Praca oryginalna

Original paper

Response of equine autonomic system to application of vibrating plate treatments – preliminary studies

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Received 26.09.2025 Accepted 16.10.2025

Wilk I., Skowerska-Wiśniewska A., Wilczyńska L., Zeliszewska-Duk P., Matysek W. **Response of equine autonomic system to application of vibrating plate treatments – preliminary studies**

Summary

The hypothesis was that stress in horses getting accustomed to a vibrating plate could be reduced within seven days. Based on this hypothesis, the aim of the study was to analyze the heart rate variability parameters of horses during their acclimation to the vibrating plate. The study involved 10 warmblood horses aged 9-21 years. The study was conducted on a Technohorse vibrating plate (platform). The experiment was planned for seven consecutive days. During the first three days, the horses were accustomed to entering the space containing the vibrating plate and to being placed on the on the device itself. During the next four days, the horses were placed on the vibrating plate, and then a three-minute program was initiated. Heart rate variability (HRV) parameters were also measured during the experiment. Based on the obtained results, it can be concluded that the assumed seven-day period is sufficient to prepare the horses for relaxation treatments on the vibrating plate. The observed ranges of heart rate variability parameters illustrate clear differences between the first and subsequent measurements, indicating lower emotional arousal of the horses on subsequent days of their acclimation to vibrotherapy. Activating the platform itself did not affect the activity of the horses' autonomic nervous system when comparing the first and last activation of the device during the experiment. The heart rate (bpm) parameter, may be useful in assessing the emotional state, which may have significant application implications in terms of the use of vibrations in the regeneration of the horse's body.

Keywords: horse, HRV parameters, vibrating plate, welfare

Contemporary animal physiotherapy and methods for supporting horse welfare increasingly utilize relaxation techniques, which until now have been used primarily in human physiotherapy (3, 17, 24). One such solution is the use of vibrating plates, which affect the body through subtle vibrations that stimulate mechanoreceptors in tissues, thus affecting the muscular, circulatory, and nervous systems (21). The use of such devices in recreational and sporting horses aims to improve the body's regeneration, reduce stress, and improve the animal's overall comfort. One of the objective and increasingly widely used indicators allowing for the assessment of the physiological state and the body's response to external stimuli is heart rate variability (HRV) (13, 16, 19). Analysis of HRV parameters enables the assessment of autonomic nervous system activity, which allows for inferences about stress levels, tension, and adaptation to new conditions or treatments (25). For horses, especially physically active ones, monitoring heart rate variability

can provide important information about their welfare and physiological reactivity. The autonomic nervous system consists of two main parts: the sympathetic nervous system (SNS) and the parasympathetic nervous system (PNS). The SNS is responsible for the body's response to stress and mobilization for action ("fight or flight"). In the initial stages of vibrotherapy, especially in horses which are new to this type of treatment, brief activation of the sympathetic nervous system may occur – visible through, among other things, an increase in heart rate, muscle tension, or anxiety. This is the body's natural adaptive response to new sensory stimuli (8). During regular vibrotherapy, the activity of the parasympathetic nervous system may predominate, especially when the horse is gradually accustomed to the treatment and appropriate vibration parameters are used. This is a sign of the body transitioning into a state of relaxation, regeneration, and reduced stress reactivity. In practice, this is manifested by reduced respiratory and heart rate rates, improved digestion,

muscle relaxation, and increased heart rate variability (HRV), a marker of autonomic balance and physiological welfare (6). Vibrotherapy affects the balance between the sympathetic and parasympathetic nervous systems, modulating the organism's response to external stressors and supporting regeneration (12, 27). However, the appropriate dosage of stimuli, regularity of treatments, and an individualized approach to the animal are crucial. The assessment of the effectiveness of such therapies is supported by HRV analysis, which allows for an objective assessment of autonomic nervous system activity (5). It was hypothesized that stress in horses accustomed to the vibrating plate could be reduced within seven days. In connection with the hypothesis put forward, the aim of the study was to analyze the parameters of heart rate variability in horses during their accustoming to treatments on a vibrating plate.

