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Impact of Temperature and Humidity on the Leukogram of Mature Thoroughbreds on Pasture

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Abstract: *Thirty retired Thoroughbreds had whole blood collected at three time points on 6 days in which various temperature and humidity conditions occurred. The ambient temperature (°F) and humidity were measured at the site at each sampling time and were added to obtain a Heat Index score (HI). A complete blood count and differential was measured on each sample and means were compared by 10 point HI ranges, published heat index ranges of potential heat stress (<130, 130-150, 150-180 >180), and day time high and low HI for the day of sampling. Although few horses had decreases out of the laboratory range of normal there was a significant decrease in total white blood cells, neutrophils, lymphocyte, monocytes and eosinophils as the HI elevated above was over 130. These decreases increased in magnitude as the HI increased.*

Keywords: *heat stress, heat index, white blood cells, leukogram*.

Abbreviations HI - Heat Index, CBC- complete blood count, WBC- white blood cell count

1. Introduction

The impact of heat stress has been studied in a myriad of animals including the horse. Environmental impact on animals on pasture has been shown to be influenced primarily by a combination of temperature and humidity.[1} The impact of these two factors is now routinely considered and the Heat Index is used to assess the potential for climatic conditions to cause stress or potential harm to animals housed in non-climate controlled housing. While complicated calculations for HI can be used, the most common method for determining the Heat Index is to add the temperature and the relative humidity.[1,2,3} If the HI during the evening decreases below stress threshold levels then recovery from heat stress may occur.[2]

Due to the horse's ability to sweat during elevated temperatures, it is generally accepted that horses are more heat tolerant than many species of domestic animals. The thermal neutral zone for mature hoses is considered to be from 40°F (5°C) to 77°F (25°F).[3] When temperatures are below the bottom threshold increased calories are required to maintain body temperature and condition. While humidity is important even at lower temperatures, the importance of the HI is most noticeable at elevated temperatures. Changes in grazing behaviors and decreased ability to thermal regulate have been observed in heat index ranges (table 1).[3] While it is believed that horses can adapt to higher Heat Index situations this takes at least 21 days in the temperature extreme and doesn't account for daily fluctuations.[3]

The environmental impact on horses has been studied to determine the impact on performance and physiological changes.[3,4,5] However, little has been done to study the impact of elevated heat indices on horses in pasture situations. This study used changes in the leukogram to indicate when environmental conditions begin to cause stress to the horse.

2. Materials and Methods

Study Animals

Thirty mature, retired thoroughbred gelding and mares were enrolled in the study. The age of the horses ranged from 5-15 years and were on the same farm pasture and rations located in central

Kentucky[a]. While the majority of the horses were bays, there were three grays and several chestnuts. One gelding became unmanageable and was replaced by another gelding of the same age during the study. The study took place during the late summer through the winter when appropriate Heat Indices were anticipated.

Table 1. *How air temperature and relative humidity affect horse cooling*

Air temperature (°F) + Relative humidity (%)	Horse cooling efficiency
Less than 130	Most effective
130-150	Decreased
Greater than 150	Greatly reduced
Greater than 180	Condition could be fatal if horse is stressed

Study Design

On days in which the heat index was anticipated to be in one of the four ranges listed in table 1, horses had blood samples taken in the morning, midday and late afternoon. All horses were sampled on the same days and time periods. Temperature and humidity were recorded in the pasture at each sampling. Two 7 ml EDTA tubes were collected from each horse at each sampling time and a complete blood count and differential were performed on the day of sample acquisition by a local laboratory.[c]

Data Management and Analysis

The laboratory results for each sampling time were collated and the mean and standard deviation were determined. The results were then analyzed utilizing a commercial statistical package.

3. Results

When results were examined in ten heat index point ranges (graph 1) the trend toward decreasing WBC parameters were seen as the heat index increased. This trend stopped at the severe heat index of >180. In order to do statistical evaluations, the changes in leukograms were evaluated by pooling the results in two different manners utilizing the previously published heat index ranges [table 1]; by the heat index high/low for the day and the heat index range at the time of sampling (table 2 and 3). While these unadjusted levels (for dehydration) rarely dropped below the normal range, significant depression in the total white blood cell count, neutrophils and monocytes occurred as the both as the heat index for the day maximum rose and the lowest value was in a higher range and if horses had samples taken during elevated indices above 130. Some significant drops were only seen when the indices were above 150 or 180 (i.e. lymphocytes and eosinophils) (graph 2). Further analysis revealed that the total white blood cell count, neutrophil count and monocyte count were also significantly depressed if the sampling occurred when the heat index was above 150 (table 2). Recovery occurred if the low for the day was below 130 (table 3).

