Brief Communication

J Vet Intern Med 2001;15:162–166

Idiopathic Hypereosinophilic Syndrome in 3 Rottweilers

Jane E. Sykes, Douglas J. Weiss, Lance C. Buoen, Melissa M. Blauvelt, and David W. Hayden

Three Rottweilers with marked peripheral eosinophilia and infiltration of the liver, spleen, lungs, and bone marrow with eosinophils were diagnosed with idiopathic hypereosinophilic syndrome (IHES). Mean serum immunoglobulin E concentrations were markedly high. On cytogenetic analysis, no evidence of karyotypic abnormalities was found in bone marrow aspirates. Despite an extensive search, no underlying cause for the eosinophilia could be identified. In this study, cytogenetic analysis and measurement of serum IgE concentrations were used to differentiate IHES and eosinophilic leukemia.

Key words: Cytogenetic analysis; Dogs; Eosinophilic gastroenteritis; Eosinophilic leukemia; Immunoglobulin E; Splendore-Hoeppli phenomenon.

Peripheral eosinophilia and multiple organ infiltration with eosinophils, without evidence of underlying parasitic, neoplastic, vascular, or allergic disease, have been described frequently in humans and cats, but reports in dogs are rare. In humans, idiopathic hypereosinophilic syndrome (IHES) is defined by sustained (>6 months) peripheral eosinophilia of >1,500 cells/μL with no apparent etiology and multiple organ involvement.[1] The difference between IHES and eosinophilic leukemia (EL) is controversial, and although molecular techniques may help distinguish these 2 disorders in humans, in some cases differentiation may not be possible.

This report describes 3 cases of canine IHES with widely differing clinical presentations. All dogs were Rottweilers, and each had large numbers of eosinophils in blood, liver, spleen, and bone marrow aspirates. Pulmonary involvement was present in 2 dogs and was suspected in the other. Karyotype analysis and measurement of serum immunoglobulin E (IgE) concentrations supported the diagnosis.

Case Reports

Dog 1

A 3-year-old female Rottweiler was referred to the Veterinary Teaching Hospital (VTH; University of Minnesota, St Paul, MN) with a 1-month history of vomiting, severe bloody diarrhea, and weight loss. Physical examination disclosed lethargy, cachexia, mucosal pallor, and abdominal enlargement; moderate ascites was detected by abdominal palpation.

A CBC disclosed moderate neutrophilia with a left shift, and marked mature eosinophilia (Table 1). A nonregenerative anemia was present (mean corpuscular hemoglobin concentration [MCHC], 32.1 g/dL, reference range [RR], 33.6–36.6 g/dL; hematocrit, 15.7%, RR 38.5–56.7%). The results of the platelet count were normal. Serum chemistry test results indicated the presence of hypoalbuminemia (1.8 g/dL, RR 2.4–3.8 g/dL), mild hyponatremia (142 mEq/L, RR 143–150 mEq/L), and high amylase activity (1,392 U/L, RR 209–973 U/L). Serum protein electrophoresis (SPE) disclosed decreased albumin (1.3 g/dL, RR 1.7–2.9 g/dL) and α-2-macroglobulin (0.38 g/dL, RR 0.41–1.11 g/dL) fractions, but γ-globulin concentration was within the RR. Mean serum IgE concentration was 658 μg/mL (RR < 50 μg/mL). Peritoneal fluid was a serosanguineous, inflammatory exudate, with a total protein concentration of 3.4 g/dL, 31,000 red blood cells (RBC)/μL, and 10,000 mature eosinophils/μL. No other nucleated cells were present. Fecal flotation and heartworm antigen test results were negative, and results of the urinalysis were unremarkable.

Abdominal ultrasound showed diffuse variation in hepatic and splenic echotexture, and an irregular splenic capsular outline. Splenic aspirates yielded large numbers of eosinophils, some of which were immature. Fewer numbers of plasma cells, small lymphocytes, neutrophils, macrophages, and hemic precursor cells were seen. A liver aspirate contained large numbers of mature eosinophils and occasional macrophages. Pre- and postprandial bile acid concentrations were mildly increased (17 μmol/L and 18 μmol/L, respectively; RR < 5 and < 15 μmol/L, respectively).

