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Sweat osmolarity shows intra-animal regional variation in the horse

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Abstract

Background

Sweating is important in regulating body temperature but can be a source of loss of both fluids and electrolytes. Although the process has been studied in horses, the variation in sweat osmolarity across the body has not been studied.

Objectives

This work describes an investigation to determine if there is regional variation in the osmolarity of sweat across different regions of the horse's body.

Animals

Ten horses were used in the study and were animals either stabled for riding lessons or had livery on-site.

Methods

Sweat samples were collected from animals following exercise and the osmolarity measurements were made using an Osmomat 030, Gonotec, Berlin. Values were analysed by paired t-tests and analysis of variance.

Results

Samples from the back and ears had statistically ($P<0.05$) lower osmolarity values than those seen in the neck and arm, with thigh values intermediate between the other two sets of values.

Conclusions and clinical importance

Previous studies have used osmolarity values based on the sweat collected from the horse's back. The current work demonstrates that these values are probably an underestimation, which may have implications for the composition and administration of rehydration compounds.

Introduction

Production of sweat is an important process in mammals as it allows for an evaporative cooling process which assists in regulation of body temperature, either at times of high ambient temperature, or during exercise when there is an increase in metabolic processes occurring in the body [1]. Horses have two different types of sweat glands; apocrine and merocrine, with the majority being apocrine [2]. In general merocrine glands are thought to be restricted to the hairless areas (e.g. the frog), whilst apocrine sweat glands are distributed across the entire body [2], meaning that all sweat seen on a horse can be regarded as being of apocrine origin. Since the apocrine sweat glands open into the hair follicles and in the horse there is one hair follicle per root hair, this means that across the normal hair distribution of the horse the hair follicle number is a representation of the number of apocrine sweat glands.

Based on this relationship between hair follicles and sweat glands, work was performed to estimate the density of sweat glands at different parts of the body at 46 loci across seven sites on the horse's body [2]. Values varied greatly across the body, from around 500 hairs cm^{-2} in the pubic region to over 1100 hairs cm^{-2} in lateral areas of the distal pelvic and thoracic limbs. Thus it is already clear that the distribution of sweat glands across the horse is not uniform.

Sweat is composed of a number of different chemicals, including ions (e.g. Cl^-, Na^+, K^+) and proteins (e.g. latherin). However, many previous publications employ generic osmolarity investigations, rather than looking at specific components of the sweat. Previous papers have been published on sweat composition in horses, including examples of osmolarity values. One such study found that the range was typically 290-320 mOsm/L [3]. There have also been differences detected in the osmolarity of sweat depending on environmental conditions e.g. 303 mOsm/L in cool dry conditions but 339 mOsm/L in hot dry conditions [4] and 535-565 mOsm/L following adrenaline infusion [5].

Regional sweat composition has been shown to vary in humans; i.e. sweat from different parts of the body have different osmolarity levels [6]. To our knowledge, no such investigation has been undertaken in the horse. This work describes an investigation to determine if there is regional variation in the osmolarity of sweat across different regions of the horse's body.

Materials and Methods

Collection and storage of sweat

Collections were carried out in Ceredigion, Wales and Florida, USA. Animals ranged from 2 to 17 years of age (mean = 9.6; SE = 3.04). Both mares and geldings were used for sampling, and animals included a range of different breeds (Quarter Horse, Irish Draft cross, British Warmblood, Welsh Section D ponies, Irish Sport Horse and Thoroughbred cross). All animals were sound at the time of sampling and samples were collected with the consent of the owners.

In a pilot study to investigate recovery of sweat, paper towels were shown to give a greater recovery than a number of commercially available swabs. Using this approach it was possible to replicate the pattern of differences in osmolarity at different parts of the human body, seen in Patterson *et al*. [6] as well as successfully collect samples from horses.

The areas of the horse which were to be used for sample collection (inner arms, back, ear, neck and inner thigh) were washed before horses were exercised for 20-30 minutes. Immediately after exercising sweat samples were collected from ten horses. During collections gloves were worn throughout to minimise the risk of contamination from human sweat. Collection of sweat was performed with towels at the five points across the horse as listed above. Immediately after collection samples were placed in individual plastic containers before being chilled at 4°C (for short-term storage) or frozen and stored in a freezer at -20°C where samples were not going to be used shortly after collection. Samples were maintained at this temperature until used.

