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ORIGINAL ARTICLE

The effect of HRV biofeedback, yoga and mindfulness training on autonomic nervous system, perceived stress, and dispositional mindfulness

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Abstract

BACKGROUND: The aim of the present article is to examine the effectiveness of three intervention methods used to stress optimalisation – heart rate variability (HRV) biofeedback, yoga training, and mindfulness training – on autonomic stress indicators, such as resting HRV, peripheral body temperature and skin conductance level as well as, perceived stress, and trait mindfulness.

METHODS: Two research studies were carried out: In study 1 we compared the effects of the three interventions, while in study 2, we tested the effectiveness of specific HRV biofeedback training on resting HRV, perceived stress, and dispositional mindfulness. The research sample consisted exclusively of healthy participants who were randomly assigned to one of the experimental groups or a control group. The efficiency of interventions was assessed using the repeated measures of the aforementioned autonomic markers, both before and after the intervention programmes.

RESULTS: The results demonstrated the effectiveness of the intervention methods on a sample of healthy individuals. The HRV biofeedback in Study 1 led to an increase in HRV, peripheral body temperature, and dispositional mindfulness; yoga exercises also led to increased HRV and dispositional mindfulness, and the mindfulness training programme had an impact only on trait mindfulness. The HRV biofeedback training in Study 2 was effective in terms of psychological variables, as it stimulated the development of the trait of mindfulness, however the data did not support the effects on autonomic indicators.

CONCLUSION: The present findings show that intervention methods aimed at effective stress management may also prove to be effective in a sample of healthy individuals exhibiting increased vulnerability to stress conditions.

Introduction

A distinctive feature of present-day society is the long-term effects of stress stimuli, often leading to a chronic stress response which negatively impacts individuals' physical and mental health. From an evolutionary point of view, humans are able to respond well to acute stress stimuli; however, prolonged, accumulated, or unmanaged stress can have harmful consequences for the functioning of the human body (Mizoguchi *et al.* 2000; Jackson, 2014).

University students constitute one of the vulnerable groups of people who have experienced an increase in perceived stress levels and mental disorders in recent years (Bayram & Bilgel, 2008; Stallman & Shochet, 2009). Several studies have identified the incidence of serious adverse effects of perceived stress on their academic performance (Vaez & Laflamme, 2008), mental health (Andrews & Wilding, 2004), nutrition and sleep (Britz & Pappas, 2010), as well as the development or aggravation of psychosomatic disorder symptoms (Chiu, Chon & Kimball, 2003; Gulewitsch *et al.* 2013). The increasing prevalence of chronic stress and stress-related disorders in university students has led to the need to seek out and investigate effective methods of stress management and the reduction of its adverse effects.

Research in the field of stress reduction programmes is currently focused on several intervention techniques that have the potential to reduce or even prevent the negative effects of excessive stress. These most often include various forms of biofeedback, relaxation, autogenous training, physical activity, mindfulness training, meditation, and yoga (Jain *et al.* 2007; Miu *et al.* 2009; Van Der Zwan *et al.* 2015).

The present article focuses on three selected intervention methods: mindfulness, yoga, and HRV feedback. These interventions were primarily chosen for their undeniable advantages, such as their availability and applicability to people regardless of age, gender, educational background, intellectual profile, and interests. In addition, these interventions are inexpensive, have a high level of control over exercise performance, and exhibit minimal side effects. Despite the philosophical, cultural, and practical differences between these methods, the results of previous studies point out their significant potential for use in the field of psychosomatics and stress-related disorder treatment (Bränström *et al.* 2010; Brown & Gerbarg, 2005; Kosuri & Sridhar, 2009; Ratanasiripong *et al.* 2015; Reiner, 2008; Tan *et al.* 2011).