A bone marrow aspirate and core biopsy disclosed marrow hyperplasia with a normal myeloid : erythroid (M : E) ratio. Approximately 40% of myeloid cells were eosinophils in various stages of differentiation. Maturation was orderly in both the myeloid and erythroid series. The dog was euthanized because of a poor prognosis. Immediately after death, bone marrow was aspirated for karyotype analysis. Examination of at least 20 chromosome spreads was not possible because of inadequate growth of bone marrow cells. However, the few chromosome spreads evaluated had diploid female karyotype with no translocations, additions, or deletions.

At postmortem examination, ascites (3,350 mL) and amber pleural effusion (685 mL) were present. The stomach was thickened (0.5–2.5 cm) with scattered serosal hemorrhages. Several deep gastric ulcers were present, the largest measuring 3.5 × 1 cm (Fig 1). Undigested and digested blood was present in the small intestine, cecum, and colon. In the colon, 4 mucosal plaques, 4 cm in diameter and 1 cm in thickness, were found proximal to the pelvic inlet (Fig 2). Peripancreatic, portal, and colonic lymph nodes were enlarged. The left medial lobe of the liver was nearly replaced by an infiltrative tan to yellow mass (Fig 3). The spleen was enlarged and irregular.

Histologically, the lungs contained interstitial infiltrates of eosinophils, lymphocytes, and plasma cells with some fibroplasia. The pleural fluid had a total protein concentration of 3.0 g/dL, 24,300 RBC/μL, and 6,900 white blood cells/μL (90% mature eosinophils, 10% macrophages and neutrophils).

The stomach wall contained dense fibrous tissue deposits that markedly thickened the submucosa and serosa and sometimes replaced smooth muscle. Eosinophil infiltrates were concentrated in the basal

From the Department of Small Animal Clinical Sciences (Sykes), the Department of Veterinary Pathobiology (Weiss, Blauvelt), and the Department of Veterinary Diagnostic Medicine (Buoen, Hayden), University of Minnesota, St Paul, MN. *An abstract of this work was presented at the 18th Annual American College of Veterinary Internal Medicine Conference, Seattle, WA, 2000.*

Reprint requests: Jane E. Sykes, BVSc (Hons), PhD, Department of Small Animal Clinical Sciences, University of Minnesota, 1352 Boyd Avenue, St Paul, MN 55108; e-mail: sykes005@tc.umn.edu.

Submitted June 27, 2000; Revised August 9, 2000; Accepted September 22, 2000.

0891-6640/01/1502-0013/$3.00/0

Table 1. Differential white blood cell count in 3 dogs with hypereosinophilic syndrome.[a]

	Dog 1	Dog 2	Dog 3	Reference Range
Total white blood cells	33,900	47,600	23,700	4,100–13,300
Eosinophils	8,480	34,748	7,820	0–1,200
Segmented neutrophils	22,030	5,236	7,580	2,100–11,200
Band neutrophils	340	0	0	0–130
Lymphocytes	2,030	4,284	7,350	300–5,100
Monocytes	1,020	1,428	950	0–1,200
Basophils	0	1,904	0	0–130

[a] All values are in absolute numbers (cells/μL).

lamina propria, submucosa, and tunica muscularis. The lamina propria was edematous and contained lymphocyte and plasma cell infiltrates. Small granulomas composed of macrophages, multinucleated giant cells, and eosinophils were scattered throughout the submucosa. Numerous granulomas contained deposits of globular fibrinoid material typical of the Splendore-Hoeppli (SH) phenomenon (Fig 4). The deposits stained with periodic acid-Schiff stain and were negative for microbial agents with Gram, Gomori's methanamine silver, and Kinyoun's acid fast stains. Serosal lymphatics were dilated, and some contained eosinophils.

In the small intestine, lymphocytes, plasma cells, and eosinophils were variably present in the villous cores and lamina propria. The colonic mucosa had increased connective tissue and infiltrates of lymphocytes, plasma cells, and eosinophils. The raised colonic lesions resulted from marked submucosal expansion by infiltrates of eosinophils, edema, hemorrhage, and fibroplasia. Large numbers of eosinophils, reactive fibroblasts, and mononuclear cells infiltrated the tunica muscularis. The serosa was edematous and infiltrated with eosinophils and plasma cells. Thrombi obstructed some of the lymphatics.

