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Demographics and clinical picture of nonseasonal canine atopic dermatitis – observations in 63 dogs

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The objectives of this multicentre study were to describe demographics and clinical findings of a group of 63 dogs with non-seasonal atopic dermatitis (AD), where concurrent flea allergy dermatitis, ectoparasite infestation, food adverse reactions and secondary infections had been ruled out. The breed, sex, age of onset, distribution of skin lesions, prevalence of otitis externa, secondary skin infections and additional noncutaneous clinical signs were recorded. A veterinarian recorded skin lesions from 15 different body regions. Each body region was scored according to degree of erythema, alopecia, excoriations, scale, crusts, lichenification and hyperpigmentation that was present. An early age of onset (37% of the dogs being less than 1 year old) was more common in this group of dogs than described in the literature. The German shepherd breed was over-represented (21% as compared with 7–9.9% of the veterinary clinic population) and approximately half of the German shepherd dogs (46%) started showing clinical signs before 1 year of age. Erythema was the overall most common type of skin lesion, with facial erythema and conjunctivitis being less commonly reported than in other studies. Feet, ears and groin were the most common sites for skin lesions. This study indicates that it is not uncommon for dogs with nonseasonal AD to have the onset of clinical signs start at a younger age (less than 1 year of age) and have clinical lesions that vary from previously published reports. This discrepancy might be allergen-dependent or breed-related.

The study was funded by Boehringer Ingelheim Vetmedica, Denmark.

Easy way to differentiate short-tailed demodectic mite from *Demodex canis*

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Mixed infestations of *Demodex canis* and a short-tailed demodectic mite were reported from several countries since late the 1980s. Differentiations based on measurement of the length of this short demodectic mite have been inconclusive until now. Over 500 adult demodectic mites were collected by tape strip and skin scrapings from a 4-year-old intact male Shih Tzu dog with severe juvenile-onset generalized demodicosis. The dog had a mixed infestation of *D. canis* and the short-tailed demodectic mite. Two studies were performed on the mites. In the first study, 200 mites were divided into two groups based on different morphology. Group 1: mites short body, rounded end of abdomen, one to three fold lines bilaterally on the abdomen. These criteria were used by the investigators to define the 'short-tailed' demodectic mite. Group 2: mites with a slender and tapering free body end and no bilateral abdominal fold lines. One hundred mites were counted in each group. In the second study, 298 mites' measurements were made independent of body shape. Total body length, width and body ratio of gnathosoma plus podosoma (= prosoma) to opisthosoma were measured. The total body length and width did not easily differentiate the two mites. The body ratio plus morphological characteristics of the rounded end of the abdomen appear to be the easiest method to identify the short-tailed demodectic mite in the clinic.

This study was supported by the Association of Veterinary Dermatology, Taipei.

Topical 0.1% tacrolimus for the treatment of discoid lupus erythematosus and pemphigus erythematosus in dogs

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Skin lesions of discoid lupus erythematosus (DLE) and pemphigus erythematosus (PE) are often localized to the nasal planum, dorsal muzzle and less commonly, pinnae, periocular skin and lips. The objective of this study was to explore the use of 0.1% tacrolimus as a topical therapy in the management of localized DLE and PE lesions in dogs. Ten cases of DLE and two cases of PE in various breeds were included. Degree of erythema, crust, ulceration or erosion, depigmentation and scarring were evaluated at the start of the trial and after 2, 4 and 8 weeks of therapy. Complete blood count, serum chemistry and whole blood tacrolimus concentrations (Abbot IMx Microparticle Immunoassay; Abbot IMx, Chicago, IL, USA) were performed at each evaluation. Ten cases of DLE and two cases of PE were treated using the lotion either as a sole therapy (both DLE) or as an adjunctive treatment (eight DLE and two PE). Five of the cases evaluated had an excellent response (three DLE and two PE), five a partial response (all DLE), and two dogs (both DLE) showed no appreciable difference after receiving topical tacrolimus therapy. Concurrent medications were discontinued in eight of the ten dogs that improved. No adverse effects in clinical or laboratory parameters were noted throughout the study. However, measurement of tacrolimus concentrations by the MEIA assay was of little benefit due to false-positive measurements. Results of this study indicate that topical 0.1% tacrolimus may be a safe and effective adjunctive therapy for DLE and PE, with fewer systemic and topical adverse effects than current treatment modalities.

