Effects of osteopathic manual therapy on the autonomic and immune systems and the hypothalamus-pituitary-adrenal axis in the horse

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Abstract

Manual therapies, physiotherapy, and modalities are often used as a treatment and prophylactic method for preventing various injuries, as well as a rehabilitation tool in the post-operative, traumatic period for horses. Significant results have been achieved with these therapies: increased joint range of motion, tissue extensibility, and reduced pain and inflammation. This study evaluated the effect of osteopathic manual therapy (OMT) on the nervous and immune systems, as well as on the hypothalamus-pituitary-adrenal (HPA) axis. To assess changes in these systems, non-invasive biomarkers were chosen: the heart rate (HR), respiratory rate (RR), temperature (T), as well as objective measurements: blood serum cortisol (BSC) concentration and white blood cell (WBC) count. A total of 30 thoroughbred horses were randomly divided into an experimental group (n = 15) and a control group (n = 15). Blood samples and physiological indicator measurements such as HR, RR, T, BSC and WBC were taken before (p0), after (p1) and one hour after (p2) the OMT treatment for the experimental group. Analogical sequence was performed for the control group without OMT. A significant increase of BSC by 0.94 µg/dl after OMT suggest a direct influence on the HPA axis. Also, a substantial decrease of WBC by 0.57×10^9/l indicates the immune system's response to the treatment. Moreover, an observed significant decrease in HR and RR shows that the autonomic nervous system was affected by activating the parasympathetic nervous system. Based on the results obtained, it is concluded that OMT has an effect on the HPA axis and on the nervous and immune systems.

Equine osteopathy, ANS, PSNS, HPA, immunity

A few decades ago, the use of equids in daily human life was different from nowadays. Now we can see the term 'athlete' more often used when work with horses is being described. An equine athlete's career commonly ends due to traumas or pathologies in the musculoskeletal system. Various manual techniques are often used as a first-tier application for treating multiple conditions in the musculoskeletal system. Chiropractic, osteopathic, and massage therapies are types of manual work techniques that have different effects on biomechanical and physiological responses in living bodies. The goal of manual therapy is to enhance the unique ability of the body to heal itself by activating various regenerative and healing processes in the body through the neuromusculoskeletal system (Haussler 2009). The osteopathic manual therapy (OMT) is becoming increasingly common in horse rehabilitation. The physical manipulations involve joints, soft tissues, bones, and muscles (Lima et al. 2020).

Although the results can be seen immediately after the therapy in the horses' dynamic and behavioural changes, it is essential to know what physiological factors have an effect on the living body and its systems.

Materials and Methods

This research was conducted in accordance with the Law of the Republic of Lithuania, Order no. 8-500 on the Protection, Maintenance, and Use of Animals, of 6 November 1997 (the Official Gazette "Valstybės žinios" number 108-6595, dated 28 November 1997); Order no. 4-361 of 31 December 1998 of the State Veterinary

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Service of the Republic of Lithuania on the Breeding, Care, and Transportation of Laboratory Animals; and Order no. 4 of 18 January 1999 of the State Veterinary Service of the Republic of Lithuania on the Use of Laboratory Animals for Scientific Tests. The study approval number was B6-(19)-1476.

A total of 30 Thoroughbred horses were included in the study. The horses were from the same stables based in Lithuania; none were concurrently participating in the racing sports season or intensive training. The median age for both groups was six years, and the median weight was 452 kg. The horses were randomly separated into two groups comprising 15 horses each: the experimental group (EG, n = 15) and the control group (CG, n = 15). Both groups were formed analogically, of seven mares and eight stallions. None of the horses (n = 30) had ever had OMT treatment sessions. During the study, all participants were treated by the same protocol. The horses were marked by numbers to hide their identities. No horses were excluded from the study.

The evaluated measurements included the heart rate (HR), respiratory rate (RR), temperature (T), blood serum cortisol (BSC) concentration and white blood cell (WBC) count. Measurements were taken three times in both groups: before the OMT session (p0), immediately after (p1), and one hour after the treatment (p2).

Data regarding HR and RR were collected by auscultation using the stethoscope 3M Littmann Classic III. The head of the stethoscope was placed from the left side of the horse's body in intercostal spaces 3–4 for HR data collection. The HR beats were counted for two minutes. For data on RR, the head of the stethoscope was placed in the first third of the horse's trachea, and the abdominal wall movements were observed. Sounds of respiration while watching the abdominal wall were counted for two minutes.