Material and methods

The study was conducted according to the guidelines of the Declaration of Helsinki. Ethical review and approval were waived due to non-invasiveness of the study. The horses were subjected to non-invasive procedures in view of European directive 2010/63/EU and Polish laws related to ethics in animal experimentation. The horses belonged to the University of Life Sciences in Lublin, Poland, and were maintained in a riding center under the care of one of the authors who monitored their welfare and veterinary state as assigned by the university. The procedures took place in a familiar environment that they experienced daily and did not cause them any pain, suffering, or damage. The horses were clinically healthy, without any signs of sense disorders, no clinical signs of lameness or musculoskeletal injury; they demonstrated a comparable condition and athletic ability. All procedures were conducted in accordance with the Polish Animal Protection Act (21 August 1997).

The study subjects consisted of 10 warmblood horses, aged 9-21 years. The horses had no known health conditions and showed no known addictions or vices. The animals were used six days a week for two hours a day, for dressage or jumping. They also spent approximately four hours a day in the paddocks. Feed was provided three times a day. The standard ration consisted of meadow hay and oats with added vitamins. They also had unlimited access to water and rock salt. The horses were housed in 3.5×3.5 m boxes equipped with automatic waterers, bedded with straw, and separated by partitions. The stable was regularly ventilated.

The study was conducted on a Technohorse vibrating plate (platform). The vibrating platform generates controlled vibrations that are transmitted to the horse's body. The vibrating plate was equipped with a programmable electronic control panel and a stable, strong, and durable metal frame. The plate weighed 210 kg, was 1025 mm wide, 2310 mm long, and had a rail height of 1490 mm. The device was located in a dedicated space (4.9 m wide, 5 m long, 5 m height) within the stable where the test horses were housed, although they had never been in this space before. The space was situated along the stable corridor and



Fig. 1. A horse standing on a vibration platform while the experiment is being conducted

belonged to a series of technical spaces that were located opposite the boxes. The platform entrance was level with the floor so that the horses did not have to use a gangplank to access the device (Fig. 1).

The experiment was planned to run for seven consecutive days. It was assumed that during the first three days, the horses would be accustomed to entering the space containing the vibrating plate and to climbing onto the device itself. Horses could be allowed to participate in the subsequent elements of the experiment once they were able to freely enter the space and onto the device itself, and once placed on the plate, they were able to remain motionless for 30 seconds. It was assumed that the number of days could be increased depending on the individual needs of horses that had difficulty accepting the new task. During the next four days, the horses were introduced to the vibrating plate, and then a three-minute program was initiated. For the first two minutes, the vibration power was set to 10% of the device's power, and for the next minute, the power was set to 20%. After the program was completed, the horses were returned to their stalls.

During the experiment, heart rate variability (HRV) parameters were noted. These were performed three times on the first, fourth, and seventh days of the experiment: on the first day the horses were introduced to the vibrating plate, on the first day the device was launched, and on the last day of the experiment. HRV measurements were taken using POLAR Vintage 2.0 telemetry devices. Transmitters were placed on rubber bands at the level of the horses' hearts, and the receiver was attached to the same rubber band at the height of the withers. The animals were accustomed to wearing the devices. During the experiment, the horses were introduced to the space by two caregivers known to

them, who were responsible for their daily care, feeding and taking them to the paddocks. During the experiment, there were other horses present in the stable that they knew and had been staying there for at least a year.

Eight heart rate variability parameters were used for the analysis:

RR – the interval between successive R waves in the ECG, i.e., the time between heartbeats;

SDNN – Standard Deviation of Normal-to-Normal RR intervals – the standard deviation of RR intervals, measuring heart rate variability over time;

HR – Heart Rate – the number of heart beats per minute; RMSSD – Root Mean Square of Successive Differences – the root mean square of the differences in successive RR intervals, reflecting short-term heart rate variability;

VLF – Very Low Frequency – very low frequencies, related to hormonal and respiratory activity;

HF – High Frequency – high frequencies, related to the activity of the parasympathetic nervous system;

LF – Low Frequency – low frequencies, related to the balance between the parasympathetic and sympathetic nervous systems;

LF/HF – the ratio of low and high frequencies, indicating the balance between the parasympathetic and sympathetic nervous systems. Heart rate variability (HRV) parameter recordings were imported to a computer using PolarSyncFlow and then analyzed in Kubios HRV 4.2.0 (Kuopio, Finland). During the analysis of the recordings, HRV parameter values were extracted for the overall measurements of the first, second, and third repetitions (the ,,subsequent measurement" factor), and for the values of the subsequent activation of the vibrating plate (the "first and last activation of the vibrating plate" factor). Additionally, the heart rate variability parameter values were analyzed during the second and third measurements, separating the phase of placing the horses on the vibrating plate from the moment the platform was activated and the duration of the entire three-minute program (the ,,horse's time on the plate" factor).