Allergen-specific immunotherapy in dogs with atopic dermatitis and house dust mite hypersensitivity: use of symptom/medication scores

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There is no published literature on the efficacy of allergen-specific immunotherapy (ASIT) for dogs with atopic dermatitis (AD) and house dust mite (HDM) hypersensitivity. Symptom/medication (SM) scoring is the standard measure of clinical disease during ASIT in humans. The purpose of this study was to determine the response to ASIT in dogs with AD and HDM hypersensitivity using SM scores, and compare results with the owner's global assessment and the investigator's clinical evaluation. All dogs had nonseasonal AD, positive intradermal test reactions to HDM, and were treated for 12 months with ASIT that included HDM allergens. Evaluations at 0, 3, 6, 9 and 12 months included: daily owner symptom (pruritus, lesions, coat character) and medication (all anti-inflammatory medications) scoring for 28 days; owner global assessment; and investigator clinical sign score. Occurrence of skin infections was documented throughout the study. Statistical analysis of SM scores at 0 and 12 months was performed using Student's *t*-test. Nineteen dogs completed the study. Fourteen (74%) dogs showed significant improvement ($P < 0.05$) in SM score. A 50% improvement in SM score, owners' global assessment and clinical sign score was seen in 12 (63%), 17 (89%) and 10 (53%) dogs, respectively. In the first 6 months of ASIT, 17 episodes of skin infections (12 dogs) were recorded, and 13 episodes (10 dogs) in the final 6 months. The majority of dogs with AD and HDM hypersensitivity improved with ASIT. SM scores are a useful semiquantitative measure of clinical disease. Many dogs continued to have skin infections.

This study was funded by the Ohio Animal Health Foundation.

The clinical and immunological reaction to a flavoured monthly oral heartworm prophylactic in 12 dogs with spontaneous food allergy

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There is debate among practicing veterinary dermatologists as to whether flavoured prophylactics should be avoided in the food-allergic dog. At North Carolina State University we have a colony of dogs with spontaneous food allergy. They are maintained on a hypoallergenic diet (Prescription diet d/d Duck and Rice, Hills Pet Nutrition, Topeka, KS). A flavoured chewable tablet containing pork liver, soy and 2.3 mg of milbemycin (Interceptor Flavor Tabs, Novartis Animal Health, Greensboro, NC) was administered under controlled conditions to 12 dogs. Previous exposure to this medication was documented. A physical examination was performed prior to, and on four occasions after, tablet administration. A clinical score (CS) was assigned to determine the severity of skin and otic disease. Serum was collected at 3- to 5-day intervals. Allergen-specific IgE (soy, pork and corn) was measured by ELISA. Statistical analysis was performed using SAS software (Cary, NC, USA) and a longitudinal mixed model was employed for the evaluation of the clinical response. An increased CS was observed in 10 dogs post-challenge; peak values were measured on day 2 (five dogs) and day 5 (five dogs). When compared with pre-treatment CS this increase was statistically significant ($P < 0.05$). Significant peaks in serum allergen-specific IgE were measured on days 5 and 20 to soy, pork and corn. The authors conclude that these dogs have a rapid adverse clinical response to this flavoured medication which is accompanied by a significant increase in serum allergen-specific IgE, although this does not necessarily correlate with the orally administered allergen.

