

The examination of biophysical parameters of skin (transepidermal water loss, skin hydration and pH value) in different body regions of ponies

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Abstract

The purpose of this study was to evaluate transepidermal water loss, skin hydration and skin pH in normal ponies. Sixteen ponies of both sexes were examined in the study. Measurements were taken from seven different sites: the neck region, the shoulder, thorax, lumbar, inguinal, lip region and the auricle. In each of the regions transepidermal water loss (TEWL), skin hydration and skin pH were measured. For transepidermal water loss, the lowest values were observed in the lumbar region (9.71g/hm²), while the highest values were observed in the lip region (22.35 g/hm²). In the case of skin hydration the lowest values were observed for the thorax region (2.13 CU), and the highest for the lip region (41.81 CU). For skin pH, the lowest results were obtained in the lumbar region (6.93), and the highest in the lip region (7.96).

Key words: skin biophysical parameters, TEWL, corneometry, skin pH, horses

Introduction

A variety of measurements of biophysical parameters, such as transepidermal water loss (TEWL), skin hydration (corneometry), as well as skin pH and erythema intensity have recently been used to complement other methods of examining skin. These non-invasive methods have been widely applied to skin examination in human medicine and, recently, in veterinary medicine, most commonly in dogs (Dirschka et al. 2004, Fluhr et al. 2006, Hightower

et al. 2008, Oh and Oh 2009, Hightower et al. 2010, Cornegliani et al. 2012). In human medicine, the parameters have been found useful to examine the skin in atopic dermatitis (Eberlein-König et al. 2000, Choi et al. 2003, Rudolph et al. 2004, Grupta et al. 2008) in order to evaluate the effectiveness of locally applied treatment (Biro et al. 2003, Loffler et al. 2003, Aschoff et al. 2009), and in case of contact dermatitis (Laudańska et al. 2003). Some of these parameters have been studied in veterinary medicine, most commonly in dogs (Beco et al. 2000, Matousek et al. 2002,

Watson et al. 2002, Young et al. 2002, Hester et al. 2004, Yoshihara et al. 2004, Oh et al. 2009, Shimada et al. 2009, Lau-Gillard et al. 2010), cats (Bourdeau et al. 2004, Szczepanik et al. 2011) and horses (Mayer et al. 1991). Changes in biophysical parameters of the skin (TEWL and skin hydration) have also been examined in dogs with atopic dermatitis (Hightower et al. 2008, Shimada et al. 2009, Hightower et al. 2010, Cornegliani et al. 2012) and pyoderma (skin pH) (Popiel et al. 2004).

Among the aforementioned methods, transepidermal water loss has been studied most frequently. Water moves passively from well hydrated layers of the skin to the less hydrated epidermis. Tewametry measures the pace of transepidermal water loss and describes the skin ability to retain water. The method is widely held to be a sensitive indicator of skin damage, and a growth of value for this parameter is attributable to epidermis damage (Shah et al. 2005, Fluhr et al. 2006, Gupta et al. 2008, Oh et al. 2009, Shimada et al. 2009).

A growth of value for this parameter, indicating epidermis damage has been observed, for example, in cases of human atopic dermatitis (Eberlein-König et al. 2000, Rudolph et al. 2004, Gupta et al. 2008) and canine atopic dermatitis (Hightower et al. 2008, Shimada et al. 2009, Hightower et al. 2010, Cornegliani et al. 2012).

Corneometry, the evaluation of skin hydration, is based on measures of electric capacitance of the stratum corneum and indicates the relative hydration of this epidermal layer. This method determines the water content of the outer layer of the stratum corneum at the depth of 10-20 μm to 60-100 μm (Rudolph et al. 2004, Aschoff et al. 2009). Changes of the value of this parameter have been confirmed in the case of trauma, metabolic disorders and local treatment of atopic dermatitis in humans and in dogs (Rudolph et al. 2004, Aschoff et al. 2009, Shimada et al. 2009).