The main research objective was to examine and compare the effectiveness of the three aforementioned intervention methods in changing resting autonomic indicators related to stress. The intention was to expand existing knowledge on the effectiveness of individual interventions aimed at coping with stress to a healthy population, which can then serve as a starting point for prevention programmes aimed at reducing maladaptive stress management and its symptoms. In order to further investigate the effectiveness of HRV biofeedback, two studies were carried out. The first study concentrated on assessing the effectiveness of yoga, mindfulness, and HRV biofeedback. The second study focused on HRV biofeedback using an experimental training protocol drawn up by Lehrer, Vaschillo, and Vaschillo (2000).

Study 1
Methods

Research sample

The research sample comprise seventy one participants (51 females, 20 males) aged from 19 to 42 years (a mean age of 20.8). All participants were university students. Participants gave their written informed consent, and afterwards received credits for participating in the research. None of the participants reported any cardiovascular or neurological conditions, and they were not taking any medication that could affect their cardiovascular or autonomic function.

The research sample was randomly assigned to three experimental groups and one control group. The experimental groups were defined on the basis of the provided intervention: a mindfulness group (n=15), a HRV biofeedback group (n=20), and a yoga group (n=20). The control group (n=16) received no intervention.

Pre- and post-intervention measurements

All experimental groups, including the control group, underwent pre- and post-intervention repeated measurements of resting autonomic indicators in order to detect changes attributable to the selected intervention method. Autonomic measurements were performed individually in the presence of one experimenter in the laboratory. The post-intervention measurement took place three months after the pre-intervention measurement. A condition of both repeated measurements was that they would take place no later than one week after the beginning or end of the intervention programme.

The NeXus-4 psychophysiological system with BioTrace software was used for calculating heart rate (HR) and heart rate variability (HRV) from ECG signal, other measured variables included skin conductance level (SCL) and peripheral temperature measured at the non-dominant hand. Resting measurement in seated position lasted 6 minutes, after which participants filled the ten-item Perceived stress scale (Cohen *et al.* 1983) and Mindfulness Attention Awareness Scale (MAAS) (Brown & Ryan, 2003).

HRV analysis was performed in Kubios HRV software.

Interventions

All three interventions took place simultaneously, and each experimental group completed one.

a) HRV biofeedback

Under the guidance of two experimenters, the participants completed a total of eight, fifteen-minute training sessions, with each one taking place once a week.

During the exercise, participants were instructed to breathe in a slow and controlled manner (six breaths per minute, as per Lehrer, Vaschillo, and Vaschillo, 2000) and to learn breathing patterns that support a high HRV. HRV biofeedback was administered using the Nexus-4 hardware and Biotrace software. In addition to the ECG sensor, the participants were connected to a respiratory sensor located in the diaphragm area which provided them with real-time information in a graphic form on whether their respiration corresponded with the target respiratory rate. The form of feedback varied across the individual exercises (graphic and numerical data, feedback in audio and video form, and games).

In addition to biofeedback training that took place in the laboratory, the participants were asked to practise slow and controlled breathing in their home environment. The participants carried out these ten-minute exercises in their home environment without receiving any feedback. The exercises were performed at least five times a week using a web application showing an adjustable breathing animation (Gibson Research Corporation, available online at https://www.grc.com/breathe.htm). The participants were instructed to record the date, time, length, and course of the exercises in an online form after each one was completed.

b) Yoga

The participants assigned to this group completed an eight-week yoga course. The sixty-minute yoga exercises were held once a week in groups and were led by a yoga instructor. Each yoga class consisted of two parts: the first forty-five minutes were focused on practising asanas and their variations with an emphasis on dynamics, strength, and endurance; the final fifteen minutes were devoted to breathing exercises and relaxation.

The ”yoga” group also performed exercises at home, including practising sun salutations (Suryanamaskar). This set of exercises included a cyclical practice of a series of twelve positions in tune with one’s own breathing rhythm (Kim, 2014). The participants were instructed to perform exercises at home at least five times a week for fifteen to thirty minutes. The participants were instructed to record the date, time, length, and progress of the exercises in an online form.

c) Mindfulness training

Mindfulness, or attention training, had the same duration as the previous two intervention techniques (i.e., eight weeks) which is also consistent with the standard duration of the MBSR course. The training comprised group exercises which were performed once a week during which the participants learned mindfulness techniques and home exercises via audio recordings. This group was also instructed to record the date, time, length, and course of the exercises on an online form.

