Seasonal Variations of Haematological and Biochemical Parameters Associated with Potential Laminitis in Apparently Healthy Horses

Ememe M. U. 1*, Ukwueze C. S., 1 Ani N. V. 2 Aloka C. H. 1

1Department of Veterinary Medicine, College of Veterinary Medicine, Michael Okpara University of Agriculture, Umudike, Abia State
2Department of Theriogenology, College of Veterinary Medicine, Michael Okpara University of Agriculture, Umudike, Abia State

Accepted May, 2022 and Published June, 2022

ABSTRACT
Early detection of laminitis in horses is valuable as it provides information that can be used as a basis for advice to trainers of race and performance horses. The study was aimed to determine the seasonal variations in haematological and biochemical parameters of horses developing laminitis at Polo Club Port Harcourt, River State, Nigeria. A total of 18 horses were sampled during the dry season (February) and rainy season (July). The horses sampled were aged between 3 and 8 years. The hoof temperature, weight, packed cell volume (PCV), red blood cell (RBC) count, hemoglobin (Hb) concentration, total and differential leucocyte counts, glucose level and fibrinogen concentration were determined using standard methods. The mean hoof temperature of the horses was 36.1± 0.11°C and 35.6 ± 0.2°C during the dry and rainy season respectively. The mean body weight of the horses was 323 ± 8.90 kg in dry season while that of rainy season was 330 ± 11.0 kg. Glucose concentration during the dry season (71.6 ± 1.5 dl/mg) was significantly lower (P ˂ 0.05) than that obtained during the rainy season (91.7 ± 1.9 dl/mg). Furthermore, the PCV, RBC count and Hb concentration were significantly higher (P ˂ 0.05) in the rainy season than the corresponding parameters in the dry season. Similarly, the total leucocyte, neutrophil and lymphocyte counts were significantly higher (P ˂ 0.05) during the rainy season than the dry season. It was concluded that variations on the parameters determined may aid to identify early warning signs of laminitis.

Key Words: Laminitis, Hoof temperature, Heamatology, Biochemistry, Season

*Corresponding author:
email: maryeneme@yahoo.com
Tel: +234 (0)803 494 7650
INTRODUCTION

Laminitis is a disease that affects the feet of hoofed animals (ungulates) and it is found mostly in all breeds of horses [1], [2]. It is one of the most common causes of lameness and disability of horses and ponies [3]. Equine laminitis is a crippling disease in which there is a failure of attachment of the epidermal laminae connected to the hoof wall from the dermal laminae attached to the distal phalanx [4]. The laminae are responsible for suspending the distal phalanx within the hoof wall, a failure in the laminar along with the downward forces of the weight of the horse and disrupting forces such as the tension from the deep digital flexor tendon usually result in a devastating displacement of the distal phalanx and severe lameness [5], [6], [7].

Severe cases of laminitis with apparently visible clinical signs are known by the colloquial term founder [2]. Progression of the disease may lead to perforation of the coffin bone through the sole of the hoof, requiring aggressive treatment or euthanasia [8], [9]. Equine affected with acute laminitis show signs of pain, foot tenderness progressing to inability to walk, increased digital pulses and temperature in the hooves [10], [11], [12].

The risk factors associated with laminitis may include high sugar and starch in feed, lack of exercise, too much food for the amount of work being performed, stress, colic, dexamethazone administration, retained placenta, unusual distribution of fat behind the shoulder and over the loin [13],[14].

The economic and welfare of a horse already showing signs of laminitis is reduced due to poor prognosis, severe pain and the possibility of recurrence which have serious effect on the future performance of the horse [9], [15]. An abnormal increase in values of erythrocyte count, total leucocyte and differential leucocyte counts, packed cell volume, hemoglobin concentration, glucose level, insulin level, fibrinogen level and triglyceride level most commonly occur in performance horses in association with septic and non-septic inflammatory conditions [16], [17]. Such increase can be used for early diagnosis of laminitis [18], [19].

Early recognition before pathologic changes occur within the hoof could be invaluable to improving equine welfare [11]. These examinations may also be beneficial as part of a preventive medical programme for such group of horses, when observed on a regular and routine basis. It may also provide valuable information that can be used as a basis for advice to trainers of race and performance horses [9], [20].

This study seeks to establish the possible use of seasonal changes in hematological and biochemical changes as an early indicator for laminitis in horses.

