase in sympathetic dominance (decreasing SDNN and rMSSD). In contrast to the research analyzing the physiological effects of the military situation, where soldiers showed increased sympathetic tone, the changes in the metrics of our participants indicated the opposite of these findings.

The individuals in our study encountered this situation for the first time in their lives, while the participants in the aforementioned studies were experienced with the situation defined by the research protocol. Our data suggest that encountering an unknown danger for the first time has a more significant impact on HRV data changes than any other factor. The situation defined by our research protocol represented extreme psychological stress for our participants, which, in proportion to the magnitude of the stress, generated significant changes in the vast majority of HRV parameters. The level of psychological stress induced in our study greatly exceeded that experienced in previous studies, due to the nature of the protocol, the participants' lack of prior knowledge, and additional stress-enhancing factors (e.g., the provision of a gun to participants). This combination of factors explains the significant deviation from the results described in the literature.

ANSWER TO THE HYPOTHESES

1. **We hypothesized that HRV values would show significant changes even during a 5-day training camp**

   Our hypothesis was confirmed, as the intensive repeated load generated significant changes in the body's autonomic regulation, and the HRV parameters proved to be suitable for reflecting these changes. Based on our findings, it can be stated that HRV parameters are capable of illustrating autonomic changes and trends in response not only to acute stress but also to cumulative, long-term stress effects.

2. **We assumed that different types of exertion would generate different degrees of changes in HRV metrics among athletes of the same fitness level.**

Our hypothesis was confirmed, as the varying training loads generated different dynamic changes in the body's autonomic regulation, which were also reflected in the HRV parameters. The quantifiability of the autonomic responses to different loads can provide valuable information for researchers working in sports science and for sports professionals involved in athlete preparation.

3. **We hypothesized that the extent of HRV metric changes would be lower in participants undergoing recovery training compared to those who did not engage in recovery training.**

   Our hypothesis was confirmed, as the HRV parameters of athletes who engaged in recovery training showed less variation during the training camp than those of athletes who did not participate in recovery sessions.

4. **We hypothesized that we would observe differences in the regulation of the body's response to physical versus psychological stress.**

   Our hypothesis was not, or only partially confirmed. Although we observed regulatory differences between the two stress protocols, when comparing our results with international literature, we concluded that prior experience in the specific situation has a greater influence than the nature of the stressor. In the present study, the participating athletes had only encountered the extreme physical stress protocol previously, while the extreme psychological protocol introduced a novel, unfamiliar stressor that triggered adaptations in their autonomic regulation. However, according to the literature, researchers observed similar autonomic changes and trends in individuals who regularly encounter extreme psychological stress (e.g., soldiers) as we measured in athletes during the extreme physical stress protocol.

5. **We assumed that due to extreme stress, the majority of HRV metrics would show significant changes compared to the resting state.**

   Our hypothesis was confirmed, as extreme stressors generated significant changes in all HRV metrics, regardless of whether participants had prior experience with the given situation. The high-intensity stress caused substantial changes in autonomic regulation, although the direction of these changes depended on the presence or absence of prior experience in the given situation.

NEW FINDINGS

The longitudinal study confirmed that the analysis of HRV parameters is suitable for evaluating stress impacts on the body in the context of training sessions as well. The examination method yields reliable results even when athletes engage in training of different types or intensities, as we observed HRV changes with differing dynamics between groups, which are logical and professionally substantiated.

The original objective of the comparative study was to isolate the two major components of complex stressors affecting athletes (physical and psychological stress) and to examine their effects separately, independently of each other. Our hypothesis was that the two types of stress would generate different changes in autonomic regulation. However, based on the results, we concluded that experience has a much more significant influence on autonomic regulation than the nature of the stressor.

KEY WORDS

stress, physical stress, psychological stress, HRV, autonomic regulation

REFERENCES

Móra, Á.; Komka, Zs.; Végh, J.; Farkas, I.; Kocsisné Szilágyi, Gy.; Bosnyák, E.; Szmodis, M.; Ligetvári, R.; Csöndör, É.; Almási, G.; Oláh, A.; Kemper, H. C. G.; Tóth, M.; Ács, P.
Comparison of the Cardiovascular Effects of Extreme Psychological and Physical Stress Tests in Male Soccer Players
INTERNATIONAL JOURNAL OF ENVIRONMENTAL RESEARCH AND PUBLIC HEALTH 19: 2 Paper: 715, 12p. (2022)

Móra, Á.; Bosnyák, E.; Elek, L.; Betlehem, J.; Szmodis, M.; Ács, P.; Tóth, M.; Oláh, A.
Kézilabdázók szívfrekvencia változékonyságának alakulása eltérő edzésterhelés esetén ötnapos edzőtábor alatt - pilot study
MAGYAR SPORTTUDOMÁNYI SZEMLE 20: 1 (78) pp. S-41-S-45. (2019)

Móra, Á.; Bosnyák, E.; Elek, L.; Betlehem, J.; Szmodis, M.; Ács, P.; Tóth, M.; Oláh, A.
Eltérő edzésterhelésre adott vegetatív válasz ötnapos edzőtábor alatt - pilot study
MAGYAR SPORTTUDOMÁNYI SZEMLE 19: 75 pp. 67-67., 1 p. (2018)

Móra, Á.; Komka, Zs.; Szendrei, E.; Végh, J.; Farkas, I.; Szilágyi, Gy.; Bosnyák, E.; Ligetvári, R.; Csöndör, É.; Far, G.; Almási, G.; Szmodis, M.; Tóth, M.; Ács, P.; Oláh, A.
Extrém fizikai és mentális stresszre adott eltérő vegetatív válasz
MAGYAR SPORTTUDOMÁNYI SZEMLE 20: 2 (79) pp. 66-67., 2 p. (2019)

Móra, Á.; Komka, Zs.; Szendrei, E.; Végh, J.; Farkas, I.; Szilágyi, Gy.; Bosnyák, E.; Szmodis, M.; Ligetvári, R.; Csöndör, É.; Far, G.; Almási, G.; Ács, P.; Tóth, M.; Oláh, A.
Differences of autonomic nervous system regulation at physical and psychological stress
In: Heazlewood, IT Asia-Singapore Conference on Sport Science 2019 : Programme & Abstract Book
EAST ASIA RESEARCH (2019) 62 p. pp. 20-22., 3 p.

Móra, Á.; Bosnyák, E.; Elek, L.; Betlehem, J.; Szmodis, M.; Ács, P.; Tóth, M.; Oláh, A.
Kézilabdázók szívfrekvencia változékonyságának alakulása eltérő edzésterhelés esetén ötnapos edzőtábor alatt: pilot study
MAGYAR SPORTTUDOMÁNYI SZEMLE 19: 5 (77) p. 69 (2018)

Móra, Á.; Bosnyák, E.
A szívfrekvencia-változékonyság (HRV) adatainak értelmezése a sporttudományban
MAGYAR SPORTTUDOMÁNYI SZEMLE 17: 4 (68) pp. 72-73., 2 p. (2016)