Statistical data analysis

As a part of the statistical analysis, the collected data were processed using the IBM SPSS 23.0 statistical software (IBM, Chicago, IL, USA). Logarithmic transformation of LF and HF HRV parameters was performed to adjust for positive skew, this is in accordance with the Task Force (1996) recommendations.

The effect of repeated measurements before and after the interventions and the interaction between repeated measurements and type of intervention were evaluated using an analysis of variance with repeated measurements (ANOVARM,) with 2 within subject repetitions and 4 groups. The significant values of the measurement x group interaction were interpreted as the effect of the intervention. The statistical significance level was defined as $p<0.05$.

Results

The interactions between the repeated measurements and the group representing the intervention effect shown in Table 1 reached statistical significance under the SDNN parameter ($F_{(3, 67)} = 5.574$, $p<0.01$), LF(log) ($F_{(3, 67)} = 5.639$, $p<0.01$) and the MAAS score ($F_{(3, 63)} = 11.098$, $p<0.01$). No significant interactions were observed for HR (BPM) ($F_{(3, 67)} = 2.355$), HF (log 10) ($F_{(3, 67)} = 1.359$), temperature ($F_{(3, 62)} = 1.173$), SCL ($F_{(3, 65)} = 0.746$) and PSS score ($F_{(3, 63)} = 0.247$).

Study 2
Methods

Research sample

A total of thirty-three participants (female n=25, male n=8) aged from twenty to thirty-eight years (a mean age of 22) participated in Study 2. All participants were university students. When the research was conducted, the same exclusion criteria as in study 1 were employed. They signed the informed consent and received credits as a reward for their research participation.

The research sample was randomly divided into an experimental group (n=17) and a control group (n=16). The experimental group underwent an intervention in the form of HRV biofeedback training. None of the participants had any previous experience with HRV biofeedback. The control group did not undergo any form of intervention but was subjected to pre- and post-intervention measurements, which were performed in both groups.

Procedure

Physiological indicators of HR, HRV were measured. The same hardware and software was used for measurement and data analysis as in study 1.

Pre- and post-intervention measurements

Both participants groups underwent repeated pre- and post-intervention measurements in order to record any psychophysiological changes induced by the HRV biofeedback training. Physiological measurements were performed in groups in the presence of one experimenter in a classroom. The final measurement took place two months after the initial measurement.

Tab. 1. Essential descriptive characteristics and intra- and inter-subject effects of repeated measurements before and after the interventions in the individual variables of the examined set

				Measurement effect			Measurement*group interaction		
		before M ± SD	after M ± SD	F	p	η^2	F	P	η^2
HR (BPM)	HRV biof.	83,6 ± 10,1	87,5 ± 14,7	4,04	0,05	0,06	2,36	0,08	0,10
	Yoga	83,1 ± 13,1	79,7 ± 16,3						
	Mindfulness	78,5 ± 10,8	86,8 ± 12,7						
	Control group	80,6 ± 8,5	84,8 ± 6,6						
	Total	81,7 ± 10,9	84,5 ± 13,5						
SDNN	HRV biof.	49,9 ± 13,8	60,1 ± 19,3	2,86	0,10	0,04	5,57	0,00	0,20
	Yoga	53,7 ± 21,5	52,8 ± 24,2						
	Mindfulness	53,1 ± 22,9	40,6 ± 15,2						
	Control group	53,7 ± 19,1	40,9 ± 15,6						
	Total	52,5 ± 19,8	49,6 ± 20,7						
rMSSD	HRV biof.	42,4 ± 21,5	42,8 ± 20,8	10,23	0,00	0,13	2,13	0,10	0,09
	Yoga	51,4 ± 28,4	47,2 ± 24,1						
	Mindfulness	49,1 ± 26,4	34,4 ± 21,8						
	Control group	46,0 ± 18,6	31,8 ± 11,7						
	Total	47,1 ± 23,9	39,8 ± 21,0						
HF (log 10)	HRV biof.	6,5 ± 1,1	6,3 ± 1,1	18,87	0,00	0,22	1,36	0,26	0,06
	Yoga	6,9 ± 1,2	6,5 ± 1,2						
	Mindfulness	6,8 ± 1,3	6,1 ± 1,2						
	Control group.	6,6 ± 1,1	5,9 ± 0,8						
	Total	6,7 ± 1,2	6,2 ± 1,1						
LF (log 10)	HRV biof.	6,9 ± 0,7	7,6 ± 0,9	0,11	0,74	0,00	5,64	0,00	0,20
	Yoga	6,8 ± 1,0	7,0 ± 1,1						
	Mindfulness	6,9 ± 1,0	6,7 ± 0,8						
	Control group	7,2 ± 1,0	6,6 ± 0,9						
	Total	6,9 ± 0,9	7,0 ± 1,0						

