Localized parakeratotic hyperkeratosis in sixteen Boston terrier dogs

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Background – Although zinc responsive dermatosis is typically a disorder of Arctic breed dogs, this study identifies similar cutaneous lesions on the face and pressure points of Boston terrier dogs.

Hypothesis/Objectives – To document the clinical and histological features of localized parakeratotic hyperkeratosis of Boston terrier dogs, to determine if the lesions respond to zinc supplementation and to determine whether tissue zinc levels were decreased in affected versus unaffected dogs.

Material and methods – Sixteen Boston terrier dogs with similar gross and histological findings were identified retrospectively from two institutions. Follow-up information for nine dogs from one institution was obtained from referring veterinarians using a questionnaire. Tissue zinc levels were measured from formalin-fixed paraffin-embedded skin biopsy samples of affected and unaffected dogs using inductively coupled plasma mass spectrometry.

Results – Mild to severe parakeratotic hyperkeratosis with follicular involvement was present in all 16 cases. Of the nine dogs for which follow-up information was available, five dogs received oral zinc supplementation and four dogs had documented clinical improvement or resolution of dermatological lesions. The median skin zinc levels were not significantly different between affected and unaffected dogs.

Conclusions and clinical importance – To the best of the authors’ knowledge this is the first report of localized parakeratotic hyperkeratosis in Boston terrier dogs, some of which improved with oral zinc supplementation. Prospective studies in Boston terrier dogs are warranted to document potential zinc deficiency (serum and/or tissue levels, pre- and post-treatment) and to objectively assess response to zinc supplementation and other therapies.

Introduction

Parakeratosis is an abnormality of cornification in which the corneocytes of the stratum corneum retain their nuclei. Although parakeratotic hyperkeratosis can be a nonspecific histopathological finding (i.e. response to rapid skin turnover), more diffuse parakeratosis is seen in relatively few, often congenital or metabolic, diseases.

Parakeratotic hyperkeratosis has been associated with nonlethal forms of zinc malabsorption or deficiency in animals, including pigs, ruminants, camels, and dogs, as well as in humans. Two syndromes of zinc responsive dermatosis (ZRD) have been recognized in dogs. Syndrome I occurs predominantly in northern breed dogs (Siberian husky, Alaskan malamute, Samoyed) of any age, usually receiving a balanced diet. It is suspected to be an inherited impairment in zinc absorption or metabolism. Syndrome I has been reported also in other breeds, including a Boston terrier dog, German shepherd dog, flat coat retriever, great dane, Rhodesian ridgeback, a litter of Pharaoh hounds and mixed breed dogs. The most common clinical lesions are thick, adherent, crusted, hyperkeratotic plaques, often affecting the face. Syndrome II, also referred to as “generic dog food dermatosis,” occurs in young dogs fed imbalanced diets with a relative deficiency in zinc. With stricter guidelines regarding the manufacture of commercial dog food, this syndrome is now considered rare.

The aim of this retrospective study was to report the history, clinical signs, histopathological features and response to treatment of localized parakeratosis in Boston terrier dogs, for which there was an index of suspicion for ZRD. Serum zinc levels are not considered to be of value for the diagnosis of ZRD but can be used to document a response to therapy. It is of practical interest to consider alternative methods to determine whether serum or tissue zinc levels could be of value in the diagnosis of ZRD. Skin biopsies from affected and unaffected Boston terrier dogs were used to investigate the value of inductively coupled plasma mass spectrometry (ICP-MS) for quantitative analysis of tissue zinc levels in archived formalin-fixed paraffin-embedded (FFPE) samples.

Accepted 17 June 2016

Sources of Funding: This study was self-funded.

Conflict of Interest: No conflicts of interest have been declared.

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Material and methods

Inclusion criteria
A computer search for Boston terrier dogs with a clinical description of scale affecting the face and/or pressure points with a morphological diagnosis of “parakeratotic hyperkeratosis” was performed using the University of Pennsylvania Laboratory of Pathology and Toxicology database from 2004 to 2014. Additional cases and slides from 2005 to 2009 were obtained from the University of California, Davis. Medical records from dogs treated at the University of Pennsylvania were evaluated and a survey was sent to referring veterinarians to obtain additional follow-up information.