The architecture of the left medial liver lobe was disrupted and largely replaced by massive infiltrations of eosinophils. Hepatic necrosis, hemorrhage, and interstitial fibrosis were accompanied by diffuse and marked accumulations of mature eosinophils with fewer numbers of macrophages, plasma cells, and lymphocytes (Fig 5). Eosinophilic granulomas with the SH phenomenon were noted in some areas.

The splenic red pulp contained eosinophils and plasma cells intermixed with neutrophils, lymphocytes, and megakaryocytes. Paracortical tissues and medullary trabeculae of the peripancreatic, portal, and mediastinal lymph nodes were infiltrated with eosinophils.

Fig 1. Stomach; dog 1. The gastric mucosa is swollen, and puffy folds contain punctate, linear, and crateriform ulcers (arrows).

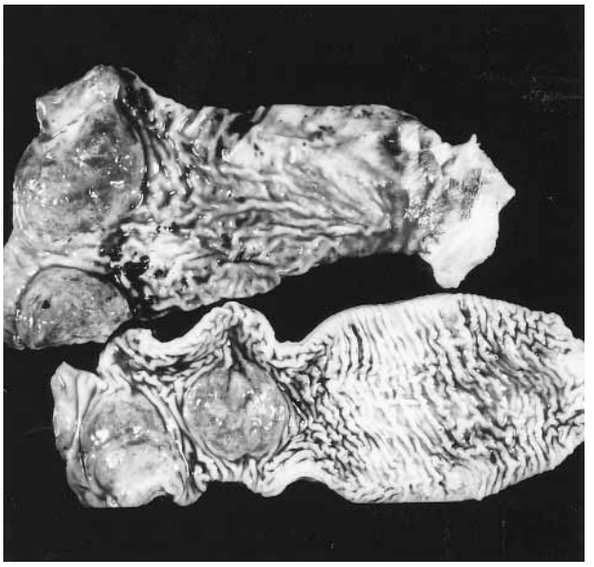

Fig 2. Colon; dog 1. Mucosa of the descending colon has multiple smooth, raised mounds of tissue and dark deposits of digested blood.

Dog 2

A 10-month-old spayed female Rottweiler was referred to the VTH for a 1-week history of inappetance. Large numbers of eosinophils previously had been seen in a blood smear, and when the dog had been spayed 6 weeks before referral, enlarged mesenteric lymph nodes, free abdominal fluid, and fibrin tags were noted. Physical examination was unremarkable except for slightly increased bilateral bronchovesicular sounds on thoracic auscultation, and splenomegaly on abdominal palpation. Rectal examination was unremarkable.

A CBC disclosed monocytosis, basophilia, and marked mature eosinophilia (Table 1). The hematocrit was normal (42.8%) but erythrocytes were hypochromic (MCHC = 30.4 g/dL). Results of serum chemistry disclosed hyperglobulinemia (4.6 g/dL, RR 2.7–4.4 g/dL) and mild hyperphosphatemia (6.9 mg/dL, RR 2.6–6.8 mg/dL). On

Fig 3. Liver; dog 1. Profile of the left medial lobe is distorted by an infiltrate of light-colored tissue.

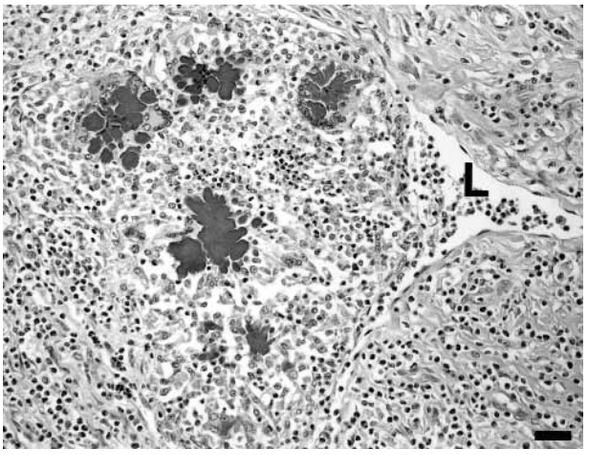

Fig 4. Stomach; dog 1. Submucosal lymphatics (L) are partly obliterated by an eosinophilic granuloma that contains globules and spicules of a homogeneous precipitate (Splendore-Hoeppli phenomenon) and multinucleated giant cells. Hematoxylin and eosin stain. Bar = 40 μm.