The participants were also asked in advance not to smoke cigarettes and drink caffeinated coffee for at least three hours before the measurements and not to drink alcohol for at least twelve hours prior to the measurements taking place (Laborde, Mosley, and Thayer, 2017).

In the introductory part of the repeated measurements, the participants were given instructions on how to properly connect the ECG sensor using disposable electrodes and how to turn it on and off. This was followed by a six-minute rest measurement in a seated position to record the baseline HRV values, followed immediately by another six-minute measurement in a standing position. This change in position during the physiological parameter measurement is usually referred to in the relevant literature as "orthostatic stress" and is a frequently-used and thoroughly-researched phenomenon characterised by changes in the ANS function. When changing the position from a seated to a standing position, there is an immediate decrease in the parasympathetic effect on the sinoatrial node as well as an increase in sympathetic activity (Paton, Boscan, Pickering, and Nalivaiko, 2005). As a result, the orthostatic test also has an effect on the HRV level and is mainly associated with a decrease in HRV values (Javorka, Turianikova, Tonhajzerova, Javorka, and Baumert, 2008; Zaidi and Collins, 2016). After the measurement, the participants were asked to fill out an online form of PSS and MAAS questionnaires within twenty-four hours.

HRV biofeedback

The experimental group completed a total of five twenty-minute HRV biofeedback exercises. The exercises took place on an individual basis once a week, always at the same time. The training was based on an

				Measurement effect			Measurement*group interaction		
		before M ± SD	after M ± SD	F	p	η^2	F	P	η^2
temperature	HRV biof.	28,3 ± 3,7	32,91 ± 1,70	76,38	0,00	0,55	1,17	0,33	0,05
	Yoga	26,5 ± 2,2	30,6 ± 2,5						
	Mindfulness	27,8 ± 3,3	32,5 ± 1,9						
	Control group	28,5 ± 3,1	30,8 ± 4,4						
	Total	27,7 ± 3,2	31,8 ± 2,7						
SCL	HRV biof.	4,1 ± 2,5	3,6 ± 2,7	0,08	0,78	0,00	0,75	0,53	0,03
	Yoga	4,1 ± 2,9	4,9 ± 6,9						
	Mindfulness	4,5 ± 3,7	4,8 ± 3,6						
	Control group	4,2 ± 3,1	2,9 ± 2,5						
	Total	4,2 ± 3,0	4,0 ± 4,4						
PSS score	HRV biof.	16,1 ± 8,5	15,2 ± 7,5	5,77	0,02	0,08	0,25	0,86	0,01
	Yoga	20,4 ± 7,2	19,2 ± 6,0						
	Mindfulness	17,4 ± 6,7	15,3 ± 5,8						
	Control group	18,2 ± 8,4	16,0 ± 9,6						
	Total	18,0 ± 7,7	16,5 ± 7,1						
MAAS score	HRV biof.	4,1 ± 0,9	4,1 ± 0,9	0,75	0,39	0,01	11,10	0,00	0,35
	Yoga	4,1 ± 0,8	4,0 ± 0,8						
	Mindfulness	3,6 ± 1,0	4,4 ± 0,8						
	Control group	3,7 ± 0,9	3,3 ± 0,7						
	Total	3,9 ± 0,9	4,0 ± 0,8						

