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Original Research

Repeated Measurements of Markers of Autonomic Tone Over a Training Season in Eventing Horses

Olivia Lorello [a,*], Alessandra Ramseyer [a], Dominik Burger [a], Vinzenz Gerber [a], Rupert M. Bruckmaier [b], Johannes H. van der Kolk [a], Cristobal Navas de Solis [a,1]

[a] Department of Veterinary Clinical Science, Swiss Institute of Equine Medicine, Vetsuisse Faculty, University of Bern and Agroscope, Bern, Switzerland
[b] Veterinary Physiology, Vetsuisse Faculty, University of Bern, Bern, Switzerland

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ABSTRACT

Autonomic nervous system tone varies with fitness, training, and conditions such as cardiac disease, hypertension, or overtraining. Normal values of autonomic tone markers and changes over a competition season are incompletely described in eventing horses. The objectives of this study were to describe normal values and changes over a competition season of markers of autonomic tone in competing eventing horses. These values were measured in conjunction with previously reported variables to monitor training and compared with non-competing breed-matched controls. Heart rate variability (HRV), non-invasive blood pressure, splenic volume, pre- and post-exercise hematocrit and cortisol, standardized exercise tests (SETs), and muscle enzyme activities were measured preseason (T1), midseason (T2), and at the peak and/or end of the competition season (T3) in eventing and control horses. Heart rate variability was lower ($P < .05$) at all times and post-exercise cortisol lower at T2 and T3 in eventing horses compared with controls. Heart rate variability and post-exercise cortisol did not change over the season in any group. Eventers had higher fitness levels during SETs than controls. Non-invasive blood pressure, splenic volume, hematocrit, pre-exercise cortisol, muscle enzyme activities, and weight were not significantly different between groups and did not change over the season. The lower HRV in competing eventers suggests a lower parasympathetic and/or higher sympathetic tone in this group. A lower post-exercise cortisol suggests a decreased stress response in eventers to the SET. Non-invasive blood pressure, splenic volume, and resting or post-exercise hematocrit did not detect differences or changes in autonomic tone in this population.

1. Introduction

The autonomic nervous system (ANS) balances sympathetic and parasympathetic tone. Changes in autonomic tone have been studied during exercise, training, and as a mechanism for arrhythmias in horses [1–3]. When athlete training loads are too high or resting periods are too short, maladaptations such overtraining may occur. Overtraining syndrome (OTS) results in long-term decrement in performance capacity with or without physiological and psychological symptoms [4].

Changes in ANS tone and dysfunction of the hypothalamic-pituitary-adrenocortical axis are proposed underlying mechanisms of OTS in both man [5–7] and horse [8–11], suggested markers of which include behavior, sleep disorders, blood pressure, and cortisol in humans [5,12,13] and hematocrit, post-exercise cortisol, and behavior changes in horses [10,14–16]. Other variables used

* Corresponding author at: Olivia Lorello, Department of Veterinary Clinical Science, Swiss Institute of Equine Medicine, Vetsuisse Faculty, University of Bern and Agroscope, Langasstrasse 124, Bern, 3012, Switzerland.
E-mail address: olivialorello@gmail.com (O. Lorello).
[1] Present address: Texas A&M University, Large Animal Clinical Sciences, College Station, TX, 77845.

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to monitor training and stress are body weight, run times, post-exercise lactate, heart rate (HR) during exercise [7], and plasma muscle enzyme concentration [14].

The horse has an intrinsically high parasympathetic tone at rest, whereas sympathetic tone dominates during exercise [17]. Sympathetic tone mobilizes the splenic reservoir, sending a large volume of concentrated red blood cells into circulation. The splenic reservoir is more pronounced in horses than in most other athletic species, and the splenic volume is correlated with the hematocrit after the contraction of its capsule [18]. Heart rate variability (HRV) estimates the activity of the sympathetic and parasympathetic systems, is used to monitor human athletes and has the potential to evaluate stress or emotional status in animals [19,20]. Blood pressure regulation is multifactorial, but high sympathetic tone increases non-invasive blood pressure (NIBP) [21]. Heart rate variability, splenic volume, NIBP, and post-exercise hematocrit could be useful, non-invasive markers of ANS activity in horses.