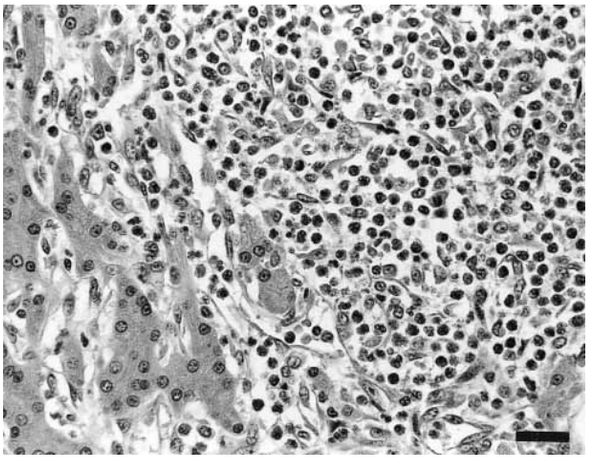

Fig 5. Liver; dog 1. Cords of hepatocytes are disrupted and replaced by an intense infiltrate of mature eosinophils. Hematoxylin and eosin stain. Bar = 30 μm.

SPE, decreased β_1-globulin (0.53 g/dL, RR 0.55–1.20 g/dL) and increased γ-globulin (1.59 g/dL, RR 0.38–1.28 g/dL) concentrations were identified. Mean serum IgE concentration was 500 μg/mL. Fecal flotation and heartworm antigen test results were negative.

On thoracic radiography, mild, diffuse bronchointerstitial pulmonary infiltrates with prominent bronchial markings were seen. Abdominal ultrasound disclosed moderate to severe splenomegaly. The liver was coarsely echogenic, and a small amount of anechoic fluid was present in the abdomen. Mesenteric and sublumbar lymph nodes were enlarged. Splenic aspirates contained large numbers of mature eosinophils (40% of nucleated cells). The remaining nucleated cells consisted of lymphoid cells (40%) and segmented neutrophils (20%). Large clusters of ''benign-appearing'' fibroblasts were seen, suggestive of fibrosis. Liver aspirates contained clusters of normal hepatocytes surrounded by mature eosinophils, with fewer numbers of neutrophils, small lymphocytes, and monocytes. Mesenteric lymph node aspirates contained 50% lymphoid cells and 50% mature eosinophils. A bone marrow aspirate and core biopsy disclosed bone marrow hyperplasia with an increased M : E ratio (5 : 1). Maturation of all cell lines was orderly and appropriate. A marked increase in immature myeloid precursors was observed, and approximately 50% of the cells had features of eosinophil differentiation. Karyotype analysis was normal.

Treatment with prednisone was begun (1 mg/kg PO q12h). The dog's appetite and activity improved markedly. No eosinophils were observed on a CBC performed 21 days later. The CBC was monitored, and the dosage of prednisone was reduced (1 mg/kg q24h on day 21, and then q48h on day 59). Clinical remission was maintained at this dosage. On day 144, the owner reported the dog was doing well, and prednisone therapy had been discontinued for the last 2 weeks. A CBC disclosed leukocytosis, consisting of eosinophilia and basophilia (Table 2). Alternate-day prednisone therapy was reinstituted, and at the time of writing (day 285), the dog was well and the CBC was within normal limits.

Dog 3

An 18-month-old male Rottweiler was referred to the VTH with a 2-month history of progressive inappetance, lethargy, and a frequent, productive cough. The sputum consisted of blood-tinged mucus. The referring veterinarian had performed a *Blastomyces* titer, results of which were negative. Culture of the sputum yielded *Klebsiella pneumoniae*, but only partial improvement resulted after treatment with clavulanic acid-amoxicillin. Eosinophilia and lymphocytosis were present on a CBC (Table 1). The only abnormalities on physical examination were thin body condition, and harsh lung sounds bilaterally.