M – mean, SD – standard deviation, F – Fisher's F-test value, *p* – statistical significance, n^2 = Eta coefficient, values close to zero signify minimal or no dependence; value 1 signifies complete dependence

experimental protocol focused on resonance frequency training designed by Lehrer, Vaschillo, and Vaschillo (2000). The term "resonance frequency" comes from the aforementioned authors and describes an individual respiratory rate at which a person is able to achieve the highest amplitude of respiratory sinus arrhythmia (RSA) and heart rate (HR) oscillations. This respiration rate is usually around six breaths per minute.

According to Lehrer, Vaschillo, and Vaschillo, the first step is identifying the client's resonance frequency. In this research, this was identified at the beginning of the first exercise using a biofeedback device. Each proband was connected to a respiratory sensor and an ECG sensor. The participants were instructed to breathe according to the breathing animation displayed on the computer screen, which determined the respiration rate and reduced it every two minutes (from seven breaths per minute to four breaths per minute). Upon completion, the experimenter evaluated the individual two-minute recordings and used them to determine the resonance frequency of each proband based on what respiration rate they had when they reached the highest RSA amplitude and the highest HRV values. The remainder of the first exercise was focused on learning to breathe at the resonance frequency with a prolonged exhalation.

Other exercises were aimed at mastering various breathing techniques: diaphragmatic breathing, breathing based on one's HR, prolonged exhalation, and exhalation with the mouth slightly open.

In addition to the training sessions led by the experimenter, the participants were asked to also practise breathing at an individually determined resonance frequency in real-life conditions outside the laboratory. The participants carried out these twenty-minute exercises in their home environment without receiving any physiological feedback. The exercises were performed once a day using a selected mobile application. The prerequisite of the selected application was its ability to set the respiration rate and the length of inspiration and exhalation, as well as the function to store historical data in order to check the frequency and length of exercises carried out by the participants outside the laboratory.

Tab. 2. Essential descriptive characteristics and intra-subject effects of repeated measurements before and after the interventions in the individual variables of the examined set and their interaction with the group

Seated position		before M ± SD	after M ± SD
HR (BPM)	HRV biof.	82,9 ± 12,0	83,3 ± 14,7
	Control	81,8 ± 15,0	80,2 ±11,2
	Total	82,3 ± 13,4	81,6 ±12,8
SDNN	HRV biof.	45,4 ±17,1	57,4 ±30,6
	Control	46,8 ± 22,3	47,0 ±19,0
	Total	46,1 ± 19,6	52,0 ± 25,4
HF (log 10)	HRV biof.	6,2 ± 1,1	6,4 ± 1,2
	Control	6,1 ± 1,3	6,3 ± 1,3
	Total	6,2 ± 1,2	6,3 ± 1,3
LF (log 10)	HRV biof.	6,8 ± 0,6	7,2 ± 1,3
	Control	6,9 ± 1,0	6,8 ± 0,8
	Total	6,9 ± 0,8	7,0 ± 1,1

Statistical data analysis

The IBM SPSS 23.0 software (IBM, Chicago, IL, USA) was used for statistical analysis, LF, HF and total power HRV were logartmicaly transformed, as in study 1.

The effect of repeated measurements before and after the interventions and the interaction between repeated measurements and groups were evaluated by an analysis of variance with repeated measurements (ANOVARM, 2 measurement x 2 group) using the Pillai's Trace (i.e., a robust and conservative test) and the Huynh-Feldt sphericity correction. The significant values of the measurement x group interaction were interpreted as the effect of the intervention.

Results

A) HRV parameters in a seated position

The essential descriptive indicators in the form of the mean and standard deviation, together with the test results on intra-subject effects and their interactions for the HRV parameters in a seated position, can be found in Table 2. None of the effects of repeated measurements and the measurement x group interactions do reach a level of statistical significance in any of the HRV parameters. The above results show that HRV values in the seated position did not differ between the experimental and control groups after completing the biofeedback training.