Measurement of osmolarity of sweat

Immediately prior to analysis samples were raised to ambient temperature and were centrifuged twice at 1000***g*** for 3 minutes. From the liquid recovered post-centrifugation 50 µl of sweat was used for analysis from each sample. Osmolarity measurements were made using a freezing point depression osmometer (Osmomat 030, Gonotec, Berlin). This machine was calibrated using 0.300 and 0.850 Osmol/kg standards and working on the principle that deionised water freezes at 0°C and a solution of 1 Osmol/kg freezes at -1.858°C.

Statistical analysis

Sweat osmolality values from each body location were compared for kurtosis and skewness to determine if data were normally distributed. Six samples posed problems due to crystallisation of the samples and were removed from analysis. In all cases, there were samples from at least 8 horses per region and in all cases each horse provided samples from at least 4 regions. Data were analysed by paired t-tests to check for differences between anatomical regions. In addition, F-test supported t-tests were performed for inter-horse variation. Analysis of variance (ANOVA) was also performed following compensation for missing data values by the method of Li [7].

Results

As can be seen from Table 1, there are significant differences in the osmolarity values of sweat collected from different regions of the body. Differences were also detected following ANOVA calculations ($P<0.05$).

The higher osmolarity values do not coincide with those areas which have the highest density of hair follicles. Likewise, based on published skin thickness values [8], there is also no apparent relationship between the thickness of the skin and the osmolarity values.

Discussion

All of the values in this table are higher than the figures quoted in the literature e.g. [3, 4], although it is worth noting that the highest figure cited previously was as a result of adrenaline infusion prior to collection [5]. In the current work the horses had just completed exercise and so their adrenaline levels may have been elevated, giving a value higher than that seen in previous publications. In addition previous work concentrated on sweat samples collected from areas such as the back, which the current study suggests is one of the areas with the lowest osmolarity value. Given the disparity in sweat production at different regions of the body, it can be suggested that the most common site for sweat collection in previous work may not have given a true reflection of electrolyte balance. In turn this may mean that estimated loss of electrolytes may have previously been underestimated.

In conclusion, the data presented here identify that there is variation in the osmolarity between different anatomical sites in the horse and by inference this means that there are regional differences in the composition of the sweat.

References

1. McEwan-Jenkinson D, Elder HY, Bovell DL. Equine sweating and anihidrosis Part 1 – equine sweating. *Veter. Dermatol.* 2006; 17: 361-392
2. Watanabe A, Kanemaki N, Matsuura K. Distribution Densities of Hair Follicles in Racehorses. *Jap. J. Equine Sci.* 1993; 4: 55-60
3. Kingston JK, Geor RJ McCutcheon LJ. Rate and composition of sweat fluid losses are unaltered by hypohydration during prolonged exercise in horses. *J. Appl. Physiol.* 1997; 83: 1133-1143
4. Geor RJ, McCutcheon LJ. Thermoregulation and clinical disorders associated with exercise and heat-stress. *Comp. Continuing Educ. Practicing Veter*. 1996; 18: 436-445
5. Beeley JG, Eason R, Snow DH. Isolation and characterisation of latherin, a surface-activated protein from horse sweat. *Biochem. J.* 1986; 235: 645-650
6. Patterson MJ, Galloway SDR, Nimmo MA. Variations in regional sweat composition in normal human males. *Exp. Physiol.* 2000; 85.6: 869-875
7. Li C. Introduction to experimental statistics. 1st Ed. New York: McGraw-Hill, 1964
8. Wakuri H, Mutoh K, Ichikawa H, *et al*. Microscopic Anatomy of the Equine Skin with Special Reference to the Dermis. *Okajimas Folia Anat. Jap.* 1995; 72: 177-184

Region	Number of samples	Mean osmolarity value	S.E.M.
Arm	10	1.321 [a]	0.153
Back	10	0.842 [b]	0.092
Ear	8	0.882 [b]	0.082
Neck	8	1.533 [a]	0.229
Thigh	8	0.969 [a, b]	0.190

Table 1.
Mean osmolarity values (Osmol/kg) for different anatomical regions of the horse, together with the standard error of the mean (S.E.M.) for these samples. Regions which are not significantly different ($P > 0.05$ for t-test calculations) have the same superscript.