Changes in skin pH have been examined in people, where an increase in pH was observed in atopic dermatitis, seborrheic dermatitis, acne, ichthyosis, contact dermatitis, and *Candida albicans* infections (Eberlein-König et al. 2000, Matousek et al. 2002, Schmid-Wendtner et al. 2006). Increase of pH in pyoderma in dogs has been shown by Popiel and Nicpoń (Popiel et al. 2004). Changes of this parameter connected with diet have also been examined in cats (Bourdeau et al. 2004).

In assessing the TEWL, skin hydration and skin pH it is vital to take into consideration other factors such as age, sex, breed, and anatomical site that may influence results in healthy animals. The influence of these factors on the biophysical parameters has been

examined in dogs (Mayer et al. 1991, Matousek et al. 2002, Watson et al. 2002, Young et al. 2002, Hester et al. 2004, Yoshihara et al. 2007, Oh et al. 2009, Shimada et al. 2009) and in cats (Bourdeau et al. 2004, Szczepanik et al. 2011).

With the exception of skin pH (Mayer et al. 1991, Matousek et al. 2002), no studies have so far investigated other biophysical parameters in horses. The purpose of this study was to examine the distribution of these biophysical parameters (TEWL, skin hydration and skin pH) in ponies of both sexes.

Materials and Methods

16 "Felin" ponies from the experimental farm of the University of Life Sciences in Lublin (Felin) were included in the study. The experimental group comprised 11 males and 5 females. The age of the animals ranged from 2 to 23 years (mean age 12.27 years). All horses were given a complete physical and dermatologic examination prior to taking the measurements. Only clinically healthy animals with no history of skin disease were included in the study. The animals were acclimatized in the test room (stable) two hours before the measurements were taken. The temperature in the room ranged from 20°C to 28°C and the relative humidity from 40% to 55%. The examination was performed in the second half of 2011. Before the measurement, hair was clipped to 1 mm length using Metzenbaum scissors. In a study by Watson et al. (Watson et al. 2002), clipping, as opposed to using a hair cutting machine, did not influence the results of TEWL. Care was taken not to damage the skin while clipping. The measurement was taken two minutes after hair clipping. Similarly, in earlier studies with cats hair was clipped before measurement in order to ensure better adherence of the probes to the skin surface (Szczepanik et al. 2010). Measurements were taken from seven different sites: the side of the neck, the shoulder region, the lateral thorax region, the lumbar region, the inguinal region, the lip region, the internal surface of the auricle. In each of the regions, transepidermal water loss, skin hydration and skin pH were measured. For each parameter six successive measurements were taken and the mean value was calculated. The assessment of the parameters was made by means of the Courage Khazaka Multi Probe Adapter 5 and the appropriate probes: the Tewameter[®]TM 300 probe (to measure TEWL, results given in g/hm^2), Corneometer[®]CM 825 (to measure skin hydration, results in corneometer units), Skin-pH-Meter[®]PH 905 (to measure skin pH, results on the pH scale). The same instrumentation was used in previous studies in dogs and cats (Young et al. 2002

Hester et al. 2004, Yoshihara et al. 2004, Shimada et al. 2009, Hightower et al. 2010, Szczepanik et al. 2011). For all parameters, the mean, standard deviation and median were calculated. Statistical analysis was conducted by the Student's t-test at P-values of $p = 0.05$ (Statistica 6.0 software). For each parameter, statistically significant differences were calculated between the results obtained in different regions. Additionally, statistically significant differences between the results for females and males were calculated, taking into consideration the distribution of parameters in the regions.

Results

For transepidermal water loss, the lowest values were observed in the lumbar region, while the highest values were observed in the lip region. The statistical analysis has shown that transepidermal water loss was

Table 1. TEWL in different body regions of ponies.