PQStat software was used to analyze the data. First, the level of normality of the data was examined. None of the tests used demonstrated normal distribution, so further analysis was performed using Kruskal-Wallis ANOVA for independent groups for the following factors: next measurement, first and last activation of the vibrating plate, and the stage of the horse's stay on the plate. Significance of differences between means was determined using the POST-HOC Dunn Bonferroni test.

Results and discussion

Analyzing the averages obtained during subsequent repetitions, differences can be observed in the RR, HR, RMSSD, and HF parameters (Tab. 1). Differences in RR and HR were visible between the first, second, and third measurements. RR was characterized by a lower value compared to the two subsequent measurements, while HR had the highest values in the first measurement compared to the other two. The RMSSD parameter differed in each measurement, reaching its highest value during measurement number two. Another differences in the RMSSD parameter differences in each measurement, reaching its highest value during measurement number two. Another differences in the RMSSD parameter differences in the RMSSD parameter

Tab. 1. Mean values of the heart rate variability parameters in horses during subsequent test repetitions

Variables	Group	Median	Lower Quartile	Upper Quartile
	1	792.06ª	752.33	849.01
Mean RR (ms)	2	909.28b	803.83	943.12
	3	914.80₺	901.53	1033.20
	1	157.41ª	110.22	331.12
SDNN (ms)	2	219.13ª	122.56	647.39
	3	122.56ª	107.86	282.78
	1	75.75ª	70.68	79.76
Mean HR (bpm)	2	65.99⁵	63.62	74.64
	3	65.59⁵	58.07	66.55
	1	208.60ª	134.56	367.51
RMSSD (ms)	2	297.21 ^b	103.10	974.68
	3	139.28⁵	118.49	419.40
	1	2663.37ª	1623.06	7989.78
VLF (ms)	2	2663.70ª	1647.80	7485.30
	3	1647.80ª	853.94	2323.40
	1	7989.00ª	3407.50	14133.50
LF (ms)	2	6969.70ª	4005.80	36226.00
	3	6969.70ª	4056.60	25995.00
	1	12896.00ª	5111.15	31811.25
HF (ms)	2	4023.70b	1191.40	12235.00
	3	4023.70b	2957.90	9615.50
	1	1.61ª	0.68	3.12
LF/HF (%)	2	1.89ª	1.70	3.23
	3	1.73ª	1.73	2.99

Explanation: Means marked with the same letters are not significantly different at $p \le 0.05$

ence concerned the HF parameter, which had the highest values compared to the other two measurements.

During the first and last start-up of the vibrating plate, no significant differences were observed between the tested parameters (Tab. 2).

The averages obtained for the heart rate variability parameters for the horses tested in the subsequent stages indicate only differences in RMSSD (Tab. 3). Higher values were obtained during the stage when the horses entered the vibrating plate.

Analyzing the average HRV parameter values during the last session on the vibrating plate, differences will be visible for the RR and HR parameters (Tab. 4). In the case of RR, lower values were recorded when the horses were placed on the vibrating plate, while the opposite was true for the HR parameter, which showed lower values during the activation of the vibrating plate.

Currently, various forms of physiotherapy constitute a very important part of equine welfare (1). There are many different types of treatments designed to regenerate horses during intense training periods or during recovery from injuries. Scientific studies have

Tab. 2. Mean values of the heart rate variability parameters in horses during the first and last activation of the vibrating plate