MATERIALS AND METHODS

Study Area

The study was conducted at the Port Harcourt Polo Club, Port Harcourt, Rivers state, South-south, Nigeria. The city of Port Harcourt is located at a coordinate of 4°45' N, 7°00'E and elevation of 468M (1535 Ft) above sea level. The climate falls within the sub-equatorial climatic belt. It has a mean annual temperature of 31.97°C in dry season and 29.10°C in rainy season, relative humidity of 80-100% and mean annual rainfall of 2138 mm [21]. Port Harcourt Polo Club was established in 1972 and it has a large Polo field and practice field, paddocks, stables, lush green lawns and serene club house.
Experimental Horses
A total of 18 apparently healthy horses comprising Argentinian, Sudanese and indigenous (Nigerian) breeds, aged between 3 and 8 years were sampled. The horses are housed in standard stable measuring 8 Ft × 12 Ft made of concrete floor, cement block wall and asbestos roof and well ventilated. The horses were fed wheat bran, sorghum, hay and fresh pasture.

Sampling Methods and Sample Collection
The horses were sampled twice during the dry season (February) and rainy season (July) between 7:00 am and 10:00 am local time to get adequate participation of grooms in restraining the horses before they were taken out to graze and exercise. Horses were individually restrained with halter and held by an assistant before each procedure was performed. They were identified by their names. Body weights were taken with the aid of a weighing band. Ages of the horses were estimated by examining the incisors teeth for eruption, wear and Galvayne's groove.

Hoof temperature was taken using infrared surface thermometer. The kit was powered on and placed on the dorsal surface of the hoof of individual horse and the result read on the screen in less than 3 seconds.

Samples for haematological examinations were taken from horses with minimal excitement. Ten milliliters (10ml) of blood was collected from each horse through a jugular venipuncture with 20ml disposable sterile syringe and 18G needle and 4ml was dispensed into an Ethylene Di-amine Tetra- Acetate-k (EDTA-K) plastic sample bottle. All samples were serially labeled. Blood samples were transported to the laboratory in cold box containing ice-pack to determine the haematological parameters, glucose level and concentration of fibrinogen.

Laboratory Analysis
Blood samples were analyzed at Department of Veterinary Medicine Laboratory, College of Veterinary Medicine, Michael Okpara University of Agriculture, Umudike. Abia State.

The anti-coagulated blood was used to determine packed cell volume (PCV), haemoglobin (Hb) concentration, red blood cell (RBC) counts, total leucocyte and differential leucocyte counts. The PCV was determined by microhaematocrit method, Hb concentration by cyanmethemoglobin method, RBC counts and total leucocyte counts by haemocytometer method. Differential leucocyte count was determined with stained blood smear using Leishman's technique and counted by battlement counting method [22].

Blood glucose level was done using Accu check® glucometer (Roche, United Kingdom). The kit is operated by battery or optional AC adapter. A test strip was inserted in the glucometer and 10µl of blood was dropped on the strip immediately the blood sign appeared on the screen. The result (glucose level) was read on the screen in less than 15 seconds.

Fibrinogen concentration was determined with the EDTA-anticoagulated blood by a heat precipitation method [23]. Plasma fibrinogen concentration was considered high when it was greater than the reference interval (2–4 g/L).

Data Analysis
Graph Pad prism 4.0 for Windows (Graph Pad Software San Diego California USA) was used for analysis. Data obtained were expressed as means ± standard error of mean (Mean ± SEM). The results were subjected to Student's t-test to determine the difference in parameters between dry and rainy season samples. Values of p <
0.05 were considered statistically significant.

RESULTS

The mean hoof temperature of the horses was 36.1± 0.11°C during the dry season and 35.6 ± 0.2 °C during the rainy season. The mean body weight of the horses was 323 ± 8.90 kg in dry season while that of rainy season was 330 ± 11.0 kg (Table 1). Blood glucose level was 71.6 ± 1.5 dl / mg during the dry season and 91.7 ± 1.9 dl / mg during rainy season. Furthermore, the PCV, red blood cell count and haemoglobin values were significantly higher (P < 0.05) in rainy season than the corresponding values in dry season (Table 1). Similarly, the total white blood cell, neutrophil, and lymphocyte counts were significantly higher (P < 0.05) during rainy season than dry season. There was no significant difference between the eosinophil count and fibrinogen level during the dry and rainy seasons.