Hyperamylasemia (1,478 U/L) was present on serum chemistry, and the urinalysis was unremarkable. Increased α_1-macroglobulin concentration (1.04 g/dL, RR 0.37–0.96 g/dL) was identified on SPE. Mean serum IgE concentration was 642 μg/mL. Fecal flotation and heartworm antigen test were negative. On thoracic radiography, severe, multifocal alveolar disease and lung consolidation were observed, primarily in the caudoventral lung fields (Fig 6). A patchy interstitial infiltrate and mild bronchial pattern were noted in the remaining lung fields. Abdominal ultrasound was unremarkable, but cytology of ultrasound-guided fine-needle aspirates of liver and spleen yielded an abundance of eosinophils. On bone marrow aspiration, bone marrow hyperplasia with an M : E ratio of 3 : 1 was present. An increase in immature myeloid precursors, 30% of which had evidence of eosinophil differentiation, was identified. Results of karyotype analysis were normal.

Treatment with prednisone (1 mg/kg PO q12h) was begun, and the dog's cough lessened and its activity improved. Three weeks after presentation, the dog was found dead. It had vomited twice in the previous 24 hours, but manifested no other abnormalities. Permission for postmortem examination was not granted.

Discussion

Eosinophils have the capacity to cause tremendous tissue destruction. Eosinophil cationic granule proteins are cytotoxic, trigger mast cell degranulation, and incite thrombo-

Table 2. Differential white blood cell count in dog 2 after treatment with systemically administered glucocorticoids.[a]

	Day 21	Day 59	Day 82	Day 144	Day 285
Total white blood cells	16,300	8,700	15,300	22,800	6,800
Eosinophils	0	520	310	9,350	816
Segmented neutrophils	13,350	5,390	12,090	7,980	5,032
Band neutrophils	0	0	0	0	0
Lymphocytes	1,790	2,090	1,070	4,790	612
Monocytes	980	700	1,840	0	340
Basophils	0	0	0	680	0

[a] All values are in absolute numbers (cells/μL).

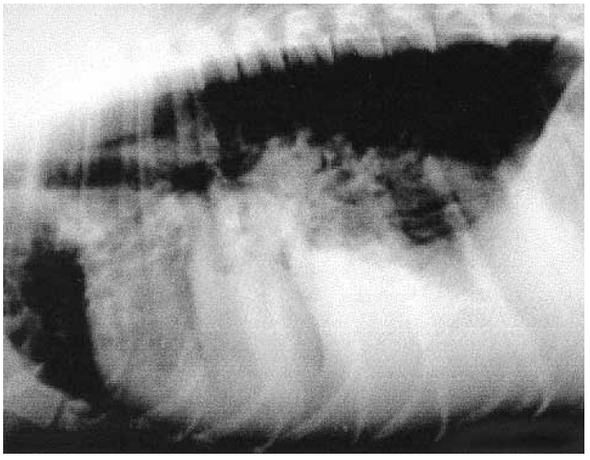

Fig 6. Lateral thoracic radiograph; dog 3. Severe, multifocal alveolar pattern in the cranioventral lung fields and consolidation of the caudoventral lung fields.

sis.[2] Eosinophils produce large amounts of 4-series leukotrienes, which cause increased vascular permeability, mucus secretion, and smooth muscle contraction.[2] Further damage is caused by reactive oxygen products generated by eosinophil peroxidase and the respiratory burst-oxidase pathway. Enhanced fibrosis may result from stimulation of fibroblast proliferation and inhibition of proteoglycan degradation by eosinophil products.[1]

Eosinophils can accumulate in response to parasitic infections, allergic diseases, vasculitides, and neoplasia. In humans, IHES is defined by sustained circulating eosinophilia of >1,500 cells/μL, with no apparent etiology and evidence of multiple organ involvement.[2] Similar definitions have been applied to feline IHES.[3] Human IHES may involve the skin, lungs, gastrointestinal (GI) tract, and, less commonly, the liver.[1] Most humans develop cardiac, ocular, and neurologic disease secondary to the release of eosinophil products. No evidence of these complications was present in the dogs described here. In cats, clinical signs usually reflect GI involvement. Disseminated eosinophilic granulomatous disease also has been described in horses.[4] Finally, IHES with GI involvement and histopathologic changes that resembled those described in dog 1 of this report has been reported in ferrets.[5]