Variables	Group	Median	Lower Quartile	Upper Quartile
Mean RR (ms)	First activation of the vibrating plate	853. 47ª	735.65	942.41
	Last activation of the vibrating plate	880.24ª	864.64	1050.10
SDNN (ms)	First activation of the vibrating plate	90.90ª	81.35	99.99
	Last activation of the vibrating plate	103.98ª	76.47	122.74
Mean HR (bpm)	First activation of the vibrating plate	70.31ª	63.82	82.87
	Last activation of the vibrating plate	68.16ª	57.14	69.39
RMSSD (ms)	First activation of the vibrating plate	94.00ª	57.03	124.40
	Last activation of the vibrating plate	99.20ª	63.88	177.72
VLF (ms)	First activation of the vibrating plate	1066.36ª	747.07	3101.40
	Last activation of the vibrating plate	1128.10ª	1047.70	1332.70
LF (ms)	First activation of the vibrating plate	3175.65ª	2033.05	9921.95
	Last activation of the vibrating plate	2710.50ª	2114.20	6756.80
HF (ms)	First activation of the vibrating plate	2848.65ª	1876.31	4053.05
	Last activation of the vibrating plate	4254.30ª	954.48	6405.20
LF/HF (%)	First activation of the vibrating plate	1.93ª	0.74	4.66
	Last activation of the vibrating plate	1.58ª	0.80	3.60

Explanation: Means marked with the same letters are not significantly different at p ≤ 0.05

Tab. 3. Mean values of the parameters of heart rate variability of horses for the factor of the stage of staying on the vibrating plate during the first activation

Variables	Group	Median	Lower Quartile	Upper Quartile
Mean RR (ms)	Placing on the vibrating plate	792. 06 ^a	752.33	849.01
	Activation of the vibration plate	853.47 ^a	735.65	942.41
SDNN (ms)	Placing on the vibrating plate	157.41 ^a	110.22	331.12
	Activation of the vibration plate	90.90 ^a	81.35	99.99
Mean HR (bpm)	Placing on the vibrating plate	75.75 ^a	70.68	79.76
	Activation of the vibration plate	70.31 ^a	63.82	82.87
RMSSD (ms)	Placing on the vibrating plate	208.60 ^a	134.56	367.51
	Activation of the vibration plate	94.00b	57.03	124.40
VLF (ms)	Placing on the vibrating plate	663.37 ^a	623.06	17989.78
	Activation of the vibration plate	1066.36ª	747.07	3101.40
LF (ms)	Placing on the vibrating plate	7989.00 ^a	3407.50	14133.50
	Activation of the vibration plate	3175.65 ^a	2033.05	9921.95
HF (ms)	Placing on the vibrating plate	12896.00ª	5111.15	31811.25
	Activation of the vibration plate	2848.65ª	1876.31	4053.05
LF/HF (%)	Placing on the vibrating plate	1.61ª	0.68	3.12
	Activation the of vibration plate	1.93ª	0.74	4.66

Explanation: Means marked with the same letters are not significantly different at $p \leq 0.05\,$

found numerous references to the positive effects of physiotherapy on the equine body. These studies primarily concern massage, lasers, cryotherapy, and magnetotherapy (23). One of the main treatments is the use of massage in various forms, such as sports massage, fascial massage, or relaxation massage (17). This method does not require any specialized tools other than appropriate manual techniques. Any type of massage applied to horses, depending on their needs,

can bring tangible benefits to the horse's body. These benefits are visible in physiological indicators such as cortisol concentration in saliva, as well as in behavioral responses that indicate stress reduction (10, 14). The literature also contains results obtained by other authors regarding physiotherapeutic treatments using appropriate equipment and infrastructure, such as magnetotherapy, laser therapy, shockwave therapy, red light therapy, and treadmills. Of all these, the most studies concern the use of magnetotherapy. The main benefits noted by researchers include accelerated tissue regeneration, reduced pain and inflammation, and improved circulation and cell oxygenation (7). This method is most often described in sport horses or horses recovering from injuries, but studies have also been conducted on geriatric horses (9). Another method that has attracted considerable interest among researchers is the use of red light therapy. It can be conducted using a solarium or special blankets placed on the horse's body. The use of red light had a positive effect on the horse's body, both physiologically and behaviorally (2, 18).

