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What is This?
Immunohistological evaluation of feline herpesvirus-1 infection in feline eosinophilic dermatoses or stomatitis

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This study used immunohistochemistry (IHC) and histopathology to evaluate the presence of feline herpesvirus-1 (FHV-1) in feline cases of ‘eosinophilic granuloma complex’ (EGC) or other eosinophilic dermatoses or stomatitis, diagnosed at the Veterinary Pathology Diagnostic Service, University of Sydney between January 1996 and June 2008. Two of the 30 cases (6.6%) examined showed positive immunoreactivity to FHV-1 using IHC. Intranuclear inclusion bodies were also detected on histopathological examination of haematoxylin and eosin stained sections of both cases but were very difficult to find. Therefore, FHV-1 is uncommonly associated with EGC or other eosinophilic dermatoses or stomatitis in Sydney. However, misdiagnosis as an EGC lesion or other eosinophilic dermatoses may occur if inclusion bodies are overlooked or absent on histopathology and this may significantly decrease the chance of a favourable treatment outcome. FHV-1 should be considered in cats with severe ulcerative cutaneous or oral lesions, unresponsive to corticosteroid treatment, with or without concurrent or historical signs of upper respiratory tract or ocular disease more typical of FHV-1. IHC may be helpful in differentiating FHV-1 dermatitis or stomatitis from other eosinophilic lesions, which is of vital clinical and therapeutic importance.

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Eosinophilic granulomas, also known as feline collagenolytic granulomas or linear granulomas, are classically marked by linear lesions on the caudal or medial thigh, or as nodular lesions anywhere on the body including the footpads, face and oral cavity. They occur more commonly in young cats, and are present as raised, erythematous, alopecic, and occasionally ulcerated lesions.

Eosinophilic plaques are typically pruritic, raised, cutaneous lesions located on the axillary, perineal, inguinal or lateral thigh areas, though they can occur almost anywhere on the body. The pruritus stimulates cats to lick at the lesions, frequently resulting in ulceration of the skin surface. It is more common in young adult cats between 2–6 years old.

Indolent ulcers, also known as eosinophilic ulcers or ‘rodent’ ulcers, are ulcerated lesions most commonly occurring on the upper lip. Lesions can be unilateral or bilateral, of variable size and severity, and are usually neither pruritic nor painful. Large lesions, however, may result in facial distortion.

Overall, there are histological and clinical similarities between EGC and FHV dermatitis and stomatitis, primarily centred on the presence of ulceration, necrosis and eosinophilic infiltrates. Therefore, the objective of this retrospective study was to examine the histopathology slides of previously diagnosed cases of EGC lesions for intranuclear inclusion bodies that may have been overlooked in the initial histopathological examination, and to use immunohistochemistry (IHC) to examine tissue sections for the presence of FHV-1 using anti-FHV-1 antibody. Presence of FHV-1 in these samples may suggest a causative role for FHV-1 in the development of some EGC lesions in cats. In addition, this study describes the development of an immunohistochemical method that may be used in excluding FHV-1 from the differential diagnosis, allowing a more accurate diagnosis of feline eosinophilic dermatoses and thus ultimately influencing treatment and prognosis.

Materials and methods

Case selection

Cases were selected from the database of the Veterinary Pathology Diagnostic Services at the University of Sydney by reviewing the records of biopsy specimens submitted between January 1996 and June 2008, for lesions diagnosed histologically as an EGC lesion, or an eosinophilic dermatitis or stomatitis of unknown cause with histological features of an EGC lesion. Medical records were requested and reviewed for signalment, information regarding clinical presentation, past medical history with particular focus on previous upper respiratory tract disease, treatment and outcome.

Histological examination

The original haematoxylin and eosin (H&E) stained tissue sections of all cases meeting the inclusion criteria were retrieved and re-examined for the presence of eosinophilic intranuclear inclusion bodies in the surface and adnexal epithelial cells. They were examined by all authors, including one pathologist (KB). The biopsy reports by the original pathologists for all cases were reviewed for mention of inclusion bodies.