In dogs, eosinophilia has been observed to accompany allergic and parasitic disease, as well as a variety of neoplasms. In pulmonary infiltration with eosinophils, eosinophilic inflammation is restricted to the lungs, and may be associated with dirofilariasis or a hypersensitivity reaction to inhaled antigens.[6] Canine eosinophilic gastroenteritis also may result from a hypersensitivity reaction. Rottweilers may be predisposed, and affected dogs often are less than 5 years of age.[7] The raised colonic lesions found in dog 1 resembled a ''colonic pseudopolyp'' similar to that described in a dog with eosinophilic colitis.[8] Some patients with eosinophilic gastroenteritis may have undetected hepatic, splenic, or bone marrow involvement. Idiopathic eosinophilic meningoencephalitis also has been reported in 3 Rottweilers.[9]

Few cases of canine IHES have been reported. Some cases have involved parasitized dogs or dogs originating from heartworm-endemic areas.[10,11] Two male Rottweilers with IHES have been described previously.[12,13] One of these Rottweilers was thought to have chronic myelogenous EL based on massive organ infiltration with eosinophils of varying degrees of maturity.[13] The respiratory and GI tracts, spleen, and bone marrow most commonly have been involved in the dogs reported. Involvement of the liver, spleen, and bone marrow was found in all of the dogs reported here. Pulmonary involvement was confirmed in dogs 1 and 3, and the interstitial lung pattern seen in dog 2 also may have represented eosinophil infiltration.

The relatively high frequency of eosinophilic syndromes in Rottweilers suggests that a breed predisposition for hypereosinophilic disorders may exist in Rottweilers. This suggestion also is supported by a recent study of 125 dogs with eosinophilia, 34% of which were Rottweilers.[14] Unfortunately, the familial relationship of the 3 Rottweilers reported here could not be determined.

The pathogenesis of the SH phenomenon is controversial. Tissue infiltration by eosinophils and the SH phenomenon have been reported in young German Shepherd Dogs in association with migrating *Toxocara canis* larvae.[15] Bacteria, fungi, and inanimate objects also have been associated with the phenomenon, which predominantly comprises antigen-antibody precipitates, eosinophil granule contents, and cell debris. In the 1st case reported here, serial sections of the granulomatous lesions failed to identify an etiologic agent.

Increased concentrations of serum immunoglobulins and lymphokines, and a therapeutic response to corticosteroids suggest a diagnosis of IHES in humans.[16] Not all patients exhibit all of these features, and several mechanisms probably account for the eosinophilia.[1] In humans, eosinophilia often is accompanied by overproduction of granulocyte-macrophage colony stimulating factor and interleukin (Il)-3, Il-4, and Il-5, with secondary induction of IgE synthesis. This overproduction may arise from premalignant or malignant T-cell clones, or activated T helper type 2 (Th2) cells associated with allergic, parasitic, vascular, or neoplastic disease. These cytokines stimulate eosinophil differentiation, proliferation, chemoattraction, and survival. The high serum IgE concentrations in the patients described in this report may indicate a dysregulated immediate-type hypersensitivity reaction to an unidentified antigen, or abnormal stimulation of IgE production due to excessive lymphokine secretion by Th2 cells, because an underlying antigenic stimulus was not found.

The eosinophilia in these 3 dogs may have resulted from EL. Although a disproportionate number of immature cells has been used to identify acute EL in humans, a lack of immature cells does not rule out chronic EL.[1] Similarly, although the presence of clonal karyotype abnormalities is diagnostic of underlying neoplasia, their absence does not rule out underlying neoplasia. This finding is particularly true in dogs, in which subtle abnormalities may be missed because of the large number of chromosomes of similar size and morphology. Also, some human patients with EL present before demonstrable cytogenetic abnormalities have occurred, and later progress to blast transformation in bone marrow and blood, with final emergence of cytogenetic ab-

normalities.[17] Thus, the lack of karyotype abnormalities in these dogs was supportive, but not diagnostic, of IHES. If karyotype abnormalities are demonstrated, techniques such as fluorescent in situ hybridization may be used to localize the abnormality to eosinophils, confirming EL.