Table 1

The dry and rainy season parameters of the horses from Polo Club, Port Harcourt, Nigeria.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Dry season</th>
<th>Rainy season</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hoof Temperature (°C)</td>
<td>36.1± 0.11</td>
<td>35.6 ± 0.2</td>
</tr>
<tr>
<td>Weight (Kg)</td>
<td>323 ± 8.90</td>
<td>330 ± 11.0</td>
</tr>
<tr>
<td>Fibrinogen (g/L)</td>
<td>2.0 ± 0.0</td>
<td>2.0 ± 0.0</td>
</tr>
<tr>
<td>Glucose (mg/dl)</td>
<td>71.6 ± 1.5&lt;sup&gt;a&lt;/sup&gt;</td>
<td>91.7 ± 1.9&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>RBC (x 10&lt;sup&gt;6&lt;/sup&gt;)</td>
<td>4.6 ± 0.36&lt;sup&gt;a&lt;/sup&gt;</td>
<td>7.2 ± 0.15&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Hb (g/dl)</td>
<td>9.2 ± 0.57&lt;sup&gt;a&lt;/sup&gt;</td>
<td>11 ± 0.30&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>PCV (%)</td>
<td>27.0 ± 1.7&lt;sup&gt;a&lt;/sup&gt;</td>
<td>32.5 ± 1.0&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>WBC (x10&lt;sup&gt;9&lt;/sup&gt;/L)</td>
<td>9.99 ± 0.42&lt;sup&gt;a&lt;/sup&gt;</td>
<td>14.8 ± 0.72&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Neutrophil (x10&lt;sup&gt;9&lt;/sup&gt;/L)</td>
<td>6.3 ± 0.34&lt;sup&gt;a&lt;/sup&gt;</td>
<td>8.89 ± 044&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Lymphocyte(x10&lt;sup&gt;9&lt;/sup&gt;/L)</td>
<td>1.4± 0.15&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.19 ± 0.28&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Eosinophil (x10&lt;sup&gt;9&lt;/sup&gt;/L)</td>
<td>1.3 ± 0.1</td>
<td>1.3 ± 0.1</td>
</tr>
<tr>
<td>Monocyte (x10&lt;sup&gt;9&lt;/sup&gt;/L)</td>
<td>1.0 ± 0.0</td>
<td>1.43 ± 0.13</td>
</tr>
</tbody>
</table>

Values with different superscripts are significantly (p < 0.05) different.
DISCUSSION

Diagnosis of acute laminitis is often confusing for the clinicians because of its close resemblance with conditions like azoturia, rupture of stomach or bladder and colic [24]. Examination of blood and biochemical parameters is among the most common methods of health assessment in animals [25], [26]. Horse owners need to identify cases of acute laminitis rapidly to minimize the life-threatening and long-term hoof damage of this serious disease [27].

In the present study, the hoof temperature during the dry season and rainy seasons did not differ significantly although they were higher than the normal value of 33°C [28]. When the ambient temperature is lower than 25°C and the hoof temperature is higher than 33°C, it could be a sign that a horse is developing laminitis [11]. The weight and fibrinogen levels of the horses did not differ during the period of study. Though the mean weight of the horses increased during the rainy season when compared with the dry season period, this may be due to increase in pasture intake at this period. The value of red blood indices increased significantly during the rainy season. This may be due to increase in pasture intake during this period [2]. The value of glucose was significantly higher during the rainy season compared with the dry season probably due to high sugar and starch in feed occasioned by excess feed intake by the horses during the rainy season. Feeds that are high in sugar and starch will cause blood glucose levels to rise sharply and quickly [29]. This is followed by a spike in insulin levels in the blood. Studies have shown the role of sugar and starch as the cause of laminitis [20]. These are carbohydrates that induce a glycaemic response that include release of insulin. The water soluble carbohydrates including sugar and fructan in cool season grasses have been used to justify the fructan theory of laminitis [30].

The significant increase in total white blood cell and differential counts during the rainy season compared with dry season agrees with the work of Moore et al. [18] but contradicts the work of Ogbanya et al. [31], who had decrease in total white blood cell and differential counts in horses showing chronic laminitis. Excessive pasture intake may contain high sugar in it. The horse may be unable to digest all the carbohydrates in the foregut, which then move into the hindgut and ferment in the caecum [32]. This results in endotoxins being absorbed into the blood stream and whole body inflammation. In the feet, this may lead to damage to other tissues as there is no expansion space for inflamed tissues, thereby resulting in laminitis [2]. Systemic inflammation leading to inflammatory injury to the laminar tissue has been reported in sepsis-related laminitis in horses [33].

Conclusion

The study highlighted increase in glucose level, RBC counts, Hb concentration, PCV, total leucocyte counts and neutrophil and lymphocyte counts during the rainy season compared to the dry season. These changes in these parameters may suggest early indicator of laminitis in horses. Future studies are required and may aid to create more robust cut off values to precisely predict future development of laminitis in individual horses.

Recommendations

Horses should be restricted on excessive consumption of high quality pasture. Increase exercise helps to maintain a healthy weight and keeps metabolism strong. High digestible fiber and low starch and sugar option feed should be given to horses.
REFERENCES