The use of vibration is one of the possibilities for equine regeneration, but it is not a widely used method, perhaps due to the need for a special platform. In the case of horses, there are few studies related to the effects of vibrating plate use on equine body function,

but it can be assumed that it can bring significant benefits considering the results of research in human medicine (4, 20). The cited studies primarily indicate improvement in balance, movement, and reduction of muscle spasticity in people affected by multiple sclerosis or cerebral palsy. Our study examined the effect of heart rate variability parameter ranges in horses accustomed to vibrating plate treatments. The noted differences were particularly noticeable when

Tab. 4. Mean values of the parameters of heart rate variability of horses for the factor of the stage of staying on the vibrating plate during the last activation

Variables	Group	Median	Lower Quartile	Upper Quartile
Mean RR (ms)	Placing on the vibrating plate	792. 06 ^a	752.33	849.01
	Activation of the vibration plate	880.24b	864.64	1050.10
SDNN (ms)	Placing on the vibrating plate	157.41 ^a	110.22	331.12
	Activation of the vibration plate	103.98 ^a	76.47	122.74
Mean HR (bpm)	Placing on the vibrating plate	75.75 ^a	70.68	79.76
	Activation of the vibration plate	68.16 ^b	57.14	69.39
RMSSD (ms)	Placing on the vibrating plate	208.60ª	134.56	367.51
	Activation of the vibration plate	199.204 ^a	63.88	277.72
VLF (ms)	Placing on the vibrating plate	663.37 ^a	623.06	17989.78
	Activation of the vibration plate	1128.10 ^a	1047.70	1332.70
LF (ms)	Placing on the vibrating plate	7989.00 ^a	3407.50	14133.50
	Activation of the vibration plate	2710.50 ^a	2114.20	6756.80
HF (ms)	Placing on the vibrating plate	12896.00 ^a	5111.15	31811.25
	Activation of the vibration plate	4254.30ª	954.48	6405.20
LF/HF (%)	Placing on the vibrating plate	1.61 ^a	0.68	3.12
	Activation of the vibration plate	1.58ª	0.80	3.60

Explanation: Means marked with the same letters are not significantly different at p ≤ 0.05

analyzing the factor of the subsequent measurement. The RR and HR parameter values clearly indicated lower emotional arousal of the horses during the second and third measurements compared to the first, which may indicate that the horses quickly accepted the new treatment, especially considering that the horses had to acclimate not only to the new form of physiotherapy but also to the new space in which the entire platform was located. Interesting values can be observed for the RMSSD parameter. Its values varied in each measurement, with the highest values recorded during the second measurement, which may indicate the highest activity of the parasympathetic nervous system, which to some extent resulted in the horses' greatest relaxation (22). In the case of the HF parameter, the highest values were visible in the first measurement compared to the two subsequent measurements. This is an extremely interesting reaction of the autonomic system, which may constitute a specific reaction of the body consisting in trying to compensate for stress in response to a new situation.

Comparison of HRV parameters during the first and last activation of the vibrating plate revealed no significant differences between the analyzed parameters. Interestingly, the range of parameter values did not differ significantly from those comparable to walking in a familiar environment (26). On the other hand, the lack of differences may indicate that the treatment itself was too short to demonstrate its relaxing effect.

An interesting relationship were observed during the first and last activation of the vibrating plate. During the first activation of the platform, differences in the RMSSD parameter were observed between the stage of

placing the horse on the plate and its activation. This was significantly lower when the platform was activated, indicating low activity of the parasympathetic nervous system and perhaps a reaction to the new experience of using vibration. Similar correlations were noted by Lee et al. (15) when analyzing HRV parameters while waiting for a new event. In the case of the last activation, the differences concerned the HR and RR parameters. In both cases, the observed values indicated lower emotional arousal of the horses during the activation of the vibrating plate. It is important that the horses showed differences in the HR parameter, because it is the most basic parameter (19), the determination of which

does not require specialized devices. This situation may have an additional positive impact in terms of the application of the results.

Based on the obtained results, it can be concluded that the assumed seven-day period is sufficient to prepare horses for relaxation treatments on the vibrating plate. The observed ranges of heart rate variability parameters illustrate clear differences between the first and subsequent measurements, indicating lower emotional arousal in the horses during the subsequent days of their acclimation to vibrotherapy. Activating the platform itself did not affect the activity of the horses' autonomic nervous system, comparing the first and last activation of the device during the experiment. The HR parameter, or heart rate, can be useful in assessing emotional state, which may have significant application implications for the use of vibration in equine regeneration. Further research should be continued to determine the effects of vibration on the horse's body.

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