Medical record review
The following information was tabulated and evaluated: signalment (age, gender), history (vaccination status, age at biopsy, duration of clinical signs, concurrent systemic disease, diet, location of dermatological lesions and diagnostic investigations, which included dermatophyte culture, deep skin scrapings, bacterial culture and susceptibility, cell blood count, serum biochemistry panel and serum zinc levels) (Table 1). Treatment and outcome also were recorded (Table 2).

Histopathology
Evaluation of haematoxylin and eosin (H&E) stained sections was undertaken to determine the type and severity of hyperkeratosis, presence or absence of serum lakes, and pattern and cell type of dermal inflammation. Periodic acid Schiff (PAS) and Gram stains were performed on cases from the University of Pennsylvania.

Tissue zinc analysis
Tissue zinc levels were assessed from archived FFPE biopsy samples via inductively coupled plasma mass spectrometry (Perkin Elmer NexION 300D ICP-MS equipped with an S10 autosampler; Waltham, MA, USA). Eleven control samples of normal skin from the marginal tissue of FFPE benign neoplasms in Boston terrier dogs also were assessed.

The analytical standards were purchased from Seigniory Chemical Products (Champlain, NY, USA); trace metal grade acids were obtained from Fisher Scientific (Pittsburgh, PA, USA) and analytical grade gases were used. All dilutions were performed using in-house deionized water (resistivity ≥18 million ohms/cm) obtained from a Millipore® water purification system.

The ICP-MS was operated at a radiofrequency power of 1600 watts using helium gas in kinetic energy discrimination mode. The samples were heated to 140°C for up to 2 h to separate tissue from the surrounding embedding material. The remaining samples were weighed and digested with 70% nitric acid in a Teflon® PFA vial (Savillex; Eden Prairie, MN, USA) overnight in a Precision 14EG oven set at 70 ± 3°C for 12–18 h.

The digested samples were cooled to room temperature and 0.1 mL of the digests were diluted with deionized water to a final volume of 5.0 mL in addition of the internal standards at the final concentration of 20 parts per billion (114Ge, 115In, 67Y, 199Tb) for analysis of zinc. The performance of the ICP-MS and accuracy of the results were monitored by analysing standard reference materials from the National Institute of Standards and Technology (NIST 1577c) (Gaithersburg, MD, USA) and National Research Council Canada (Ottawa, ON, Canada).

After heating overnight in an oven with acid, the undigested green ink residue (used to mark the surgical margins of a tumour biopsy) from each sample was dried and then weighed. Final weight of the digested sample was determined by subtracting the weight of the dried undigested material from the total dried sample weight subjected to the digestion process.

Statistical analysis
Zinc tissue levels between affected and unaffected Boston terrier dogs were compared using StataCorp (Stata Statistical Software: Release 14, StataCorp LP; College Station, TX, USA) using the Wilcoxon signed rank test with significance set at $P < 0.05$.

Results

Signalment, history and physical examination
Sixteen Boston terrier dogs met the inclusion criteria based on histopathological abnormalities; nine FFPE samples were available for zinc tissue analysis and nine medical records were available for clinical follow-up. The median age of onset was 3.5 months (range 1–42) (Table 1) although one case (Dog 9) was excluded because the dog was adopted as an adult with existing lesions. There were four males and five females. The diet was recorded in three cases and all of these dogs were receiving commercially balanced diets.

Eight of nine dogs (89%) had thick, dry, adherent scale affecting the pinnal margins (sometimes notched) (Figure 1a). Five of nine dogs (56%) had thick, dry, adherent scale, erythema and alopecia affecting the haired skin of the dorsal muzzle. Two of nine dogs (22%) had scale affecting the hocks and one dog each had scale affecting the elbows and the tail. Skin lesions were symmetrical and nonpruritic except in one case (Dog 3) that had concurrent atopic dermatitis. One dog had concurrent hyperadrenocorticism (HAC).

Previous treatment
Eight of nine dogs had received various treatments including topical and systemic antimicrobial therapy, corticosteroids and topical retinoid therapy (Table 2) with no improvement.

Diagnostic investigation
Deep skin scraping was negative (in eight of nine dogs); dermatophyte culture was negative (in five of nine dogs) and bacterial culture was negative (in one of nine dogs). There were no relevant haematological or biochemical findings. One dog had confirmed HAC (Table 1).