Blood samples for WBC and BSC determination were taken by puncturing the jugular vein. All samples were collected in less than three minutes to avoid rising cortisol levels. The samples were tested in the Lithuanian University of Health Sciences Veterinary Academy laboratories. Blood analysis was made using two machines: AIA-360 (Tosoh Bioscience, Inc., San Francisco, JAV) and MS4s VET (MS Labors, Austria).

The BSC concentration was assessed to evaluate the OMT's effect on the HPA axis (Henderson et al. 2010) and the WBC count was determined to assess implications for the immune system (Walkowski et al. 2014). The physiological factor measurements were kept as autonomic nervous system biomarkers (Henderson et al. 2010).

High velocity and low amplitude thrusts were used to manipulate joints according to the Pascal's osteopathic technique (Pascal 2004). The OMT treatments were performed by an Equine Diplomat Osteopath (EDO) in the horse stall box to reduce stress. Each treatment took approximately 45 min. None of the horses were sedated during the treatment.

Statistical data analysis was made using the 24.0 version of the Statistical Package for Social Science programme. Spearman correlation was used for evaluation of data. Significance was considered when $P < 0.05$.

Results

All results are shown in Table 1.

When evaluating changes in the WBC values in EG, a significant difference was found between p0-p1 and p1-p2. There were no significant differences when evaluating the WBC values in CG. Evaluating the temperature data of EG, a significant difference was found comparing the p0-p1 to p1-p2 values; no significant change was found in CG. No significant changes were observed when evaluating the HR values in EG. In CG, a significant difference was found between p1 and p2 values. No significant changes were observed in either group when evaluating the RR values. A significant difference was found when evaluating the BSC concentrations in EG between p0-p1 and p1-p2. No significant changes were found in BSC concentrations in CG.

Spearman correlation in EG (Table 2) showed a significant correlation between WBC and T immediately after therapy ($r = -0.694$; $P = 0.004$), as well as between the HR and RR values ($r = 0.678$; $P = 0.005$).

Discussion

Evaluating the results of measurements and statistical analysis showed that the mean BSC concentrations in EG were 4.83 μg/dl, 5.77 μg/dl, and 5.21 μg/dl at p0, p1, and p2, respectively. Analysing the results, the differences in BSC concentrations in EG between p0 and p1, and also between p1 and p2 were significant. According to published research regarding the effect of OMT, an increase in BSC after osteopathic treatment can be directly

Table 1. Leukocyte and blood serum cortisol values determined before (p0), right after (p1), and one hour after (p2) the treatment.

Measurements	Group	Measuring time	Mean ± SD	Min	Max	*P*
WBC × 1000/mm^3	CG	p0	8.43 ± 1.39	6.30	11.29	-
		p1	8.40 ± 1.99	6.48	13.83	-
		p2	8.45 ± 1.70	6.88	12.97	-
	EG	p0	8.32 ± 1.42	5.76	10.27	0.027
		p1	7.76 ± 1.37	4.98	10.20	0.023
		p2	8.20 ± 1.25	5.64	10.30	-
BSC (µg/dl)	CG	p0	4.48 ± 1.38	1.16	5.84	-
		p1	4.84 ± 1.34	3.04	8.29	-
		p2	4.43 ± 1.61	2.69	8.00	-
	EG	p0	4.84 ± 1.79	2.20	8.04	0.031
		p1	5.77 ± 1.63	3.87	8.75	0.020
		p2	5.21 ± 2.00	3.02	9.02	-
HR/min	CG	p0	42.00 ± 7.75	24.00	56.00	-
		p1	38.67 ± 7.04	28.00	56.00	-
		p2	36.80 ± 8.03	28.00	56.00	0.017
	EG	p0	39.33 ± 6.40	24.00	48.00	-
		p1	35.07 ± 6.13	24.00	44.00	-
		p2	35.33 ± 6.17	28.00	48.00	-
RR/min	CG	p0	13.33 ± 3.83	8.00	20.00	-
		p1	11.47 ± 3.87	8.00	20.00	-
		p2	11.87 ± 3.27	8.00	16.00	-
	EG	p0	14.13 ± 3.25	10.00	20.00	-
		p1	12.00 ± 3.02	8.00	20.00	-
		p2	13.73 ± 4.59	8.00	20.00	-
Temperature (°C)	CG	p0	37.52 ± 0.28	37.00	38.00	-
		p1	37.55 ± 0.34	37.00	38.30	-
		p2	37.61 ± 0.27	37.20	38.20	-
	EG	p0	37.51 ± 0.39	37.00	38.20	0.040
		p1	37.68 ± 0.49	37.20	38.70	0.018
		p2	37.58 ± 0.43	37.00	38.30	-

WBC - white blood cell count; BSC - blood serum cortisol concentration; HR - heart rate; RR - respiratory rate; CG - control group; EG - experimental group
$P < 0.05$ comparing values in the same group

Table 2. Correlation between the monitored indicators.