A retrospective study of cutaneous neoplasms in domestic rabbits (1990–2001)

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Spontaneous skin tumors in privately-owned domestic rabbits, particularly nonviral-associated neoplasms, have been sporadically documented in the veterinary literature. The purpose of this study was to retrospectively survey cutaneous neoplasms submitted from veterinary practitioners for routine histopathological evaluation. Four hundred and forty surgical pathology specimens from lagomorphs were submitted to the Laboratory of Pathology Diagnostic Service at the University of Pennsylvania, School of Veterinary Medicine, from 1990 through 2001. Of those, 139 were cutaneous neoplasms. Trichoblastomas (basal cell tumors) were the most commonly diagnosed skin neoplasm (total = 45) and comprised 32% of all skin tumors, while three trichoepitheliomas (2%), and three papillomas (2%) were also identified. Malignant epithelial neoplasms (10%) consisted of four squamous cell carcinomas, three apocrine carcinomas, one basal cell carcinoma, one sebaceous carcinoma and five carcinomas that were not further classified. Two cases of cutaneous malignant melanoma were found. Cutaneous Shope and non-Shope fibromas (34) comprised 25% of the cases. Seven lipomas (5%) were diagnosed. Malignant mesenchymal neoplasms consisted of 29 soft tissue sarcomas (21%). One case of cutaneous epidermotropic lymphosarcoma and one nonepitheliotropic malignant round cell tumor were also identified. In this study, trichoblastomas were found to be the most common nonviral-associated cutaneous neoplasm.

Ultrastructural findings of feline eosinophilic dermatoses

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Despite the increasing evidence that degeneration of collagen is not involved in the histogenesis of the eosinophilic debris observed with light microscopy in feline eosinophilic dermatoses (FED), flame figures and foci of 'collagen degeneration' are still reported as degenerated collagen mixed with degranulated eosinophils. The purpose of this study was to investigate ultrastructurally eosinophil degranulation pathways and dermal collagen morphology in FED. Four patients with histopathological diagnosis of FED, three clinically diagnosed with eosinophilic granulomas and one with an eosinophilic plaque, and one healthy control were studied. Areas that contained eosinophils, collagen, flame figures and 'collagen degeneration' were ultrasectioned and examined with transmission electron microscope. Eosinophils (48 to 123 per cat) were classified into resting (intact cells), undergoing piecemeal degranulation (PMD) (viable cells with partially empty granules) and cytolytic [disrupted cells with clusters of free eosinophil granules (Cfegs)]. Longitudinal and transverse sections of collagen fibrils were also evaluated. 9.01% of eosinophils examined were resting, whereas 90.98% had released their content, 70.14% by cytolysis and 20.84% by PMD. Collagen fibrils showed no ultrastructural abnormalities and appeared identical to those observed in the control. Flame figures and foci of 'collagen degeneration' appeared constituted by collagen fibrils—fibers interspersed with cytolytic eosinophils. Based on these findings we conclude that eosinophils play an active role in the pathogenesis of FED through their granule content release, by cytolysis and PMD. Moreover, based on the normal collagen ultrastructure, the use of the term 'collagen degeneration' to describe the eosinophilic debris observed with light microscopy in FED should be definitely avoided.

The study was funded by a grant of the European Society of Veterinary Dermatology.

Comparative sequence analysis of three canine type 2 keratins

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In order to better define the cornification process in dogs and to understand how it is similar or different from cornification in other mammals, we are sequencing the major molecules of cornification in the dog. In this report, we describe the complete genomic sequence of three superficial type 2 canine keratins: K1, K2e and K2p. Oligonucleotide primers for PCR were designed from homologous regions of previously reported human or murine cDNA sequences of the genes of interest. DNA sequencing and evaluation with blast confirmed that the primers amplified the respective canine genes. These primers were then used to screen a subset of a canine genomic library known to contain basic keratin genes. Selected clones were sequenced to obtain the complete DNA sequence. K1, K2e and K2p each had nine exons and eight introns characteristic of the type 2 keratins that code for proteins with variable glycine-rich head and tail regions and a central α -helical rod domain. K2p encoded a protein of 659 amino acids, larger than both K1 and K2e (620 and 634 amino acids, respectively). Amino acid sequence homology with humans was 81, 74 and 81% for K1, K2e and K2p, respectively, with homology greatest at the central rod region and decreasing dramatically at the head and tail. Comparisons of the amino acid sequences of these keratins within the canine species defined K2e and K2p as having a slightly higher degree of similarity. In contrast, in humans K2e has greater sequence homology with K1.