Body region	Mean g/hm ²	Standard deviation
The neck region	12.15	4.95
The shoulder region	18.21	5.53
The thorax region	14.21	6.21
The lumbar region	9.71	4.72
The inguinal region	11.89	7.01
The lip region	22.35	11.76
The auricle	9.96	7

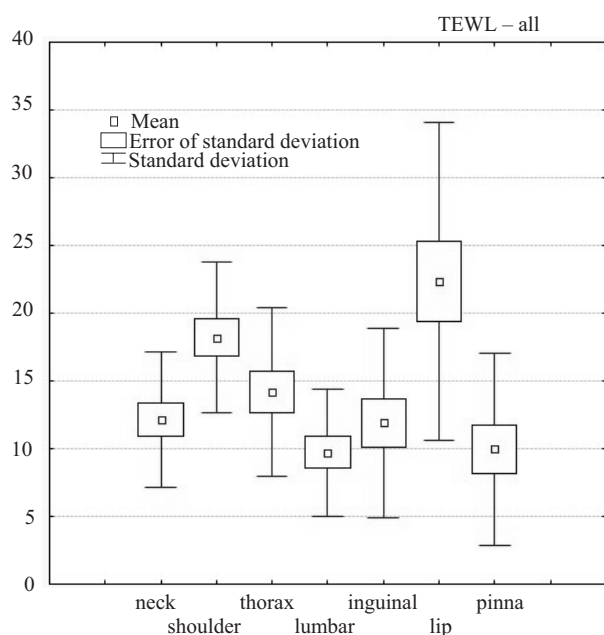


Fig. 1. Mean TEWL in different body regions of ponies

statistically significantly lower in the neck region as compared to the shoulder region ($p = 0.0028$), and the lip region ($p = 0.0032$). In case of the shoulder region, statistically significant differences were found in comparison to the lumbar region ($p = 0.000059$), the inguinal region ($p = 0.0082$) and the auricle ($p = 0.00092$). Statistically significant differences were also observed between the values for the thorax region and the lumbar region ($p = 0.028$) as well as the lip region ($p = 0.02$). There were statistically significant differences between the results obtained for the lumbar region and the lip region ($p = 0.0004$), the inguinal region and the lip region ($p = 0.0046$), the lip region and the auricle ($p = 0.001$). The results are presented in Table 1 and Fig. 1.

TEWL was slightly higher for males (14.24) than for females (13.7), but the difference was not statistically significant. No statistically significant differences were observed for the results between males and females for TEWL in corresponding body regions (the neck $p = 0.49$, the shoulder $p = 0.427$, the thorax $p = 0.06$, the lumbar region $p = 0.83$, the inguinal region $p = 0.59$, the lip $p = 0.31$, the auricle $p = 0.79$).

In case of skin hydration the lowest values were observed for the thorax region, and the highest for the lip region. It was found that the value for this parameter is statistically higher in the lip region compared to the neck region ($p = 0.0028$), the shoulder region ($p = 0.0011$), the thorax region ($p = 0.00074$), the lumbar region ($p = 0.0029$), the inguinal region ($p = 0.0025$), and the auricle ($p = 0.022$). Statistically significant differences were also found between the thorax and the auricle ($p = 0.047$). The results obtained are presented in Table 2 and Fig. 2.

Similarly to TEWL, there were no statistically significant differences between the values for males and females although skin hydration value in males (13.7) was higher than in females (6.61).

No statistically significant differences were observed between the results for males and females when comparing the results of skin hydration for corresponding body regions (the neck region $p = 0.95$, the shoulder region $p = 0.23$, the thorax region $p = 0.86$, the lumbar region $p = 0.19$, the inguinal region $p = 0.08$, the lip region $p = 0.33$, the auricle region $p = 0.61$).

For skin pH, the lowest results were obtained in the lumbar region (6.93), and the highest in the lip region (7.96). Statistical analysis has shown that skin pH is statistically significantly different in the neck region when compared to the shoulder region ($p = 0.0028$) and the lip region ($p = 0.0032$). Statistically significant differences were observed between the lip and the neck region ($p = 0.032$), the shoulder region ($p = 0.011$), the thorax region ($p = 0.0065$), the lum-

bar region ($p = 0.00370$) and the inguinal region ($p = 0.049$). The results are presented in Table 3 and Fig. 3.

Table 2. Skin hydration in different body regions of ponies.