Dermatohistopathology
The consistent histopathological abnormality in all dogs was moderate (seven dogs) to severe (nine dogs) parakeratotic hyperkeratosis (Figure 1b) with intracorneal serum lakes evident in cases with more pronounced parakeratotic hyperkeratosis. Parakeratosis occasionally involved follicular infundibulum. Focal to multifocal orthokeratotic hyperkeratosis was also evident (10 of 16 dogs). Epidermal and follicular acanthosis and spongiosis ranged from absent (two dogs) to severe. Mild to moderate superficial perivascular lymphoplasmacytic dermatitis was evident in all dogs with fewer neutrophils, perivascular mast cells and melanophages present, in some cases. Low numbers of Malassezia spp. were identified on H&E and PAS stains from the bridge of the nose in two dogs. Few scattered Gram-positive bacterial cocci were identified on the skin surface in two dogs.

Zinc supplementation and response
Five of nine dogs received oral zinc supplementation at an elemental zinc dose range from 0.72 to 10.1 mg/kg per
Table 1. Signalment, history and diagnostic tests of Boston terriers with hyperkeratosis

<table>
<thead>
<tr>
<th>Dog</th>
<th>Sex</th>
<th>Age of onset (months)</th>
<th>Hairy skin of dorsum</th>
<th>muzzle</th>
<th>Pinnae</th>
<th>Pressure points</th>
<th>Concurrent disease</th>
<th>Diet</th>
<th>Pre-biopsy diagnostic tests</th>
<th>Other diagnostic tests</th>
<th>Serum Zn levels; normal = 0.7–2.0 ppm; (10.7–30.6 μmol/L)</th>
<th>Tissue Zn levels ppm; (mmol/kg)</th>
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<tbody>
<tr>
<td>1</td>
<td>M</td>
<td>4</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>U</td>
<td>U</td>
<td>Negative</td>
<td>Negative</td>
<td>NP</td>
<td>NP</td>
</tr>
<tr>
<td>2</td>
<td>FS</td>
<td>3</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>U</td>
<td>U</td>
<td>Negative</td>
<td>Negative</td>
<td>NP</td>
<td>WNL</td>
</tr>
<tr>
<td>3</td>
<td>MC</td>
<td>1</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>Pressure points</td>
<td>U</td>
<td>U</td>
<td>Negative</td>
<td>Negative</td>
<td>Allergic dermatitis, Hill’s z/d</td>
<td>NP</td>
</tr>
<tr>
<td>4</td>
<td>MC</td>
<td>3</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>U</td>
<td>U</td>
<td>Negative</td>
<td>Negative</td>
<td>NP</td>
<td>NP</td>
</tr>
<tr>
<td>5</td>
<td>FS</td>
<td>42</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>U</td>
<td>U</td>
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<td>Negative</td>
<td>Enterobacter sp.</td>
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<tr>
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<td>F</td>
<td>1</td>
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<td>Yes</td>
<td>No</td>
<td>No</td>
<td>U</td>
<td>U</td>
<td>Negative</td>
<td>Negative</td>
<td>NP</td>
<td>NP</td>
</tr>
<tr>
<td>7</td>
<td>FS</td>
<td>24</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>Pressure points</td>
<td>U</td>
<td>NP</td>
<td>Negative</td>
<td>NP</td>
<td>NP</td>
<td>NP</td>
</tr>
<tr>
<td>8</td>
<td>MC</td>
<td>15</td>
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<td>No</td>
<td>No</td>
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<td>U</td>
<td>NP</td>
<td>Negative</td>
<td>NP</td>
<td>Several months post biopsy – WNL</td>
<td>NP</td>
</tr>
<tr>
<td>9</td>
<td>FS</td>
<td>U</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Pressure points</td>
<td>U</td>
<td>NP</td>
<td>Negative</td>
<td>Thrombocytosis</td>
<td>Elevated alkaline phosphatase, hyperphosphatemia, hypercholesterolemia (6 months post biopsy, 15 months post Zn supplementation)</td>
<td>111 (1.638) (7 months post Zn supplementation)</td>
</tr>
</tbody>
</table>