IHC

Four micron tissue sections were cut from the original archived paraffin blocks. Slides were dried for at least 12 h at 37°C after sections were cut in order to improve tissue adherence to the slide. IHC was performed on all slides within 7 days of being cut to optimise results. Deterpafinisation and rehydration was achieved by submerging the slides for 3 min in two changes of each of the following solutions: 100% xylol, 100% ethanol, 95% ethanol, 70% ethanol and then water. Care was taken not to allow the tissues to dry out once rehydrated.

Monoclonal mouse antibodies to FHV-1 (AbD Serotec, Mouse FHV-1 antibody, MCA 2490) were used for antigen detection using a 1:200 antibody dilution. Fixed tissue samples known to be positive for FHV-1 were not available at the time of our testing, therefore, commercial positive and negative control slides were used. The control slide (VMRD, catalogue number SLD-FAC-FVR, US) consisted of two wells containing fixed, unstained FHV-1-infected Crandell feline kidney cell (CrFK) cultures grown on the surface of teflon-masked slides. Each slide contained one positive and one negative control cell culture well.

Two different antigen retrieval methods were compared: heat-induced antigen retrieval and enzymatic antigen retrieval (proteinase K). In regards to heat-induced antigen retrieval, all slides were completely submerged in a working dilution of commercially available antigen retrieval solution (Target Retrieval Solution, 10× concentrate, code S1699, DakoCytomation, Carpinteria, CA, USA) and microwaved for 10 min as per manufacturer’s instructions. Once cooled the slides were placed in the Dako Autostainer, which was pre-programmed to automatically perform the remaining procedures as outlined below.

Endogenous peroxidases were blocked using 0.03% hydrogen peroxide (Peroxidase Block, code K4007, DakoCytomation, Carpinteria, CA, USA) for 15 min at room temperature. The sections were then incubated for 60 min at room temperature in 1:200 diluted primary FHV-1 antibody. FHV-1 antibody was substituted by diluted (1:100) universal mouse serum (item number 004335, DakoCytomation, Carpinteria, CA, USA) for one slide from each biopsy specimen, in order to provide a negative reagent control to evaluate non-specific and undesirable staining. DakoCytomation Antibody Diluent (code S0809, DakoCytomation, Carpinteria, CA, USA) was used for all dilutions. Envision anti-mouse labelled polymer (labelled polymer-HRP anti-mouse, code K4007, DakoCytomation, Carpinteria, CA, USA) was used as the secondary antibody. All
slides were incubated in the secondary antibody for 30 min at room temperature. Lastly, the slides were incubated for 5 min at room temperature in 3,3’-diaminobenzidine (DAB) chromogen solution (DAB + Chromogen, code K4007, DakoCytomation, Carpinteria, CA, USA). Slides were thoroughly rinsed with DakoCytomation phosphate-buffered saline between each step.

Enzymatic antigen retrieval was achieved by pre-treatment with proteinase K (code S3020, DakoCytomation, Carpinteria, CA, USA) for 10 min at room temperature immediately prior to the application of the primary antibody.

Once the Dako Autostainer finished the DAB step and the slides were rinsed in distilled water, the slides were removed, manually counterstained with haematoxylin, dehydrated and then coverslipped. The positive and negative tissue control slide was examined first under a microscope to ensure the reagents were functioning properly. All slides were thoroughly examined by two separate individuals (including one pathologist – author KB) for the presence of any brown cytoplasmic or intranuclear staining indicating FHV-1-infected cells.

Results

Study population

Thirty cases selected from the database of the Veterinary Pathology Diagnostic Services at the University of Sydney met the inclusion criteria and were used in this study. One additional case diagnosed histologically with eosinophilic dermatitis and eosinophilic folliculitis on the nasal bridge was unable to be included in the study due to the absence of the original paraffin block. The block had been used for polymerase chain reaction, which had detected FHV-1 DNA in the biopsy specimen. However, no inclusion bodies were identified on histopathological examination by two pathologists. Cases selected consisted of a mix of internal cases from the University of Sydney Veterinary Teaching Hospital and external cases from various nearby veterinary clinics in the Sydney region.

The age of the cat at submission of the biopsy samples was known for 27/30 cats and ranged from 5 months to 17 years (average 7.1 years; median 7 years, interquartile range 3–10) (Table 1). The majority of cats (20, 67%) were domestic shorthair (DSH) cats. Other breeds included an Exotic Shorthair, Manx, Himalayan, Maine Coon, Abyssinian, Persian cross, Burmese, Persian and Tonkinese. There were 13 females and 16 males. Of the 30 cats, 14 had oral lesions, four had facial lesions, 11 had lesions on the feet, body or limbs, and one had both an oral lesion and abdominal skin lesion submitted for histological examination.