Table 2. *Impact of elevated Heat Index for the day of sampling on blood parameters*

Day TI range	WBC Mean /SD	RBC Mean/SD	HGB Mean/SD	HCT Mean/SD	PLT Mean/SD	MCV Mean/SD	MCH Mean/SD	MCHC Mean/SD
155-182	6.607778b 0.959656	8.251 0.84946	13.58778 1.115031	39.9 3.293833	165.9333b 37.6899	48.61111 2.58927	16.52222 0.859299	34.04778a,.06 0.545533
147-160	6.528848b 1.124062	8.15214 1.151384	13.42587 1.720518	39.4415b 5.069098	164.1418b 40.16472	48.12585 5.50495	16.36384 1.862498	33.6561 3.570475
124-160	7.183146c.06 1.0712	8.163708 0.861623	13.45393 1.458494	39.64719 4.197059	178.4045c 37.39186	48.67416 2.443865	16.55056 0.793389	33.91798b.06 0.371925
125-138	7.486667d.06 1.099111	8.372556 0.854954	13.78667 1.268613	40.74d 3.644716	157.3444 38.54583	48.81111 2.529358	16.54444 0.832147	33.83778c 0.357329

Table 2. (cont.) *Impact of elevated Heat Index for the day of sampling on blood parameters*

Day TI range	NEUT% Mean /SD	LYMPH % Mean /SD	MONO% Mean /SD	BASO% Mean /SD	NEUT# Mean /SD	LYMPH# Mean /SD	MONO# Mean /SD	EOS# Mean /SD	BASO# Mean /SD
155-182	57.02222 8.457709	35.41111 8.2676	4.311111b 1.53944	0.044444 0.20608	3.68b 0.814616	2.328889 0.642434	0.27b 0.115902	0.182222b 0.13045	0.001111b 0.010482
147-160	56.81438 9.693788	34.78291 8.560161	4.256748b 1.562497	0.036523 0.18131	3.65949b 0.862123	2.280577b.07 0.646901	0.266134b 0.116731	0.183288b 0.128967	0.00013b 0.00111
124-160	57.80899 5.928178	33.88764 6.724829	4.842697c 2.01064	0.101124 0.301492	4.044944c 0.839477	2.398876 0.632899	0.324719c 0.130066	0.223596c 0.104958	0.001124b 0.01054
125-138	57.64444 8.000432	33.78889 7.947738	5.3c 1.834545	0.055556 0.229061	4.135556c 0.885979	2.463333d.07 0.695294	0.373333d 0.128927	0.221111c 0.102734	0.004444d 0.020608

Table 3. *Impact of elevated Heat Index by the temperature range at time of sampling on blood parameters*

TI range at sample collection	WBC Mean /SD	RBC Mean/SD	HGB Mean/SD	HCT Mean/SD	PLT Mean/SD	MCV Mean/SD	MCH Mean/SD	MCHC Mean/SD
Greater than 180	6.773333a 0.906274	8.41a 0.804449	13.87333a 1.029876	40.68a 3.166353	169.4333 44.15387	48.56667 2.661129	16.56667 0.897634	34.12b 0.761758
150-180	6.608667a 0.954424	8.0366b 0.854856	13.26267b 1.261404	38.95067b 3.664146	172.4533 36.7847	48.59333 2.52216	16.58b 0.873842	34.04267b,c 0.401223
130-150	7.424444b 1.144097	8.451333c 0.886278	13.95222c 1.337097	41.00444c 3.83307	168.6667 40.2147	48.64444 2.531188	16.57778c 0.868943	34.02444c 0.430842
Under 130	7.240609b 1.296313	8.449739d 1.140402	14.12435ad 3.332778	42.09826d 13.05597	166.0702 41.76621	48.88696 4.787239	16.97913b 3.558835	33.80174c 2.090641

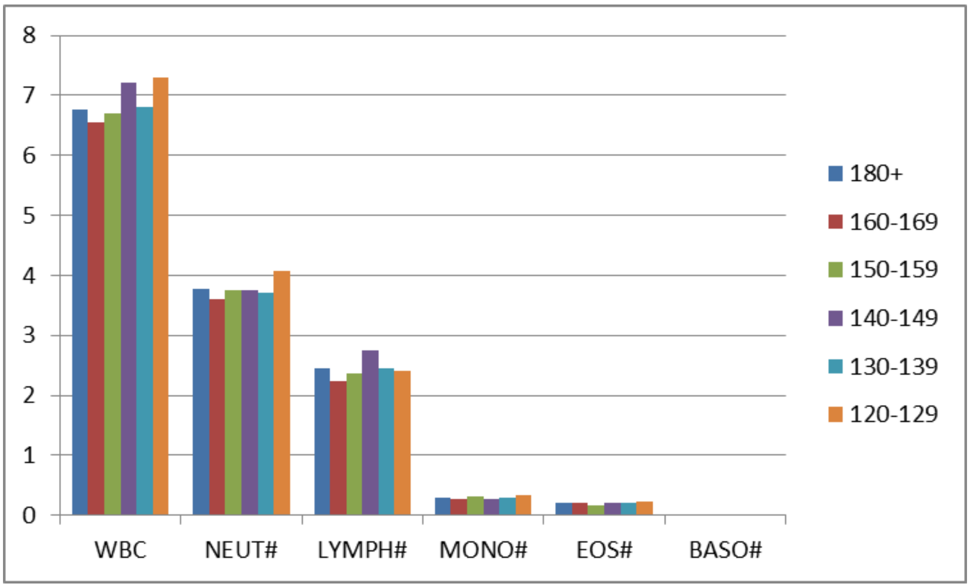
Table 4. (cont.) *Impact of elevated Heat Index by the temperature range at time of sampling on blood parameters*