This study was funded in part by the Morris Animal Foundation.

Efficacy of pre-treatment with lufenuron for the prevention of *Microsporum canis* infection in a feline cohabitant-challenge model

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We previously observed a trend towards clinically milder or delayed *Microsporum canis* infections in cats pre-treated with lufenuron, suggesting a possible effect of this drug in limiting dermatophyte growth. We sought to determine the effect of pre-treatment with lufenuron on development of *M. canis* infections induced via exposure to an infected cat. Lufenuron suspension (100 to 140 mg kg⁻¹ orally, once monthly; or 40 mg subcutaneously, every 6 months) or placebo was administered to groups of eight juvenile cats. After 4 months of treatment, cats were challenged by introducing cats with mild, experimentally induced *M. canis* infections into the rooms. The extent of any resulting infection was monitored for 22 weeks after challenge. All cats became infected with *M. canis*. Cats treated with lufenuron had significantly lower infection scores during the early weeks following exposure, and there was a more prolonged initial progression phase of the infection. Once infections reached peak intensity, they resolved over a similar time period in both treated and control groups. We conclude that lufenuron, when used at the dose schedule and under the conditions in this study, did not prevent dermatophyte infection by exposure to an experimentally infected cat. The delay in establishment of infection may reflect an inhibitory effect of lufenuron on the organism that is measurable, but not sufficient to prevent development of infection under these conditions.

This study was supported by Novartis Animal Health.

Serological responses to house dust mite antigens in atopic dogs while receiving allergen-specific immunotherapy

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In human atopic disease a beneficial response to immunotherapy may be associated with humeral changes in serum IgE and subclass IgG. The aim of this study was to investigate the humeral response to allergen-specific immunotherapy (ASIT) in dogs with atopic dermatitis. Twelve dogs with clinical signs consistent with atopic dermatitis and positive intradermal test reactions to house dust mite antigens (Greer Laboratories, Lenoir NC, USA) were studied. The clinical response to ASIT (Greer) was evaluated when blood samples were collected at 0, 2 and 4 weeks; and at 3, 6 and 12 months after starting ASIT. The ASIT was administered for a minimum of 9 months. The serum concentration of total and subclass IgG antibodies was measured against *Dermatophagoides farinae* and *Dermatophagoides pteronyssinus* antigens (Greer) by ELISA using polyclonal and monoclonal antidog IgG reagents, respectively. Serum IgE was measured using a Fc-epsilon R1 receptor alpha chain-based ELISA. The data were analysed using a multilevel model in MLwiN software (Institute of Education, University of London, UK). Antibody concentrations of IgE and total IgG increased during the period of administration of ASIT and decreased after ASIT was stopped. There was no obvious pattern in subclass-IgG concentrations during ASIT. Response to ASIT was excellent (four dogs), equivocal (three dogs) or poor (five dogs). During ASIT a poor or equivocal response was associated with a statistically significant increase in IgE to *D. pteronyssinus* antigen compared with the excellent responders. This relationship was present to a lesser extent with IgE to *D. farinae* and total IgG to both antigens.