Histological examination

The original H&E stained tissue sections were reviewed for amorphous eosinophilic intranuclear inclusion bodies within surface and adnexal epithelial cells for all 30 cases. Inclusion bodies were only detected in two cases, which were also the only cases that had convincing positive reactivity with IHC for FHV-1 antigen. The original pathologist report had not noted the presence of any inclusion bodies. Histologically, these lesions had severe epidermal ulceration and necrosis, with the necrosis extending into the follicular epithelium and underlying superficial dermis. Intranuclear inclusion bodies were found in epithelial cells of hair follicles near or within areas of necrosis, and were associated with margination of chromatin on the nuclear membrane and a peripheral clear halo (Fig 1). In the deeper dermis there was an infiltration of macrophages and eosinophils. All slides were scanned thoroughly and no inclusion bodies were detected in the remaining 28 cases.

IHC

Convincing positive immunohistochemical staining for FHV-1 antigen was present in only two cats (cats 17 and 18 in Table 1). Staining was present on the two slides incubated with the FHV-1 antibody, but absent on the slide incubated with the negative reagent control, indicating non-specific background staining to be unlikely. Both heat-induced antigen retrieval and enzymatic antigen retrieval resulted in positive staining. However, utilisation of proteinase K resulted in significantly darker and stronger staining (Fig 2). Positive immunoreactivity was present in surface and adnexal epithelial cells adjacent to or within areas of necrosis (Figs 2 and 3). Both intranuclear and cytoplasmic staining were present. In addition, fine granular staining not associated with specific cells was present throughout areas of necrosis. This staining is likely karyorrhectic debris as a result of viral cytolysis of epithelial cells.

The commercial control slide was not affected by heat antigen retrieval. Proteinase K however, caused the fixed cell cultures on the commercial control slide to detach from the slide, even when the proteinase K treatment period was reduced to 5 min. Proteinase K had no such effect on the tissue samples used in this study.

Sections from all other cats revealed either no staining or irrelevant background staining with IHC, with the exception of one case (cat 2). This case displayed unusual cytoplasmic staining of random cells scattered within the dermis on sections where heat-induced antigen retrieval was used, but was not present on corresponding sections using proteinase K antigen retrieval. These cells appeared to be plasma cells, but cell identification was difficult due to the cytoplasmic staining. Staining of these cells was present on the two slides incubated with the FHV-1 antibody, but absent on the slide incubated with the negative reagent control. IHC was repeated for this case using heat-induced antigen retrieval and similar staining was again achieved. The biopsy specimen was of a sublingual mass, originally diagnosed as an oral eosinophilic granuloma histologically by a pathologist.
### Table 1. Summary of the histopathology and immunochemistry results for all 30 cats.