TI range at sample collection	NEUT% Mean /SD	LYMPH% Mean /SD	MONO% Mean /SD	BASO% Mean /SD	NEUT# Mean /SD	LYMPH# Mean /SD	MONO# Mean /SD
Greater than 180	55.56667a 6.196124	36.33333 5.749563	4.566667 1.304722	0.1 0.305129	3.773333a 0.699721	2.45a 0.514446	0.303333a 0.099943
150-180	56.98667 7.485084	35.10667 7.796706	4.533333(.07) 1.934482	0.08 0.271293	3.659333a 0.815361	2.288b 0.616865	0.283333a 0.127758
130-150	56.07778 8.068082	35.81111(.08) 8.02066	4.833333 1.973435	0.111111 0.31427	4.03b 0.896468	2.592222c(.09)c 0.772762	0.345556b 0.150673
Under 130	57.3687b 8.417026	34.5913(.08) 9.944975	4.626087(.07) 1.98893	0.608696 1.241988	4.0629b 0.8607	2.651304(.09)d 0.882227	0.817391c 1.003495

EOS# Mean /SD	BASO# Mean /SD
0.21 0.14704	0a 0
b0.193333b (.06) 0.117568	0.002667 0.016111
0.218889 0.109437	0.005556 0.022906c
0.218261c(.06) 0.108414	0.033043d 0.074275

4. Discussion

It is important to note that the numbers were not adjusted for dehydration as the heat indices rose so the changes in the leukogram, while significant, are not as low as would be expected if corrected for dehydration**.** This is demonstrated when the leukogram, at heat indices above 180, are considered (tables 2,3; graph 1). Although significantly lower WBC are seen when compared to samples collected when the heat index is below 150, in almost every parameter there is a slight, though non-significant increase when compared the samples in the 150-180 range. This certainly could be attributed to hemo-concentrating due to dehydration and yet the decline is still significant when compared to lower temperature/humidity conditions. One previous study was performed in resting horses, in which the temperature and humidity were recorded. However, this study was complicated by the fact that the horses were also transported via aircraft for a long flight.[6] The horses demonstrated some increased white blood cell counts but both study groups were actually in the same HI range (130-150). As the researchers increased the temperature, they decreased the humidity.



Graph 1. *Outcomes by TI 10 point increments at time of sampling*

Various studies have suggested that heat stress in animals may not only decrease immune responses to vaccination but may be a factor in increased adverse reactions to systemic vaccines, in particular gram negative vaccines.[7,8] The onset of heat related stess in horses when environmental conditions are over a TI of 130 may be an indication of the potential for increased reactions and/or decreased responses to vaccination. Further studies evaluating immune responses in horses vaccinated in the various TI indices will add to the information regarding heat stress in horses and vaccine responses.

The impact of exposure to these environmental extremes should also be assessed in different breeds of horses. The ranges may be different in heavy horse breeds or ponies when compared to the Thoroughbreds used in this study. Furthermore studies have shown that horses can adapt to elevated temperatures over time.[9,10] These studies have shown that horses need to be in the elevated temperatures for at least 14 days and often did not take into consideration the humidity when assessing the ability to adapt. However at least one study did acclimate horses to elevated HI of 166.[11] The horses in this study were housed and tested in Kentucky and yet didn't show adaption. This is most likely due to the fact that Kentucky has significant variability in day and night temperatures and humidity that may make it more difficult for horses to heat acclimate. Repeating the study in horses housed in areas with constant HI above 130 would give an indication of the degree of adaption that occurs.

Finally, evaluating the WBC of horses that have samples drawn when undergoing heat stress may impact the levels of WBCs is important when assessing the leukogram of horse. While successive WBC sampling is often done to monitor horses for infection, a rising leukogram post admittance may actually be recovery from heat stress and not the onset of an infection

5. Conclusion

The horse has been considered to be heat tolerant so the majority of studies on the impact of environmental stress on horses has been performed during exercise. This study demonstrates that resting horses also show a degree of stress due to elevated temperature and humidity as measured by significant decreases in white blood cell count while dehydration is occurring. This may be an indication of times in which vaccination of horses may not give an optimal response or increase the risk of adverse reactions occurring. The decreases in the leukogram should also be taken into consideration when evaluating blood work collected in elevated climatic conditions.

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