B) HRV parameters in a standing position

The test results on intra-subject effects and their interactions showed that neither of the HRV parameters reached statistically significant values after the repeated measurements or after the measurement x group interactions. In light of the above, it can be concluded that there were no significant differences between the repeated measurements of HRV parameters in the standing position before and after the biofeedback intervention, and no differences between the groups were observed.

C) Psychological variables

The repeated measurement x group interaction reached statistical significance under the MAAS score ($F_{1,31}$=10.494), These results suggest that the experimental procedure was an important factor for trait mindfulness. Based on means and standard deviations, it is clear that the experimental group had an increase in the MAAS score in the second repeated measurement, while the MAAS score decreased slightly in the control group. No significant interactions were observed for PSS score ($F_{1,31}$ = 3,737) (Table 3).

Discussion

The main objective of this research was to examine the effect of three selected intervention methods that are generally used to efficiently manage stress and eliminate its negative consequences on people's mental and physical health. The research was focused on mindfulness, yoga, and HRV biofeedback. The effectiveness of these three methods was tested in two research designs: Study 1 focusing on all three methods and Study 2 concentrating only on HRV biofeedback and its specific form of training as designed by Lehrer, Vaschillo, and Vaschillo (2000). The effectiveness of the methods was evaluated in terms of physiological indicators (HRV, SCL, and peripheral skin temperature) and psychological variables (perceived stress – PSS and dispositional mindfulness – MAAS).

In order to allow for a comprehensive understanding and evaluation of the results, one summarising discussion for both studies is presented.

Tab. 3. Descriptive characteristics and intra-subject effects of repeated measurements before and after the interventions in the individual variables of the examined set and their interaction with the group

				Measurement effect			Measurement*group interaction		
		before M ± SD	after M ± SD	F	p	η	F	P	η
PSS score	HRV biof.	18,9 ± 7,5	15,8 ± 5,9	4,06	0,05	0,12	3,74	0,06	0,11
	Control	17,9 ± 5,8	17,8 ± 6,0						
	Total	18,4 ± 6,6	16,8 ± 6,0						
MAAS score	HRV biof.	3,6 ± 0,6	3,9 ± 0,7	0,69	0,41	0,02	10,49	0,00	0,25
	Control	3,7 ± 0,7	3,6 ± 0,6						
	Total	3,7 ± 0,6	3,7 ± 0,7						
	Total	6,2 ± 1,2	6,3 ± 1,3						

As a basic parameter of the autonomous regulation of heart activity, the heart rate represents the interaction between vagal and sympathetic function. An increased heart rate is associated with arousal or an activation of the body (Azarbarzin, Ostrowski, Hanly, and Younes, 2014), which gives the HR measurement significant importance given the fact that the remaining HRV indicators mostly evaluate the parasympathetic function. The results of either study did not show effects of the interventions on HR, which means that the intervention techniques did not lead to systematic changes in HR values. This finding is inconsistent with previous research results, some of which report changes in the ANS function and the HR as a result of the HRV biofeedback, yoga, or the MBSR programme (Brown, and Gerbarg, 2005; Posadzki, and Parekh, 2009; Siepmann, Aykac, Unterdörfer, Petrowski, and Mueck-Weymann, 2008). One possible explanation is provided by Matthews, Jelinek, Vafaeiafraz, and McLachlan (2012), who found that in young and healthy people, stress can lead to changes at the HRV level without changes in the HR values. With regard to this result, they present a theory that there is likely a parasympathetic reserve in young adults that counters changes in ANS activity to ensure that the HR values are stable. For the present results, this means that if the HR values were to some extent resistant to stressors, they could also be more difficult to modify using intervention methods. However, this hypothesis requires further research into the autonomic regulation of HR in young and healthy individuals.