<table>
<thead>
<tr>
<th>Cat</th>
<th>Age at diagnosis (years)</th>
<th>Breed</th>
<th>Sex</th>
<th>Region of biopsy sample</th>
<th>Histological diagnosis by pathologist</th>
<th>Inclusion bodies</th>
<th>IHC- heat</th>
<th>IHC- PK</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>16</td>
<td>DSH</td>
<td>MN</td>
<td>Oral lesion</td>
<td>Ulcers caudal hard palate, base of tongue, glossopalatine arch</td>
<td>ECG</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>2</td>
<td>9.5</td>
<td>DSH</td>
<td>M</td>
<td>Sublingual mass</td>
<td>Oral eosinophilic granuloma</td>
<td>—</td>
<td>Unusual cytoplasmic staining in dermis</td>
<td>—</td>
</tr>
<tr>
<td>3</td>
<td>0.4</td>
<td>Exotic shorthair</td>
<td>NA</td>
<td>Pharynx-tonsil</td>
<td>EGC</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>4</td>
<td>10</td>
<td>DSH</td>
<td>F</td>
<td>Mass base of the tongue</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>5</td>
<td>12.5</td>
<td>DSH</td>
<td>MN</td>
<td>Oral mucosa</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>6</td>
<td>NA</td>
<td>DSH</td>
<td>MN</td>
<td>Oral mucosa</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>7</td>
<td>9</td>
<td>DSH</td>
<td>MN</td>
<td>Tongue mass</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>8</td>
<td>12</td>
<td>DSH</td>
<td>FN</td>
<td>Tongue</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>9</td>
<td>5</td>
<td>Manx</td>
<td>FN</td>
<td>Tongue mass</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>10</td>
<td>7</td>
<td>DSH</td>
<td>MN</td>
<td>Oral mucosa</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>11</td>
<td>10</td>
<td>DSH</td>
<td>FN</td>
<td>Tongue</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>12</td>
<td>9</td>
<td>DSH</td>
<td>MN</td>
<td>Tongue</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>13</td>
<td>11</td>
<td>DSH</td>
<td>M</td>
<td>Oropharynx</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>14</td>
<td>NA</td>
<td>Himalayan</td>
<td>M</td>
<td>Tonsils and soft palate</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>15</td>
<td>0.6</td>
<td>Maine Coon</td>
<td>FN</td>
<td>Face</td>
<td>Upper lip</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>16</td>
<td>3</td>
<td>DSH</td>
<td>M</td>
<td>Nasal plane</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>17</td>
<td>17</td>
<td>Abyssinian</td>
<td>FN</td>
<td>Left lower eyelid</td>
<td>Eosinophilic inflammatory lesion</td>
<td>+</td>
<td>—</td>
<td>++</td>
</tr>
<tr>
<td>18</td>
<td>7</td>
<td>DSH</td>
<td>MN</td>
<td>Nasal bridge and nasal mucosa</td>
<td>Eosinophilic granuloma (nasal skin), eosinophilic rhinitis (nasal mucosa)</td>
<td>+</td>
<td>—</td>
<td>++</td>
</tr>
<tr>
<td>19</td>
<td>7</td>
<td>Persian Cross</td>
<td>FN</td>
<td>Feet/body/limbs</td>
<td>Digital pad of 3rd phalanx</td>
<td>EGC and furunculosis</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>20</td>
<td>3</td>
<td>Burmese</td>
<td>FN</td>
<td>Skin elbow</td>
<td>Eosinophilic plaque/miliary dermatitis</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>21</td>
<td>15</td>
<td>DSH</td>
<td>MN</td>
<td>Skin LF foot</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>22</td>
<td>12</td>
<td>DSH</td>
<td>F</td>
<td>Skin axilla, chest, leg</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>23</td>
<td>1</td>
<td>Persian</td>
<td>MN</td>
<td>Food pad</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>24</td>
<td>3</td>
<td>DSH</td>
<td>F</td>
<td>Caudal thigh and ventral abdomen</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>25</td>
<td>6</td>
<td>DSH</td>
<td>FN</td>
<td>Skin caudal abdomen</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>26</td>
<td>9</td>
<td>NA</td>
<td>M</td>
<td>Skin inner thigh</td>
<td>Eosinophilic granuloma</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
</tbody>
</table>

(continued on next page)
Clinical details of the two cats positive for FHV on IHC

Cat 17 (17-year-old female neutered Abyssinian) had positive immunohistochemical staining on the biopsy of a left lower eyelid mass. Relevant medical history included an extensive erosive nasal squamous cell carcinoma with concurrent EGC, treated initially with radiation therapy using gold implants, and then intralesional carboplatin when it recurred a year later. The squamous cell carcinoma had been in remission and stable over the past 3 years with a residual erosive lesion on the left dorsal nasal plane. Presence of upper respiratory tract signs consistent with FHV-1 in the past was not reported. The lower eyelid mass involved more than 75% of the eyelid margin and had developed rapidly over 2 weeks. The lesion was diagnosed histopathologically as an eosinophilic inflammatory lesion with extensive necrosis, possibly consistent with an eosinophilic granuloma. Tongue ulcers were discovered at the time of the eyelid biopsy. Treatment was started with prednisolone, amoxicillin-clavulanate and chlorambucil, however, the cat declined rapidly and was euthanased a month after the eyelid biopsy. By this time, lesions had surrounded both eyes, 75% of the tongue, and the nose was extensively ulcerated.