Body region	Mean CU	Standard deviation
The neck region	5.87	13.31
The shoulder region	3.64	3.93
The thorax region	2.13	1.14
The lumbar region	5.32	16.35
The inguinal region	6.06	10.4
The lip region	41.81	42.81
Pinna	13.38	22.34

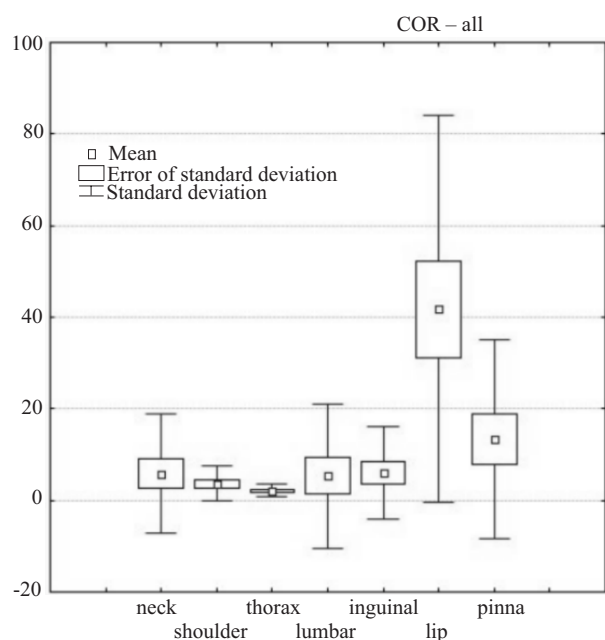


Fig. 2. Mean skin hydration (CU) in different body regions of ponies

Table 3. Skin pH in different body regions of ponies.

Body region	Mean pH	Standard deviation
The neck region	7.27	0.53
The shoulder region	7.09	0.71
The thorax region	7	0.68
The lumbar region	6.93	0.71
The inguinal region	7.29	0.71
The lip region	7.96	1.12
The auricle	7.56	1.45

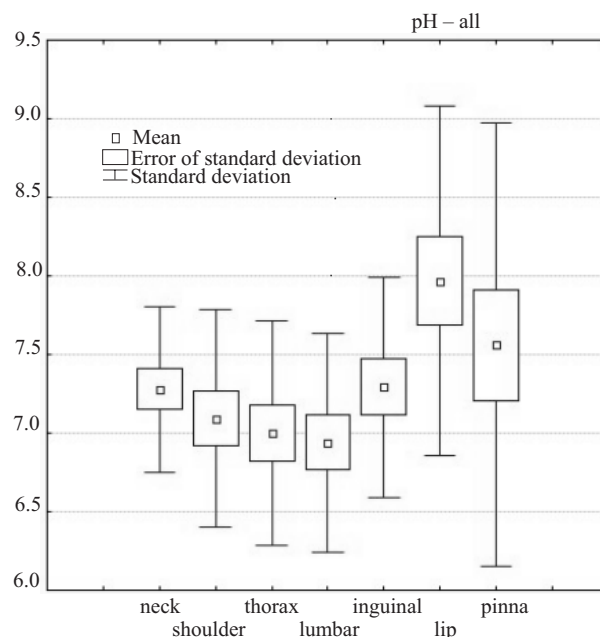


Fig. 3. Mean skin pH in different body regions of ponies

Similarly to the abovementioned parameters, no statistically significant differences were observed between males (pH 7.26) and females (pH 7.4). No statistically significant differences were found between the results for males and females comparing skin hydration in corresponding body regions (the neck region $p = 0.49$, the shoulder region $p = 0.12$, the thorax region $p = 0.36$, the lumbar region $p = 0.36$, the inguinal region $p = 0.63$, the lip region $p = 0.63$, the auricle region $p = 0.87$).

Discussion

Transepidermal water loss, skin hydration and skin pH measurements are considered to be useful techniques to assess the damage of skin in humans and are widely used to evaluate the skin barrier function in patients with atopic dermatitis as well as to evaluate the therapeutic efficacy of locally administered treatments (Eberlein-König et al. 2000, Dirschka et al. 2004, Grupta et al. 2008, Aschoff et al. 2009). Additionally, the measurement of the abovementioned parameters has recently been used to evaluate the skin condition in atopic dermatitis and pyoderma in dogs (Popiel et al. 2004, Hightower et al. 2008, Shimada et al. 2009, Cornegliani et al. 2012).