Correlation		BSC	WBC	Temperature	HR	RR
BSC	r	---	0.158	-0.286	-0.239	-0.312
	P		0.575	0.302	0.390	0.258
WBC	r		---	-0.694**	0.220	-0.006
	P			0.004	0.431	0.984
Temperature	r			---	-0.015	-0.157
	P				0.958	0.577
HR	r				---	0.678**
	P					0.005
RR	r					---
	P					

WBC - white blood cell count; BSC - blood serum cortisol concentration; HR - heart rate; RR - respiratory rate; r - Spearman correlation coefficient
**Significant correlation at $P < 0.01$

related to the HPA axis activation (Sampath et al. 2019). Moreover, BSC was found to be significantly increased after equine chiropractic treatment (Lima et al. 2020).

The mean WBC values in EG were 8.32 × 10^9/l, 7.76 × 10^9/l, and 8.2 ×10^9/l at p0, p1, and p2, respectively. The WBC values in CG remained relatively stable, ~8.40 × 10^9/l at all measuring points. Change in the numbers of leukocytes in blood is controversial; an increase in the WBC count could be seen in healthy individuals after OMT (Noll and Johnson 2005). However, a few years later it was discovered that the change in WBC was much more statistically significant than was thought before (Noll et al. 2008). A significant decrease in WBC was also observed 5 min and 30 min after OMT (Walkowski et al. 2014).

Analysing the HR and RR values showed a significant difference in HR of CG when comparing the p1 and p2 measurements. Other HR and RR values showed no significant differences. Applying Spearman correlation analysis, medium-strength correlation between HR and RR was discovered (P = 0.005), which agrees with the hypothesis that OMT affects the nervous system. A significant decrease in HR was found after OMT in healthy individuals. These results have been discussed as the effect of OMT on the autonomic nervous system, which activates and modulates the parasympathetic nervous system (Giles et al. 2013). Moreover, according to Henderson et al. (2010), OMT activates the parasympathetic nervous system by decreasing HR and RR in healthy individuals, and the HPA axis by increasing blood serum cortisol levels.

Analysing temperature values in EG shows significant differences between p0 and p1. According to other research (Osilla et al. 2022), this change suggests that the autonomic nervous system was affected, and the applied therapy impacted the homeostasis of the horses' bodies.

In conclusion, our study shows that OMT affects the immune and nervous systems as well as the HPA axis in the body. Although not much research has been done on equids in the fields of rehabilitation and manual techniques, some relevant body of scientific work is analysed. In future studies, more measurement points could be made and additional markers could be added to make the results of our study more specific. Overall, more research is needed to determine the benefits of manual work in animals.

References

Giles PD, Hensel KL, Pacchia CF, Smith ML 2013: Suboccipital decompression enhances heart rate variability indices of cardiac control in healthy subjects. J Altern Complement Med **19**: 92-96

Haussler K 2009: Review of manual therapy techniques in equine practice. J Equine Vet Sci **29**: 849-869

Henderson AT, Fisher JF, Blair J, Shea C, Li TS, Bridges KG 2010: Effects of rib raising on the autonomic nervous system: a pilot study using noninvasive biomarkers. J Am Osteopath Assoc **110**: 324-330

Lima CR, Martins DF, Reed WR 2020: Physiological responses induced by manual therapy in animal models: A scoping review. Front Neurosci **8**: 430

Noll DR, Johnson JC 2005: Revisiting Castlio and Ferris-Swift's experiments testing the effects of splenic pump in normal individuals. Int J Osteopath Med **8**: 124-130

Noll DR, Johnson JC, Brooks JE 2008: Revisiting Castlio and Ferris-Swift's experiments on direct splenic stimulation in patients with acute infectious disease. J Am Osteopath Assoc **108**: 215

Osilla EV, Marsidi JL, Sharma S 2022: Physiology, Temperature Regulation. Treasure Island (FL): StatPearls Publishing. Available at: https://www.ncbi.nlm.nih.gov/books/NBK507838/

Pascal E 2004: Lehrbuch der strukturellen Osteopathie beim Pferd. Georg Thieme Verlag KG, Stuttgart, Germany, 329 p.

Sampath KK, Katare R, Tumilty S 2019: Stress axis and osteopathy: A dual hormone approach. Int J Osteopath Med **33-34**: 24-30

Walkowski S, Singh M, Puertas J, Pate M, Goodrum K, Benencia F 2014: Osteopathic manipulative therapy induces early plasma cytokine release and mobilization of a population of blood dendritic cells. PLoS One **9**: e9013