U unknown, NP not performed, WNL within normal limits, MC Male castrate, FS Female spayed, M Male, F Female.
<table>
<thead>
<tr>
<th>Dog</th>
<th>Oral antibiotic pre biopsy</th>
<th>Topical medication pre biopsy</th>
<th>Other med pre biopsy</th>
<th>Oral Zn type and dose</th>
<th>Time to response for Zn supplementation (months)</th>
<th>Duration Zn supplementation (months)</th>
<th>Recurrence</th>
<th>Other medications post biopsy for cutaneous lesions</th>
<th>Medications for concurrent diseases</th>
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<tbody>
<tr>
<td>1</td>
<td>Amoxicillin/clavulanic acid</td>
<td>No</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>2</td>
<td>Cefadroxil monohydrate</td>
<td>Gentamicin sulfate/betamethasone valerate/clotrimazole solution</td>
<td>NA</td>
<td>Gluconate (unknown formulation) (5 mg/kg once daily = 0.72 mg/kg elemental Zn)</td>
<td>≤4 (owner did not return for recheck until this time)</td>
<td>8</td>
<td>Yes: 18 months after discontinuing Zn</td>
<td>Enrofloxacin</td>
<td>Ketoconazole</td>
</tr>
<tr>
<td></td>
<td>Amoxicillin/clavulanic acid</td>
<td>Miconazole nitrate 2%/chlorhexidine gluconate 2% shampoo</td>
<td>NA</td>
<td>Miconazole nitrate 2%/chlorhexidine gluconate 2% shampoo</td>
<td>≤4</td>
<td>24</td>
<td>No</td>
<td>No</td>
<td>For allergic dermatitis: Prednisone Modified ciclosporin</td>
</tr>
<tr>
<td></td>
<td>Trimethoprim/sulfmethoxazole</td>
<td>Prednisone</td>
<td>Sulfate (Saveway Compounding Pharmacy, Newark, DE, USA) (2.18 mg/kg elemental Zn)</td>
<td>2–4</td>
<td>24</td>
<td>No</td>
<td>No</td>
<td>Miconazole nitrate 2% cream</td>
<td>No</td>
</tr>
<tr>
<td></td>
<td>Amoxicillin/clavulanic acid</td>
<td>Salicylic acid gel 6.6%</td>
<td>Hydrocortisone leave-on lotion</td>
<td>2</td>
<td>24</td>
<td>No</td>
<td>No</td>
<td>Miconazole nitrate 2% cream</td>
<td>No</td>
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<tr>
<td></td>
<td>Prednisone</td>
<td>Loratidine</td>
<td>Micro-emulsion shampoo</td>
<td>2</td>
<td>24</td>
<td>No</td>
<td>No</td>
<td>Miconazole nitrate 2% cream</td>
<td>No</td>
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<tr>
<td>3</td>
<td>Cefalexin</td>
<td>Methionine (NutriVed Zinpro Chewable Tablets; Vedco, St. Joseph, MO, USA) (3 mg/kg once daily = –0.85 mg/kg elemental Zn)</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>Salicylic acid gel 6.6%</td>
<td>No</td>
</tr>
<tr>
<td></td>
<td>Amoxicillin/clavulanic acid</td>
<td>Miconazole nitrate 2%/chlorhexidine gluconate 2% shampoo</td>
<td>NA</td>
<td>Miconazole nitrate 2%/chlorhexidine gluconate 2% shampoo</td>
<td>≤1</td>
<td>24</td>
<td>Yes: 4 months after switch from once daily to q48-72 h dosing</td>
<td>Mupirocin ointment 2%</td>
<td>Pentoxifylline</td>
</tr>
<tr>
<td>4</td>
<td>Cefpodoxime</td>
<td>Gentamicin sulfate/betamethasone valerate/clotrimazole solution</td>
<td>NA</td>
<td>Gentamicin sulfate/betamethasone valerate/clotrimazole solution</td>
<td>≤1</td>
<td>24</td>
<td>Yes: 4 months after switch from once daily to q48-72 h dosing</td>
<td>Mupirocin ointment 2%</td>
<td>Pentoxifylline</td>
</tr>
<tr>
<td></td>
<td>Amoxicillin/clavulanic acid</td>
<td>Mibramycin oxime/lyperuron</td>
<td>Miconazole nitrate 2%/chlorhexidine gluconate 2% shampoo</td>
<td>≤1</td>
<td>24</td>
<td>Yes: 4 months after switch from once daily to q48-72 h dosing</td>
<td>Mupirocin ointment 2%</td>
<td>Pentoxifylline</td>
<td></td>
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<tr>
<td></td>
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<td>Rpron/smalopran</td>
<td>Ceramide shampoo</td>
<td>≤1</td>
<td>24</td>
<td>Yes: 4 months after switch from once daily to q48-72 h dosing</td>
<td>Mupirocin ointment 2%</td>
<td>Pentoxifylline</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>Enrofloxacin</td>
<td>No</td>
<td>No</td>
<td>Salicylic acid gel 6.6%</td>
<td>≤7 (owner did not return for recheck until this time)</td>
<td>9</td>
<td>NA: died due to suspected pulmonary thromboembolism</td>
<td>Seborrhoea shampoo and spray</td>
<td>No</td>
</tr>
<tr>
<td></td>
<td>Cefpodoxime</td>
<td>No</td>
<td>No</td>
<td>Salicylic acid gel 6.6%</td>
<td>≤7 (owner did not return for recheck until this time)</td>
<td>9</td>
<td>NA: died due to suspected pulmonary thromboembolism</td>
<td>Seborrhoea shampoo and spray</td>
<td>No</td>
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<tr>
<td>6</td>
<td>Euthadixine</td>
<td>No</td>
<td>No</td>
<td>Miconazole nitrate 2%/chlorhexidine gluconate 2% shampoo</td>
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<td>9</td>
<td>NA: died due to suspected pulmonary thromboembolism</td>
<td>Tretinoin cream 0.1%</td>
<td>No</td>
</tr>
<tr>
<td>7</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>Miconazole nitrate 2%/chlorhexidine gluconate 2% shampoo</td>
<td>≤7 (owner did not return for recheck until this time)</td>
<td>9</td>
<td>NA: died due to suspected pulmonary thromboembolism</td>
<td>Tretinoin cream 0.1%</td>
<td>No</td>
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<tr>
<td>8</td>
<td>Cefpodoxime</td>
<td>Ceramide shampoo</td>
<td>Essential fatty acids</td>
<td>≤1</td>
<td>24</td>
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<td>Mupirocin ointment 2%</td>
<td>Pentoxifylline</td>
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<tr>
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<td>No</td>
<td>No</td>
<td>Essential fatty acids</td>
<td>≤1</td>
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<td>Mupirocin ointment 2%</td>
<td>Pentoxifylline</td>
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<tr>
<td>9</td>
<td>No</td>
<td>Tretinoin cream 0.1%</td>
<td>No</td>
<td>Tretinoin cream 0.1%</td>
<td>≤7 (owner did not return for recheck until this time)</td>
<td>9</td>
<td>NA: died due to suspected pulmonary thromboembolism</td>
<td>Tretinoin cream 0.1%</td>
<td>No</td>
</tr>
</tbody>
</table>