In veterinary medicine, information regarding these biophysical parameters in different diseases is limited (Popiel et al. 2004, Hightower et al. 2008, Shimada et al. 2009, Cornegliani et al. 2012), and most studies focused on TEWL in the course of the diseases in dogs only. There is also little information

available concerning the baseline values of these parameters in different animal species with most information obtained from canine studies (Oh et al. 2009, Shimada et al. 2009, Hightower et al. 2010, Cornegliani et al. 2012) and feline studies (Bourdeau et al. 2004, Szczepanik et al. 2011). With the exception of pH, TEWL and skin hydration have not been investigated in horses (Mayer et al. 1991). Neither TEWL nor skin hydration have so far been the object of research for this species.

Research conducted by other authors has pointed to statistically significant differences in case of TEWL between different dog breeds. Hestler et al. (2004) determined that TEWL values differ significantly between beagles and basset hounds. Differences in TEWL between breeds have also been described by Young et al. (2002) (beagles, fox terriers, Labrador retrievers and Manchester terriers were examined). Lau-Gillard et al. (2010) observed statistically significant differences between different dog breed with long and short hair. The observations made by these authors suggest that the TEWL values obtained should be interpreted according to breed, hence the single breed examined in the present study.

Numerous authors point to the fact that transepidermal water loss may differ according to body region. Research of this kind was conducted in humans (Marrakchi et al. 2007) as well as dogs (Watson et al. 2002, Oh et al. 2009, Yoshihara et al. 2009, Lau-Gillard et al. 2010) and cats (Szczepanik et al. 2011).

Oh et al. (2009) found that TEWL in beagles is the lowest for ear pinnae and for the lumbar region, as compared to other body regions, with the highest values found on the head and tail. Yoshihara et al. (2007), who also took measurements with Tewameter[®] TM 300, showed that the lowest values of transepidermal water loss are found in the lumbar region. In studies involving cats the lowest values for the parameter were also observed in the lumbar region (Szczepanik et al. 2011). A similar relationship was found in the present study in that TEWL was lowest in the lumbar region, which was statistically significantly different from values obtained for other regions. In contrast, Watson et al. (2002), determined that the lowest TEWL values were found in the ventrum as well as in the shoulder region and the limbs in Labrador dogs. Lau-Gillard et al. (2010) observed the lowest values in the ventrum and the highest between the shoulders in short-haired dogs. In long-haired dogs, the lowest values were noted on the side of the thorax and the highest in the axilla region. Due to the fact that TEWL has not so far been examined in horses, there is no possibility of direct comparison of the present results with the results obtained by other authors.

The studies of Hester et al. (2004) have also revealed that, similarly to TEWL, skin hydration is determined by breed. The authors have proved that the parameter has different values for basset hounds and beagles. Similar results were obtained by Young et al. (2002) who observed differing values for this parameter between the 4 breeds of dogs (beagles, fox terriers, Labrador retrievers and Manchester terriers). The studies of these authors indicate that, similarly to TEWL, skin hydration examination should be performed considering the breed of the animal. In humans, similarly to TEWL, skin hydration differs according to the body region (Marrakchi et al. 2007). Additionally, Oh et al. (2009) found statistically significant differences in skin hydration according to the body region in dogs. The highest value of the parameter was found in the pinna and the lowest value for the tail. In the present study, the best hydration was observed in the lip and the pinna came as the second. Similarly to TEWL, owing to the lack of studies pertaining to horses, there is no possibility of direct comparison of the present results with the results obtained by other authors.

Skin pH was the subject of a study conducted by Mayer and Neurad (1991), who investigated pH in several horse breeds in different body regions. The authors obtained the results ranging from 5.9 to 7.04 depending on the region assessed, which are values lower than those obtained in the present study. Similarly to our findings, Mayer and Neurad (1991) observed diverse pH depending on body region. The highest value was found in the lip/nasal region which corresponds with the results our study, where the highest pH was observed in the lip region. The lowest pH was found by Mayer and Neurad (1991) in the dorsal region while in our study the highest values were obtained for the lumbar region which was not investigated by the abovementioned authors. The differences of skin pH according to the body region was also described by Oh et al. (2009) in dogs. The authors confirmed statistically significant differences in skin pH in different body regions.