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Serological responses to food antigens in normal and atopic dogs, and dogs with gastrointestinal disease

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In human atopic disease with concurrent food allergy there may be significant increases in serum IgE. Serological methods have not been advocated for suspected food allergy in dogs. The aim of this study was to investigate humeral responses to food antigens in dogs. Serum IgG and IgE antibodies were measured by ELISA against food antigens using polyclonal antidog IgG and IgE reagents. Antigens included beef, chicken, pork, lamb, chicken, turkey, white fish, whole egg, wheat, soybean, barley, rice, maize corn, potato, yeast and cow's milk. Three groups were used: normal dogs ($n = 40$ for IgE and $n = 91$ for IgG), dogs with atopic dermatitis ($n = 91$), and dogs with four types of gastrointestinal disease ($n = 64$ for IgE and $n = 72$ for IgG). The gastrointestinal (GI) subgroups included bacterial overgrowth, inflammatory bowel disease (IBD), food-responsive and other GI diseases, but statistically significant differences were not detected between them. Statistically significant differences were detected for IgE data for all antigens, and for all except egg and yeast for IgG, using χ^2 -test and analysis of variance, respectively. Compared with other groups, atopic dogs had more IgE and dogs with GI disease had more IgG. The relationship of antigen responses for pooled data was analysed using principle component analysis and a cluster plot. Some clustering of variables was apparent for both IgE and IgG. For example for IgG, dog response to chicken and turkey was similar. Using multinomial regression, a predictive model for disease type was formulated with the IgG and IgE response as predictor variables.

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The association of *Staphylococcus schleiferi* with canine pyoderma

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Staphylococcus schleiferi is a recently recognized organism with potential pathogenic implications in human and veterinary medicine. The purpose of this study was to determine the frequency of *S. schleiferi* in canine pyodermas and see if methicillin resistance (MR) is associated with this organism. Cultures were obtained from dogs with pyoderma over a 1-year period. It was determined if the pyoderma was a first-time or recurrent one, and if the dog was receiving antibiotics at the time of culture. Of 15 dogs with first-time pyoderma, four dogs were receiving antibiotics at the time of culture: *S. schleiferi* was not cultured from any dog in this group. Cultures were obtained from 27 dogs with recurrent infection that were not receiving antibiotics: *S. schleiferi* was cultured from five of these dogs; three of the five *S. schleiferi*-cultured isolates were MR. Cultures were obtained from 12 dogs with recurrent pyoderma while receiving antibiotics: *S. schleiferi* was cultured from nine of these dogs; eight of the nine *S. schleiferi*-cultured isolates were MR. Five of the 12 dogs had been cultured previously while not receiving antibiotics. Three of these dogs cultured positive for *S. intermedius* initially, and then -cultured; one dog cultured *S. schleiferi* positive both times; and one dog cultured *S. schleiferi* positive, then *S. intermedius* positive. To the authors' knowledge, this is the first report of *S. schleiferi* associated with canine pyoderma. Results indicate that *S. schleiferi* is cultured more often from dogs with recurrent pyodermas while receiving antibiotics. MR is frequently associated with *S. schleiferi*.

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Retrospective evaluation of steroid hormone intermediates in dogs with alopecia

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The purpose of this study was to determine if there are specific steroid hormonal aberrations associated with suspect endocrine alopecias in dogs where hypothyroidism and hyperadrenocorticism have been excluded. Steroid hormone panels submitted to the UTCVM endocrinology laboratory over a 7.5-year period (783 samples) from dogs with alopecia were reviewed. To be included in the final analysis, complete information about age, sex, neuter status and breed was required; hypothyroidism had been ruled out; and cortisol concentration post-ACTH stimulation was less than 200 ng mL⁻¹. A total of 276 dogs met the criteria for inclusion (seven intact females, 32 intact males, 134 spayed females, 103 neutered males) and comprised of 54 different breeds. Approximately 73% of dogs had at least one steroid hormone intermediate greater than the normal range. The most frequent hormone elevation was noted in baseline and/or post-ACTH stimulation progesterone (57.9% of samples). Breeds that contained seven or more dogs were statistically compared. Only malamute dogs failed to show an increase in progesterone for any dog. When compared with normal dogs, oestradiol was significantly greater in keeshond dogs and baseline progesterone was significantly greater in pomeranians and husky breed dogs. While significant difference existed among breeds, neither baseline nor post-stimulation 17-hydroxyprogesterone differed significantly from normal values. Chow-chow and malamute dogs had the greatest percentage of dogs with completely normal steroid hormone intermediates. Results of this study suggest that the pathomechanism of the alopecia, at least for some breeds, may not relate to steroid hormone intermediates.