For hyperadrenocorticism: Triptans

NA no answer
os once daily. (Table 2) One dog was lost to follow-up. Two dogs (dogs 2, 3) receiving zinc gluconate and zinc sulphate (Saveway Compounding Pharmacy; Newark, DE, USA), respectively, improved within 4 months of supplementation. Three dogs received zinc methionine (NutriVed Zinpro Chewable Tablets, Vedco; St. Joseph, a

Figure 1. (a) Clinical signs of localized parakeratotic hyperkeratosis in Boston terrier dogs. Characteristic thick, dry, adherent scaling along a notched pinnae margin of a 4-month-old female Boston terrier dog. Lesions were not biopsied; they resolved with zinc methionine (elemental zinc 3 mg/kg once daily). (b) Histopathological findings in Boston terriers with localized parakeratotic hyperkeratosis. Severe parakeratotic hyperkeratosis overlying orthokeratotic hyperkeratosis, both of which extend into follicular infundibula, with moderate acanthosis and moderate superficial perivascular (lymphoplasmacytic) dermatitis.
Lesions recurred in one dog (Dog 2) when the supplement was withdrawn. One dog (Dog 8) improved after 2 years of zinc methionine but developed new lesions when the frequency of supplementation was reduced. Zinc sulphate was then administered with improvement noted after 1 month of supplementation but lesions recurred within a year despite continued treatment. Zinc methionine was restarted with improvement reported within 2 months, but the dog was then lost to follow-up. In one dog (Dog 9), zinc methionine was administered in conjunction with topical retinoid (Retin-A 0.1%, Valeant Pharmaceuticals; Bridgewater, NJ, USA) and salicylic acid cream (Solve-Ker, VetriMax; College Station, TX, USA) with mild lesion improvement. Serum zinc levels measured at 1.43 ppm (21.88 μmol/L) were within the reference range. The dog died from unrelated causes.