Cat 18 (7-year-old male neutered DSH) had positive immunohistochemical staining on a nasal bridge and nasal mucosa biopsy. The cat was initially presented with a raised irregular lesion on the nasal bridge, purulent ocular discharge and snuffling. The cat had a chronic history of upper respiratory signs that commenced as a kitten. The nasal lesion was considered to be a reoccurrence of an EGC lesion in the same location diagnosed by histopathology 1-year prior. The prior lesion did not respond to prednisolone but appeared to completely resolve on megestrol.
acetate, thus this medication was trialled again but with no success. In addition, there was no response to prednisolone, methylprednisolone, amoxicillin-clavulanate or cephalexin. The nasal lesion progressed to severe ulceration of the nasal planum and nasal bridge. Tongue ulcers were later noticed accompanied by increasing difficulty in breathing through the nose. About 6 weeks after the initial signs, biopsies of the nasal cavity and nasal bridge lesions were taken confirming a diagnosis of eosinophilic granuloma and eosinophilic rhinitis. Due to the lack of response to treatment and the severity of disease the cat was eventually euthanased. A post mortem examination was performed and samples of the nasal skin, tongue, nasal cavity, lung and mandibular lymph node were submitted for histopathology. The histopathology report concluded the lesions to be a particularly severe case of EGC. Periodic acid-Schiff staining of samples of the nasal cavity did not reveal any fungi and a *Cryptococcus* species antigen test from serum was negative.

**Discussion**

In this study FHV-1 was detected using IHC and histopathology in EGC biopsy specimens from 2/30 cats (6.6%). This suggests that FHV-1 dermatitis or stomatitis is an uncommon cause of eosinophilic dermatitides in Sydney. However, these case studies illustrate the importance of excluding FHV-1 as an aetiological agent in order to optimise the likelihood of a favourable treatment outcome.

Previous reports on the prevalence of FHV-1 dermatitis and stomatitis are lacking. Hargis et al. reported nine feline cases of ulcerative facial or nasal dermatitis and one feline case of focal proliferative ulcerative stomatitis, located on the soft palate, associated with FHV-1. These cases were documented in the USA between 1996 and 1997, and suggest a higher prevalence than expected from the paucity of cases noted in previous literature. In six of these cats, intranuclear inclusion bodies were initially missed in histopathology samples and the eosinophilic inflammation was originally associated with EGC or allergic dermatitis. A small number of cases in domestic cats have since been reported. FHV-1 has also been reported to cause similar erythematous, ulcerated cutaneous lesions in cheetahs, primarily on the face and forelimbs. As with domestic cats, these lesions in cheetahs are characterised by infiltrations of eosinophils and plasma cells, epithelial hyperplasia, necrosis and intraepithelial inclusion bodies.

One cat with positive immunohistochemical staining for FHV-1 in the current study had concurrent and chronic history of upper respiratory tract disease consistent with FHV-1. Most, but not all, cases of FHV dermatitis or stomatitis reported in the literature have had concurrent or previous signs of upper respiratory tract disease. Subsequently it may be suggested...
that a history of concurrent or past upper respiratory or ocular signs may be a predisposing or contributing factor to FHV-1 dermatitis or stomatitis. FHV-1 establishes latency primarily in the trigeminal ganglia and FHV-1 dermatitis or stomatitis may be a form of recrudescence triggered by stress or immunosuppression.

Epithelial intranuclear inclusion bodies were found in the cats with positive immunostaining on IHC in the current study. However, only low numbers were found and could have been easily overlooked if the pathologist was not specifically looking for inclusion bodies. In addition, intranuclear inclusion bodies are most commonly observed histopathologically during the initial period of active viral replication and are rarely detected beyond this stage when there are often vast areas of necrosis present. Thus the absence of inclusion bodies does not exclude a diagnosis of FHV-1 dermatitis or stomatitis and in the absence of inclusion bodies, if histological and clinical findings are consistent with FHV-1 dermatitis or stomatitis, further diagnostic investigation utilising IHC may be rewarding.