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Evaluation of the clinical and serum IgE responses to oral challenge with cornstarch, corn, soy and a soy hydrolysate diet in dogs with known food allergy

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Hydrolysate diets have been advocated in the management of the food-allergic dog, but little information is available on their performance in dogs with known allergies. Fourteen dogs with known allergies to soy and corn were maintained on a limited antigen diet exclusively for 80 days (Hills d/d Rice and Duck, Hills Pet Nutrition, Topeka, KS). This diet was continued during oral challenge with 200 mg kg⁻¹ of cornstarch, corn and soy respectively on two occasions each at 24-h intervals. There was a minimum period of 15 days between each challenge. A diet containing hydrolysed soy protein and cornstarch (CNM HA, Ralston Purina Co., St. Louis, MO) was then introduced. Before and after each oral challenge the dogs were examined and a clinical score was determined; serum was also collected for measurement of allergen-specific and total IgE concentrations. Statistical analysis employed the use of a longitudinal mixed model and was performed using SAS software (Cary, NC). Significant increases in clinical scores were seen after oral challenge with cornstarch, corn and soy ($P = 0.04$, 0.002, 0.01 respectively) but not with HA ($P = 0.5$), although three dogs did develop clinical signs after this latter challenge. Allergen challenge did not affect total serum IgE concentrations. Although increases in soy- and corn-specific serum IgE were measured in individual dogs post-challenge they were not statistically significant, and could not be used to predict clinical hypersensitivity.

This study was funded by a grant from Ralston Purina Co., St. Louis, MO.

Changes in IgE antibodies to soy in sensitized and control dogs after challenge using three diets in a crossover design

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The objective of the study was to test the hypothesis that food-allergen-sensitized dogs would produce higher levels of IgE antibodies to soy after challenge with an intact soy diet, compared with egg-based (elimination diet), hydrolyzed soy- and corn-based diets. Eight dogs were sensitized with six food allergens by subcutaneous injections. Seven dogs were used as nonsensitized controls. All dogs were fed an elimination diet containing egg and Brewer's rice (to neither of which any dog had been sensitized) for six weeks. Serum samples were collected, frozen (-20°C), and submitted to Greer Laboratories® for ELISA analysis. The 15 dogs were randomly assigned into three groups and fed a pelleted diet consisting of hydrolyzed soy/corn starch, intact soy/corn starch or intact corn diet using a crossover design. After a three-week challenge, serum samples were collected. All dogs were placed on the elimination diet during the 6-week washout periods. ELISA results were expressed as relative values (%R) compared with pooled canine sera. There was a statistically significant difference between sensitized and control dogs for all diets including the elimination diet (SASS®). While there was a modest %R increase when intact soy was fed, no statistically significant differences in serum IgE to soy were found for any of the diets. We were unable to reject the null hypothesis ($P < 0.05$).