Young et al. (2002) assessed the influence of sex on TEWL, skin hydration, and skin pH in beagles, fox terriers, Labrador retrievers and Manchester terriers. In this study, the sex did not influence significantly the parameters, which correlates with the results obtained in the present study. Also Mayer and Neurad (1991), who assessed skin pH in horses, and Bourdeau et al. (2004) in cats, did not observe statistically significant differences between males and females. Opposite results were obtained in cats, where statistically significant differences between males and females were determined (Szczepanik et al. 2011).

The evaluation of biophysical parameters of the skin may be useful in case of atopic dermatitis. Studies conducted on dogs have shown that TEWL and skin hydration values differ between healthy and diseased animals, as well as undergo changes in the course of treatment. Therefore, it can be used to monitor its effectiveness (Shimada et al. 2009, Hightower et al. 2010, Cornegliani et al. 2012). There have not been any studies conducted so far concentrating on the biophysical parameter disorders in horses. Therefore, further research into the problem appears justified.

Similarly to dogs and cats, the values of biophysical parameters vary according to the body region examined. Further investigation into other horse breeds should aim at specifying the influence of breed and the reference values of the parameters discussed, as well as at assessing their appearance in the course of various skin diseases.

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References

- Aschoff R, Schwanebeck U, Bräutigam M, Meurer M (2009) Skin physiological parameters confirm the therapeutic efficacy of pimecrolimus cream 1% in patients with mild-to-moderate atopic dermatitis. *Exp Dermatol* 18: 24-29.
- Beco L, Fontaine J (2000) Corneometry and transepidermal water loss measurements in the canine species: validation of these techniques in normal beagle dogs. *Ann Med Vet* 144: 329-333.
- Biro K, Falk DT, Ochsendorf R, Kaufmann R, Wolf-Henning B (2003) Efficacy of dexpanthenol in skin protection against irritation: a double-blind, placebo-controlled study. *Contact Dermatitis* 49: 80-84.
- Bourdeau P, Taylor KW, Nguyen P, Biourge V (2004) Evaluation of the influence of sex, diet and time on skin pH and surface lipids of cats. *Vet Dermatol* 15: 41-69.
- Choi SJ, Song MG, Sung WT, Lee DY, Lee JH, Lee ES, Yang JM (2003) Comparison of Transepidermal Water Loss, Capacitance and pH Values in the Skin between Intrinsic and Extrinsic Atopic Dermatitis Patients. *J Korean Med Sci* 18: 93-96.
- Cornegliani L, Vercelli A, Sala E, Marsella R (2012) Transepidermal water loss in healthy and atopic dogs, treated and untreated: a comparative preliminary study. *Vet Dermatol* 23: 41-44
- Dirschka T, Tronnier H, Folster-Holst A (2004) Epithelial barrier function and atopic diathesis in rosacea and perioral dermatitis. *Br J Dermatol* 150: 1136-1141.
- Eberlein-König B, Schäfer T, Huss-Marp J, Darsow U, Möhrenschrager M, Herbert O, Abeck D, Krämer U, Behrendt H, Ring J (2000) Skin Surface pH, Stratum Corneum Hydration, Trans-epidermal Water Loss and Skin Roughness Related to Atopic Eczema and Skin Dryness in a Population of Primary School Children. *Acta Derm Venerol* 80: 188-191.
- Fluhr JW, Feingold KR, Elias PM (2006) Transepidermal water loss reflects permeability barrier status: validation in human and rodent in vivo and ex vivo models. *Exp Dermatol* 15: 483-492.
- Grupta J, Grube E, Ericksen MB, Stevenson MD, Lucky AW, Sheth AP, Assa'ad AH, Khurana Harshey GK (2008) Intrinsically defective skin barrier function in children with atopic dermatitis correlates with disease severity. *J Allergy Clin Immunol* 121: 725-730.