The reason for the absence of positive immunoreactivity in the other cases may have one of several explanations: FHV-1 may not be involved in the pathogenesis of disease; FHV-1 may not be present in the tissue samples tested, or the FHV-1 present may be below the level of detection by IHC (ie, false negatives). The latter suggestion seems less likely due to the strong staining present on the control slides as well as the sections of the positive cases. For future research other methods for detection of FHV-1 in biopsy specimens could include in situ hybridisation, polymerase chain reaction (PCR) and transmission electron microscopy. As with IHC, in situ hybridisation demonstrates the distribution of FHV-1 in tissue biopsies, however, instead of detecting FHV-1 antigen it detects FHV-1 DNA and therefore does not rely on the presence of whole virus. Amplification of FHV-1 nucleic acid by PCR would allow for detection of trace amount of viral DNA. However, it does not differentiate between viable DNA and avirulent virus or viral DNA fragments that are not contributing to disease, as demonstrated in a study where FHV-1 was detected in 31% of apparently normal cats using PCR. In addition, these techniques may have limited availability in veterinary diagnostic laboratories compared with IHC. Hence, from a clinical perspective, IHC was chosen as the method of choice for the detection of FHV-1 in this study with the view that it could be applied as a diagnostic test for future cases.

The method of IHC used in this study was previously described in detail in materials and methods and differs from methods used by previous researchers. With regard to the method of antigen retrieval, the results of this study suggest proteinase K to be more suitable to retrieve FHV-1 antigen. Proteinase K produced stronger staining in positive sections in comparison to heat-induced antigen retrieval. In addition, heat-induced antigen retrieval resulted in unusual cytoplasmic staining of random cells scattered within the dermis of one biopsy specimen, which was not present on corresponding sections using proteinase K antigen retrieval. As FHV-1 is an epitheliotropic DNA virus, and is not known to infect cells in the dermis, we concluded that the staining was most likely a result of cross-reactivity by the antibody and less likely a reflection of FHV-1-infected cells. Lack of staining when using proteinase K antigen retrieval may be explained by proteinase K being unable to ‘unmask’ the particular component causing the cross-reactivity. These theories could be verified using in situ hybridisation methods.

Despite the uncommon occurrence of FHV-1 dermatitis or stomatitis suggested by the present study, ruling out FHV-1 as a differential is an important consideration as these diseases have very different treatment options. This is reflected in the cats with FHV-1 dermatitis identified in this study, which did not respond to various treatments for EG lesions and were subsequently euthanased. If IHC had been available for these cats, the use of antiviral agents such as famciclovir may have avoided the need for euthanasia.

Treatment of EG lesions are usually based on systemic corticosteroid therapy and identification and avoidance of potential allergens, including strict flea control. Other treatments that have been used include antibiotics, cyclosporine, chlorambucil, antigens, oral gold, megestrol acetate, surgical excision, cryotherapy and laser excision or ablation. While response supports an underlying hypersensitivity, many cases are non-responsive, possibly indicating further unidentified factors involved in the aetiopathogenesis of EG lesions or inaccurate diagnosis.

Reports on the treatment and prognosis of FHV-1 dermatitis or stomatitis are limited. Treatment options which have been reported to be beneficial include oral interferon, surgical excision and antibiotics to treat secondary bacterial infection. Interferons are a group of related cytokines with multiple antiviral, antiproliferative and immunomodulatory properties, and have been used to treat various viral infections. In addition lysine, and numerous antiviral agents (such as acyclovir) developed for the treatment of human herpesvirus have been used to treat cats with respiratory and ocular signs of FHV-1 with varying success. More recently, the use of famciclovir, a systemic antiviral agent has shown favourable clinical responses in the management of diseases attributable to FHV-1, including conjunctivitis, keratitis, corneal sequestra, rhinosinusitis and FHV-1-associated dermatitis. Controlled studies using these various agents in cats are limited. Unlike EG lesions, corticosteroids have not been shown to be beneficial and may exacerbate FHV-1 lesions.

In summary, differentiation of FHV-1 dermatitis or stomatitis from EG lesions or other eosinophilic
dermatitides is of vital clinical and therapeutic importance. Increased suspicion of FHV-1 dermatitis or stomatitis should occur in cats with severe ulcerative cutaneous or oral lesions unresponsive to corticosteroid treatment, with or without concurrent or historical signs of upper respiratory tract or ocular disease more typical of FHV-1. Furthermore, the present study has outlined a relatively fast and easy to interpret protocol for the detection of FHV-1 using IHC, which is particularly useful if inclusion bodies are not detected on histopathology.

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