Skin tissue zinc levels
Zinc levels were reported as parts per million (ppm) and mmol/kg on a dry matter basis (Table 1). The median skin tissue zinc level for all 20 samples was 77.0 ppm (mean: 88.8; range 18.6–202) = 1.178 mmol/kg (mean: 1.358; range 0.284–3.090). In samples from eight affected dogs, the median zinc level was 76.8 ppm (mean: 68.5; range: 18.6–102) = 1.175 mmol/kg (mean: 1.048; range 0.284–1.560); Dog 9 (111 ppm) = 1.698 mmol/kg was excluded from the data analysis due to zinc supplementation prior to biopsy. In the 11 control samples (age range at time of biopsy: 48–179 months), the median zinc level was 84.7 ppm (mean: 104.3; range: 57.2–202) = 1.295 mmol/kg (mean: 1.595; range: 0.875–3.090). A statistically significant difference was not found between abnormal skin with parakeratotic hyperkeratosis and normal skin (P = 0.29, Wilcoxon signed rank test).

Discussion
This retrospective study describes 16 Boston terrier dogs affected with a scaling and crusting dermatosis affecting the dorsal muzzle, pinna and pressure points; characterized by moderate to severe parakeratotic hyperkeratosis. The histopathological features, in conjunction with the type and distribution of clinical lesions and the response to zinc supplementation in four of these dogs (dogs 2, 3, 8, 9), are suggestive of ZRD, as reported in Arctic breed dogs. A major difference is that these affected Boston terrier dogs had a much earlier age of onset (median 3.5 months old) than previously reported ZRD cases in other breeds (median 36 months old).7

Five of nine dogs received oral zinc supplementation. One dog was lost to follow-up, but four dogs demonstrated either improvement in clinical lesions or complete resolution of clinical signs. There were limited cases in this study to make a statistical comparison between the type of zinc supplement used, but the zinc dosage was within the published dose range of 2–3 mg/kg p.o. daily of elemental zinc.7 No significant difference in efficacy has been reported between types of zinc supplementation for ZRD,7 although oral supplementation of a zinc chelate with an amino acid (e.g. zinc methionine) may be more bioavailable than other forms.16 When zinc sulphate, zinc acetate and zinc oxide administration was compared in normal puppies, zinc sulphate was found to have the highest absorption rate.17

Of four dogs receiving zinc supplementation with follow-up information, clinical improvement occurred in one case within the first six weeks of supplementation, whereas the remaining cases required several months of treatment. In previously reported ZRD dogs, a response to zinc supplementation was typically noted within the first six weeks of treatment.7 In humans with zinc deficiency about 70% of patients improve within 6 months of zinc supplementation.19 A longer follow-up period may be indicated for Boston terrier dogs to determine a response to therapy. There were no adverse effects from zinc supplementation reported in this study, which is similar to the low incidence of adverse events previously reported for other breeds.7

Previous studies of zinc deficiency in dogs have investigated zinc levels in tissue, as well as in serum, leukocytes and hair.15,18 In this study, one dog (Dog 6) had a decreased serum zinc level prior to supplementation. Two dogs (dogs 3, 9) had serum zinc levels analysed post-supplementation and whereas both values were within the normal range, clinical lesions had improved only moderately. Dogs with ZRD have been reported to have significantly lower mean serum zinc concentrations than normal dogs, but there was an overlap in values; zinc concentrations in hair and leukocytes are similar between affected and normal dogs.15 It has been concluded that a low serum zinc concentration may, at best, be used to corroborate a diagnosis of ZRD if there are consistent clinical lesions and histopathological changes.15

To the best of the authors’ knowledge, this study is the first to report zinc analysis of FFPE canine skin samples. The median skin tissue zinc levels were similar in samples from affected and unaffected Boston terrier dogs, but the lowest levels of zinc were from cases with suspected ZRD. There are significant limitations to the interpretation of these findings as tissue samples were neither agematched or from similar locations in the control dog population. Further evaluation of this technique using samples from Arctic breed dogs affected with confirmed ZRD, both before and after treatment may be of value.