The objective of this study was to describe normal values and potential changes over a competition season of markers of autonomic tone in eventing horses and to compare these with a matched population of non-competitive horses. We hypothesized that at peak season, competing horse ANS balance would shift toward higher sympathetic tone and lower parasympathetic tone. This would cause decreased HRV, decreased splenic volume, decreased post-exercise hematocrit and cortisol, and increased NIBP in otherwise clinically healthy horses. Variables previously suggested to change with training including body weight, HR during and after exercise, blood lactate during exercise, creatinine kinase (CK), and aspartate aminotransferase (AST) were also obtained.

2. Materials and Methods

2.1. Subjects

A prospective longitudinal cohort study, performed with the approval of the appropriate committees for animal use and experimentation (VD2809 February 2014–2015), and the informed consent of all owners was received. Inclusion criteria in the eventing group at each time point was normal physical and lameness examinations, normal complete blood count (IDEXX ProCyte Dx Hematology Analyzer, IDEXX Diavet), and the goal to compete in one or more International Federation for Equestrian Sports sanctioned eventing competition CCI*-CCI**** (Concours Complet International) during 2014. Control horses were breed-matched horses with normal physical and lameness examinations, normal complete blood counts, considered able to complete the standardized exercise test (SET) described below and ridden less than 5 hours per week. All horses were enrolled by convenience sampling.

Three examinations were scheduled during the 2014 competition season based on each horse's competition schedule. The first time point (T1) was at least 4 weeks before the first competition of the season. The second time point (T2) was approximately halfway through the season. The third time point (T3) was within 4 weeks of the final and/or most demanding competition. Control groups were examined within 3 weeks of competition horses. Pre-exercise weight on a digital scale, CK and AST (IDEXX Catalyst Dx Chemistry Analyzer, IDEXX Diavet AG), pre- and post-exercise hematocrit, and pre- and post-exercise plasma cortisol were measured at each examination. Total cortisol was measured by radioimmunoassay after extraction with methanol, as previously described [22].

2.2. SETs

Standardized exercise tests, adapted from previously reported protocols in eventers [23], were performed on a fiber-sand 1,490 m racetrack and consisted of 10 minutes walking, 10 minutes trotting, 1,490 m of cantering at 450 m/minute, 1,490 m of galloping at 500 m/minute, 1,490 m of galloping at 550 m/minute, and 10 minutes of walking. Gallops were separated by 5 minutes of trotting. Temperature and humidity were recorded at the start of each test [39]. Blood samples were obtained by direct jugular venipuncture before and after exercise (30 mL) and after each gallop (0.5 mL). Hematocrit was measured by microcentrifugation before and after exercise. Lactate was measured immediately on whole blood before the test and after each gallop (Lactate Pro, Axonlab). An exponential line of best fit (Microsoft Excel 2010) was used to determine the speed at a lactate of 4 mmol/L (VL4) [23]. The cardiac rate and rhythm was recorded throughout the exercise test with a digital telemetry unit (Televet, Engel Engineering Services GmbH). Electrodes (Phillips M2202A, Philips Medical Systems) were placed in a modified base apex lead following manufacturer's recommendations. Electrodes were secured with adhesive foam (Animal Polster, Snogg AS) and glue (Medical Adhesive B, Ulrich Swiss). A GPS system, synchronized with the telemetry unit, was carried by the rider (Televet Mobile Kit, Engel Engineering Services GmbH) and the speeds were also manually timed. Heart rates during the last 30 seconds of each gallop were averaged and plotted against speed; a linear line of best fit equation (Microsoft Excel 2010) was used to calculate the speed at a HR of 200 beats per min (V200). The HR 10 minutes after the SET was recorded (HR10min). Results for exercising electrocardiograms (ECGs), 24-hour continuous ECG rhythm and echocardiograms are described elsewhere (unpublished data).

2.3. HRV

After the SET horses were equipped with a digital ECG monitor (Televet, Engel Engineering Services GmbH). Electrodes (Phillips M2202A, Philips Medical Systems) stabilized with cardboard were secured according to the user manual for horses without a saddle (Televet, Engel Engineering Services GmbH). The ECG was stored on a Secure Digital card. Horses trailered for 0 minutes to 3 hours to home. The period between 0 and 4 AM was used for HRV analysis. The N-N intervals were corrected manually using ECG analysis software (Televet, Engel Engineering Services GmbH) and exported into HRV software (Kubios HRV software version 2.2, Biosignal Analysis and Medical Imaging Group). Artefacts, premature complexes (RR interval 20% shorter than the previous R-R interval), and