In addition to HR, the individual HRV parameters were measured: the overall HRV level was determined using the SDNN parameter and the total power in Study 2, the parasympathetic activity was assessed using the rMSSD time analysis and HF frequency analysis parameter, and sympathetic and parasympathetic activity were both measured using the LF band. The research presented in the theoretical part of this work, which highlights the positive effect of the selected intervention methods on the regulation of the cardiovascular system and the ANS function, led to the assumption that the selected techniques would have an effect on increasing both the parasympathetic activity and the HRV.

The results of the research showed the effect of the HRV biofeedback from Study 1 on physiological variables: within HRV, the SDNN and LF values increased, as did the peripheral body temperature. These changes can be interpreted as an increase in the overall HRV level and the parasympathetic tone (Reyes del Paso, Langewitz, Mulder, Van Roon, and Duschek, 2013; Billman, 2013; Ferreira and Zanesco, 2016). Study 2 showed that the HRV biofeedback had no effect on the examined somatic markers.

The finding that the biofeedback training in Study 1 had an effect on the HRV parameters, whereas the training in Study 2 did not show statistically significant effect on these variables, is interesting. One of the reasons for this inconsistency in the results may be the protocol itself, as it differed in the two studies. The HRV biofeedback training in Study 1 focused on breathing at a rate of six breaths per minute and on alternating between different forms of feedback (e.g., graphs, numerical data, audio, video, and games), whereas the training in Study 2 focused on individual resonance frequency breathing and was based on an experimental protocol drawn up by Lehrer, Vaschillo, and Vaschillo (2000). The efficiency of this protocol has been demonstrated in several studies (Lehrer *et al.* 2003; Karavidas *et al.* 2007; Lagos *et al.* 2008; Vaschillo, Vaschillo, and Lehrer, 2006) but the HRV evaluation in these cases was based on HRV values that were recorded during exercises, which is not in accordance with the HRV measurement standards (Task Force, 1996). The increase in RSA as well as the HRV level recorded in these studies, was most likely the result of the decrease in the breathing rate during the biofeedback exercise. The effect of the HRV biofeedback on HRV values should be measured outside the exercises, which is also important for the practical use of this method. In this research, the HRV values were measured before and after the spontaneous breathing training, which could have led to discrepancies in the results of Study 2 and other studies examining the effect of the protocol.

In addition to the differences in the procedure of the two biofeedback training sessions documented in Study 1 and Study 2, it is also important to mention the differences in the number of exercises and the duration of the training sessions, which are an important factor in the success of training programmes in general. In Study 1, the HRV biofeedback training consisted of eight exercises, whereas the participants participating in Study 2 completed a total of five exercises. The results of several studies (Dudášová, 2008; Swanson *et al.* 2009), including the present one, show that five exercises may not be sufficient to impact the HRV parameters. In connection to this, a clear problem is that there is currently no verified standardised protocol determining both the course of training and the minimum number of HRV biofeedback exercises which would lead to positive changes in the HRV parameters.

As another intervention method, yoga proved to be effective in terms of physiological parameters; the SDNN values increased, which saw an increase in the overall HRV level and a decrease in the rMSSD value that was, however, significantly lower when compared to the control group. Although yoga exercises (unlike biofeedback training) do not directly stimulate HRV, the research showed that they can have an effect on HRV, especially in terms of increasing its overall level. Similar results were found in a study conducted by Muralikrishnan, Balakrishnan, Balasubramanian, and Visnegarawla (2012), who interpreted the increase in SDNN values detected in a yoga-practising group as an improvement of the ANS's ability to more effectively regulate cardiovascular activity in response to ever-changing conditions.