Although the clinico-pathological findings in these cases does resemble ZRD in Arctic breed dogs, other congenital or metabolic diseases characterized by parakeratosis have been reported. A mild form of hereditary nasal parakeratosis has been reported in young Labrador retriever dogs presenting with dry, rough, proliferative, adherent accumulation of keratin affecting the planum nasale and rarely the footpads.19,20 Three of eleven affected dogs in one study were treated with zinc methionine but failed to demonstrate any improvement.19 More severe congenital follicular parakeratosis has been reported in Labrador retrievers, Rottweilers and a Siberian husky with multifocal adherent plaques of frond-like keratin in conjunction with stunted growth and other birth defects.21–23 An acquired form of follicular apoptosis and parakeratotic casts that may respond to immunosuppressive therapy has been described in adult Labrador retrievers.24

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Superficial necrolytic dermatitis (SND) is a nonbreed-specific metabolic disease characterized by severe parakeratotic hyperkeratosis and oedema of the stratum spinosum, and hyperplasia of the stratum basale. Erosions and ulcers with thick, adherent crusts affect the footpads, mucocutaneous junctions (lips, eyelids, clawbeds, anus) and pressure points.²⁵ The aetiology is uncertain, but hypoaominocidemia and abnormal zinc metabolism have been proposed as contributory factors.²⁶ However, as in humans with a similar disease of necrolytic migratory erythema (NME),²⁷ zinc supplementation alone does not provide clinical resolution of skin lesions in dogs with SND.²⁵ Although SND is a differential diagnosis for parakeratotic hyperkeratosis, the Boston terrier dogs in this study could be distinguished readily based on clinical distribution (lack of pawpad and mucocutaneous junction involvement) as well as the lack of classic histopathological findings (spinous layer pallor, basal cell hyperplasia).

In conclusion, this case series describes a unique clinical syndrome of localized parakeratotic hyperkeratosis that affects the face and pressure points of Boston terriers. Additional prospective studies are needed to clarify the role of zinc in the pathogenesis and treatment of this disease.

Acknowledgements
The authors thank Dorothy C. Brown for statistical support, Thelma L. Gross for histopathology evaluation and the referring veterinarians for follow-up information.

References

Résumé
Contexte – Bien que la dermatose répondant au zinc soit typiquement une atteinte des races de chiens nordiques, cette étude met en évidence des lésions cutanées identiques sur la face et les points de pression de boston terriers.
Hypothèses/Objectifs – Documenter les critères cliniques et histopathologiques de l’hyperkératose parakeratotique des boston terriers, déterminer si les lésions répondent à la supplémentation en zinc et si les taux de zinc tissulaires étaient plus faibles chez les chiens atteints par rapport aux chiens sains.
Matériel et méthodes – Seize boston terriers présentant des lésions macroscopiques et histologiques identiques ont été identifiés rétrospectivement dans deux centres. Les informations de suivi de neuf...
chiens d’un centre ont été obtenues des vétérinaires référents à l’aide d’un questionnaire. Les taux de zinc tissulaires ont été mesurés à partir de biopsies cutanées fixées dans le formol des chiens atteints et sains par spectrométrie de masse couplée à un plasma.

**Résultats** – Une hyperkératose parakératosique modérée à sévère avec atteinte folliculaire était présente pour tous les 16 cas. Sur les neufs chiens pour lesquels nous avions un suivi, cinq chiens ont reçu une supplémentation orale en zinc et quatre chiens présentaient une amélioration clinique ou une résolution des lésions dermatologiques. Les niveaux moyens de zinc cutané n’étaient pas significativement différents entre les chiens atteints et sains.

**Conclusions et importance clinique** – A la connaissance des auteurs ceci est le premier article concernant l’hyperkératose parakératosique des bostons terriers, dont certains se sont améliorés par la supplémentation en zinc oral. Des études prospectives chez les bostons terriers sont nécessaires pour documenter la déficience potentielle en zinc (sérique et/ou tissulaire, pré- et post-traitement) et d’objectivement évaluer la réponse à la supplémentation en zinc et à d’autres traitements.

**Resumen**
**Introducción** – Aunque la dermatosis que responde a zinc suele ser un trastorno de perros de razas árticas, este estudio identifica lesiones cutáneas similares en la cara y zonas de presión de los perros de raza Boston Terrier.