The mainstay of treatment for IHES in humans has been glucocorticoids, which suppress cytokine gene transcription and inhibit cytokine-dependent eosinophil survival.[2] Some patients are resistant to glucocorticoids and require treatment with hydroxyurea, vincristine, interferon-α (IFN-α), or cyclosporine. IFN-α promotes a shift from a Th2 cell-dependent response to a T helper type 1 cell-dependent response, and appears especially promising.[2] Other treatments, such as antibodies directed against Il-5, also are being investigated.

The lack of immature eosinophils and karyotype abnormalities, presence of the SH phenomenon in case 1, increased serum IgE concentration, and lack of an apparent underlying cause in the dogs reported here support a diagnosis of IHES. The SH phenomenon in case 1 and high serum IgE concentrations suggest that the condition may represent a dysregulated immediate-type hypersensitivity reaction to an unidentified antigen. The severity of GI involvement in case 1 and the chronicity of the gastric lesions suggest that initial antigen exposure may have occurred in the stomach. In the future, cytogenetic analysis and measurement of serum IgE and cytokine concentrations may prove useful to further characterize IHES and clonal eosinophilic disorders in both dogs and cats.

Acknowledgments

We acknowledge Heska Corporation for performing the serum IgE assay, and T. Rohman, K. Anderson, and F. Bell for their assistance with dogs 2 and 3.

References

1. Weller PF, Bubley GJ. The idiopathic hypereosinophilic syndrome. Blood 1994;83:2759–2779.

2. Rothenberg ME. Eosinophilia. N Engl J Med 1998;338:1592–1600.

3. Huibregtse BA, Turner JL. Hypereosinophilic syndrome and eosinophilic leukemia: A comparison of 22 hypereosinophilic cats. J Am Anim Hosp Assoc 1994;30:591–599.

4. Latimer KS, Bounous DI, Collatos C, et al. Extreme eosinophilia with disseminated eosinophilic disease in a horse. Vet Clin Pathol 1996;25:23–26.

5. Fox JG, Palley LS, Rose R. Eosinophilic gastroenteritis with Splendore-Hoeppli material in the ferret (*Mustela putorius furo*). Vet Pathol 1992;29:21–26.

6. Corcoran BM, Thoday KL, Henfrey JI, et al. Pulmonary infiltration with eosinophils in 14 dogs. J Small Anim Pract 1991;32:494–502.

7. Regnier A, Delverdier M, Dossin O. Segmental eosinophilic enteritis mimicking intestinal tumors in a dog. Canine Pract 1996;21:25–29.

8. van der Gaag I, van der Linde-Sipman JS. Eosinophilic granulomatous colitis with ulceration in a dog. J Comp Pathol 1987;97:179–185.

9. Bennett PF, Allan FJ, Guilford WG, et al. Idiopathic eosinophilic meningoencephalitis in rottweiler dogs: Three cases (1992–1997). Aust Vet J 1997;75:786–789.

10. Calvert CA, Mahaffey MB, Lappin MR, et al. Pulmonary and disseminated eosinophilic granulomatosis in dogs. J Am Anim Hosp Assoc 1988;24:311–320.

11. Goto N, Kawamura M, Inoue M, et al. Pathology of two cases of canine disseminated hypereosinophilic disease. Jpn J Vet Sci 1983; 45:305–312.

12. Jensen AL, Nielsen OL. Eosinophilic leukemoid reaction in a dog. J Small Anim Pract 1992;33:337–340.

13. Ndikuwera J, Smith D, Obwolo MJ, et al. Chronic granulocytic leukaemia/eosinophilic leukaemia in a dog? J Small Anim Pract 1992; 33:553–557.

14. Lillihehöök I, Gunnarsson L, Zakrisson G, et al. Diseases associated with pronounced eosinophilia: A study of 105 dogs in Sweden. J Small Anim Pract 2000;41:248–253.

15. Hayden DW, Van Kruiningen HJ. Eosinophilic gastroenteritis in German Shepherd Dogs and its relationship to visceral larva migrans. J Am Vet Med Assoc 1973;162:379–384.

16. Brito-Babapulle F. Clonal eosinophilic disorders and the hypereosinophilic syndrome. Blood Rev 1997;11:129–145.

17. Juneja S, Stewart J, McKenzie A, et al. Hypereosinophilic syndrome or chronic eosinophilic leukemia: Report of a case with a lytic bone lesion. Leukemia 1997;11:765–766.