Mindfulness has not been shown to be effective in terms of physiological variables, as there were no significant effects in any of the measured physiological variables. Previous studies report inconsistent findings in researching the effect of the mindfulness programme on the HRV level. Many studies that focused on this issue confirmed partial changes in HRV after completing the mindfulness programme, but there are still discrepancies in the affected HRV parameters. In a study carried out by Joo, Lee, Chung, and Shin (2010), the MBSR programme led to an increase in SDNN, rMSSD, and total power values, indicating an overall increase in HRV. Krygier *et al.* (2013) reported changes in the levels of the HF and LF bands in relation to mindfulness meditation, whereas Bhatnagar *et al.* (2013) recorded an increase in pNN50 after completing eight weeks of MBSR training. Studies that showed that the MBSR programme did not have any effect on HRV (Nyklíček, Mommersteeg, Van Beugen, Ramakers, and Van Boxtel, 2013; Owens *et al.* 2016; Zimmermann-Schlegel *et al.* 2019) were also taken into account. This inconsistency in results stems mainly from the shortcomings in methodology and research design, i.e., the low number of participants or design without a control group. These circumstances point to the increased need for high-quality research in the field of mindfulness and HRV, which would clarify the extent of the impact of mindfulness programmes on the autonomic and cardiovascular systems.

These findings suggest that the HRV biofeedback in Study 1 and yoga have the highest (albeit limited) efficiency in terms of somatic markers in healthy people. Due to the limited number (or limited frequency) of exercises included in the research design, future research ought to focus more on the effect of longer-term training with a higher frequency of exercises, which appears to be more effective according to previous studies (Hasset *et al.* 2007; Pusenjak, Grad, Tusak, Leskovsek, and Schwarzlin, 2015; Sutarto, Wahab, and Zin, 2010).

The results of the effect of intervention methods on psychological variables were also limited. The perceived stress level evaluated by PSS decreased in Study 1 in all groups (including the control group). Intergroup differences just above the significance threshold were observed in Study 2, which means that although the PSS score decreased in the HRV biofeedback group as opposed to the control group, such a decrease was not sufficient to be regarded as statistically significant. This result is contrary to expectations and to the results of the HRV parameters and peripheral body temperature, which showed an increase in the overall HRV level and the parasympathetic activity in groups participating in the HRV biofeedback and yoga exercises. Discrepancies between the subjective assessment of the stress level and the objective stress indicators, such as HRV, also emerged in previous studies (Riese, Van Doornen, Houtman, and De Geus, 2004); therefore, this is not an isolated phenomenon and the present result may be related to the participants' impaired ability to evaluate their own stress level. Moreover, the intervention methods in the two studies differed in the number and duration of exercises as well as in the course and activities performed during the exercises, which may have affected their impact on psychophysiological parameters relevant to the stress levels.

However, the intergroup differences were manifested in the dispositional mindfulness measured by the MAAS questionnaire; the MAAS score of all experimental groups increased after the interventions. This result suggests that the trait of mindfulness is developed through mindfulness programmes as well as through other intervention techniques, such as HRV biofeedback and yoga exercises. Although these techniques are different in terms of philosophy, content, and culture, they share certain common features, which is reflected by the finding regarding the trait mindfulness. All three methods are based on attention focusing, self-regulation, and conscious breathing control that lead to the reduction of anxiety and the stimulation of feelings of relaxation and unwinding (Van Diest, Verstappen, Aubert, Widjaja, Vansteenwegen, and Vlemincx, 2014), which likely contributed to the effect of the individual interventions on dispositional mindfulness.

Conclusion

The results of the two studies demonstrated the effectiveness of intervention methods in a healthy population; the HRV biofeedback in Study 1 led to an increase in HRV, peripheral body temperature, and dispositional mindfulness; yoga exercises also increased HRV and MAAS scores; the mindfulness programme had an impact only on increasing the level of mindfulness as a trait; and the biofeedback training in Study 2 was effective in terms of psychological variables, as it stimulated a decrease in the perceived stress level and the development of the trait of mindfulness. These studies are the first published research evaluating the effectiveness of several intervention techniques from an interdisciplinary point of view. As such, the results and findings may serve as motivation for further research into intervention techniques aimed at effective stress management.

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Conflict of interest
On behalf of all authors, the corresponding author states that there is no conflict of interest.

Ethical approval
All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Informed consent
Informed consent was obtained from all individual participants included in the study.

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