- Hester SL, Rees CA, Kennis RA, Zoran DL, Bigley KE, Wright AS, Kirby NA, Bauer JE (2004) Evaluation of Corneometry (Skin Hydration) and Transepidermal Water-Loss Measurements in Two Canine Breeds. *J Nutr* 134: 2110S-2113S.
- Hightower K, Marsella R, Creary E, Dutcher P (2008) Evaluation of trans-epidermal water loss in canine atopic dermatitis: a pilot study in beagle dogs sensitized to house dust mites. *Vet Dermatol* 19: 108.
- Hightower K, Marsella R, Flynn-Lurie A (2010) Effects of age and allergen exposure on transepidermal water loss in a house dust mite-sensitized beagle model of atopic dermatitis. *Vet Dermatol* 21: 88-95.
- Ludańska H, Reduta T, Szmitkowska D (2003) Evaluation of skin barrier function in allergic contact dermatitis and atopic dermatitis using method of the continuous TEWL measurement. *Rocz Akad Med Białymst* 48: 124-127.
- Lau-Gillard PJ, Hill PB, Chesney CJ, Budleigh C, Immonen A (2010) Evaluation of a hand-held evaporimeter (VapoMeter) for the measurement of transepidermal water loss in healthy dogs. *Vet Dermatol* 21: 136-145.
- Löffler H, Steffes A, Happle R, Effendy I (2003) Allergy and Irritation: An Adverse Association in Patients with Atopic Eczema. *Acta Derm Venereol* 83: 328-331.
- Marrakchi S, Maibach HI (2007) Biophysical parameters of skin: map of human face, regional, and age-related differences. *Cont Dermatitis* 57: 28-34.
- Matousek JL, Campbell KL (2002) A comparative review of cutaneous pH. *Vet Dermatol* 13: 293-300.
- Meyer W, Neurand K (1991) Comparison of skin pH in domesticated and laboratory mammals. *Arch Dermatol Res* 283: 16-18.
- Oh WS, Oh TH (2009) Measurement of transepidermal water loss from clipped and unclipped anatomical sites on the dog. *Aust Vet J*: 87: 409-412.
- Popiel J, Nicpoń J (2004) The correlation of skin pH of dogs with the pyoderm treatment before and after usage of surface-acting agents of known pH reaction. *Acta Sci Pol Med Vet* 3: 53-60.
- Rudolph R, Kownatzki E (2004) Corneometric, sebumetric and TEWL measurements following the cleaning of atopic skin with a urea emulsion versus a detergent cleanser. *Contact Dermatitis* 50: 354-358.
- Schmid-Wendtner MH, Korting HC (2006) The pH of the skin surface and its impact on the barrier function. *Skin Pharmacol Physiol* 19: 296-302.
- Shah JH, Zhai H, Maibach HI (2005) Comparative evaporimetry in man. *Skin Res Technol* 11: 205-208.
- Shimada K, Yoon J, Yoshihara T, Iwasaki T, Nishifuji K (2009) Increased transepidermal water loss and decreased ceramide content in lesional and non-lesional skin of dogs with atopic dermatitis. *Vet Dermatol* 20: 541-546.

- Szczepanik MP, Wilkołek PM, Adamek ŁR, Pomorski ZJ (2011) The examination of biophysical parameters of skin (transepidermal water loss, skin hydration and pH value) in different body regions of normal cats of both sexes. *J Feline Med Surg* 13: 224-230.
- Watson A, Fray T, Clarke S, Yates D, Markwell P (2002) Reliable Use of the ServoMed Evaporimeter EP-2 to Assess Transepidermal Water Loss in the Canine. *J Nutr* 132: 1661S-1664S.
- Yoshihara T, Endo K, Konno K, Iwasaki T (2004) A new method for measuring canine transepidermal water loss. *Vet Dermatol* 15: 39.
- Yoshihara T, Shimada K, Momoi Y, Konno K, Iwasaki T (2007) A new method of measuring the transepidermal water loss (TEWL) of dog skin. *J Vet Med Sci* 69: 289-292.
- Young LA, Dodge JC, Guest KJ, Cline JL, Kerr WW (2002) Age, Breed, Sex and Period Effects on Skin Biophysical Parameters for Dogs Fed Canned Dog Food. *J Nutr* 132: 1695S-1697S.