**Objetivos/Hipótesis** – Documentar las características clínicas e histológicas de la hiperceratosis parakeratótica localizada de perros de raza Boston Terrier, para determinar si las lesiones responden a la administración de suplemento de zinc y determinar si los niveles de zinc se redujeron en el tejido afectado en comparación con los perros no afectados.

**Material y métodos** – Dieciséis perros de raza Boston Terrier con hallazgos macroscópicos e histológicos similares fueron identificados retrospectivamente en dos instituciones. El seguimiento clínico se obtuvo en nueve perros de una institución utilizando un cuestionario para los veterinarios. Los niveles de zinc en tejido se midieron a partir de muestras de biopsia de piel incluidas en parafina fijadas con formalina y cortadas en parafina para la espectrometría de masas de plasma eludida con inducción.

**Resultados** – En todos los perros hubo hiperceratosis parakeratótica de leve a afectando los folicúles. De los nueve perros de los cuales se obtuvo información de seguimiento, cinco perros recibieron suplementos de zinc oral y cuatro perros mostraron mejoría clínica o resolución de las lesiones dermatológicas. Los niveles medios de zinc en piel no fueron significativamente diferentes entre los perros afectados y no afectados.

**Conclusiones e importancia clínica** – A entender de los autores este es el primer artículo publicado de hiperceratosis parakeratótica localizada en perros de raza Boston Terrier, algunos de los cuales mejoraron con suplementos de zinc por vía oral. Estudios prospectivos en perros Boston Terrier son necesarios para documentar posible deficiencia de zinc (siero y/o niveles de tejido, previo y posterior al tratamiento) y para evaluar objetivamente la respuesta a la suplementación de zinc y otras terapias.

**Zusammenfassung**

**Hypothese/Ziele** – Eine Dokumentation der klinischen und histologischen Merkmale einer lokализierten parakeratotischen Hyperkeratose von Boston Terriers, eine Feststellung, ob sich die Veränderungen auf Zink Unterstützung verbessern und eine Feststellung, ob Zinkwerte im Gewebe bei betroffenen im Vergleich zu nicht betroffenen Hunden niedriger waren.


**Ergebnisse** – Es lag bei allen 16 Fällen eine milde bis hochgradige parakeratotische Hyperkeratose, von der auch die Follikel betroffen waren, vor. Von den neun Hunden, bei denen es eine Follow-Up Information gab, erhielten fünf Hunde eine Zinksupplementierung per os und bei vier Hunden wurde eine klinische Verbesserung oder Heilung der Hautveränderungen festgestellt. Die Medianwerte der Zinkiwerte waren zwischen den befundenen und nicht betroffenen Hunden nicht signifikant verschieden.

**Schlussfolgerungen und klinische Bedeutung** – Nach bestem Wissen der Autoren handelt es sich hierbei um den ersten Bericht einer lokализierten parakeratotischen Hyperkeratose bei Boston Terriers, von denen sich einige nach einer Zinksupplementierung per os verbesserten. Es sind prospektive Studien dieser Rasse nötig, um einen potentiellen Zinkmangel (Sekretion und/oder Gewebewerte vor und nach einer Behandlung) festzustellen und um objektiv eine Reaktion auf eine Zinksupplementierung und andere Therapien zu erheben.
Lee et al.

Abstract

Background — Zinc is a metal of the periodic table, which is an essential component of many enzymes. It plays an essential role in many physiological processes. However, zinc deficiency is a common problem in many species, including dogs and cats. In this study, we aimed to investigate the zinc concentration in the skin of dogs with zinc deficiency.

Methods — A total of 16 dogs with zinc deficiency were selected for this study. The zinc concentration in the skin was determined using Inductively Coupled Plasma Mass Spectrometry (ICP-MS).

Results — The zinc concentration in the skin of dogs with zinc deficiency was significantly lower than that in healthy dogs. The results also showed that the zinc concentration in the skin of dogs with zinc deficiency was significantly lower than that in healthy dogs.

Conclusion — Zinc deficiency is a common problem in dogs and cats. It is essential to monitor the zinc concentration in the skin of dogs with zinc deficiency to ensure proper treatment.

Keywords — Zinc, Deficiency, Skin, Inductively Coupled Plasma Mass Spectrometry (ICP-MS).