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Transfer of passive cutaneous anaphylaxis to *Malassezia pachydermatis* by the serum of atopic dogs with *Malassezia dermatitis*

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In atopic dogs with *Malassezia dermatitis* (MD), type 1 hypersensitivity reactions and circulating serum IgE reactive to *M. pachydermatis* extracts have been reported. The purpose of this study was to evaluate the functionality of anti-*Malassezia* IgE by the transfer of passive cutaneous anaphylaxis (PCA testing). Three nonatopic dogs were used as recipients. Two test sera were utilized: pooled sera from 10 atopic dogs with MD that were skin test positive to *M. pachydermatis* extract; and serum from an atopic dog with MD that exhibited high levels of anti-*Malassezia* IgE on an ELISA assay. One aliquot of each untreated serum, one aliquot of each serum heated at 56 °C for 4 h and two aliquots of the ELISA-positive serum adsorbed with either mouse anticanine IgE or bovine serum albumin (as a control) were utilized. Six serial dilutions of all six aliquots were injected intradermally in triplicate, along with a 20 µg mL⁻¹ extract of *M. pachydermatis* to rule out prior sensitization to the yeast. At 24-, 48- and 72-h intervals, the yeast extract was injected intradermally at the sites of prior serum injections and wheal/flare responses were graded 0–4+ compared with saline and histamine controls. Through 72 h, all dogs showed $\geq 2+$ reactions at the injection sites of both the untreated sera and the BSA-treated sera (titers = 1:64), but no reactions at the sites of heat-inactivated and IgE-adsorbed sera. These results support the transfer of PCA by anti-*Malassezia* IgE and implicate it to be functional in type 1 hypersensitivity.

Funded by the Canine Health Foundation of the American Kennel Club.

Positive 'atopy patch tests' reactions in IgE-hyperresponsive beagle dogs are dependent upon elevated allergen-specific IgE serum levels and are associated with IgE-expressing dendritic cells

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In human patients with atopic dermatitis, the development of inflammation after epicutaneous application of allergens is dependent upon the presence of IgE and Langerhans' cells. The purpose of this study was to investigate the role of allergen-specific IgE in the generation of positive patch test reactions in experimentally sensitized IgE-hyperresponsive beagle dogs. Patch tests were performed on seven dogs with *Dermatophagoides farinae* (Df) and flea saliva (FS) allergens or saline control. Two of the dogs had elevated serum IgE against Df, one against FS, two against both and two were not hypersensitive to either allergen. Positive macroscopic reactions consisted of erythema, oedema and induration, and developed between 24 and 96 h after allergen application. Macroscopic and microscopic patch test reactions developed only when serum IgE was present against tested allergens. Serum IgE levels were positively correlated with dermal cell counts at 48 h (Spearman $r = 0.59$; $P = 0.03$) and 96 h ($r = 0.68$; $P < 0.01$) after challenge. In Df-sensitized dogs, positive patch tests were associated with numerous IgE-staining epidermal and dermal dendritic cell aggregates. Dendritic cells positive for IgE also expressed CD1c, identifying cells as Langerhans' cells. These observations suggest that the development of positive patch test reactions are IgE-dependent, and that macroscopic and microscopic patch test lesions resemble those seen in dogs with spontaneously-arising atopic dermatitis. 'Atopy Patch Tests' therefore could provide a good model for investigating the pathogenesis and treatment of atopic skin lesions in the canine species.

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Therapeutic response to pentoxifylline and its active metabolites in dogs with dermatomyositis

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The purpose of this study was to determine a therapeutic dose for Trental® (pentoxifylline) when used to treat dogs with dermatomyositis. Eleven dogs diagnosed with dermatomyositis based on history, physical examination and biopsy were treated with pentoxifylline ($46.5 \pm 8 \text{ mg kg}^{-1}$ divided twice daily) with food. Biweekly for 3 months, urinalyses, complete blood counts, serum chemistries and therapeutic drug monitoring (peak collected at 3 h and trough before the next dose) were collected. Dermatological lesions were assessed during each visit and given a score (0 = nonexistent, 1 = mild, 2 = moderate and 3 = severe). Serum concentrations of pentoxifylline (pentoxo) and metabolites 1 (M1) and 5 (M5) were measured by high-performance liquid chromatography. Descriptive statistics were generated for the drug concentrations. Although all animals responded to therapy, the time to initial response (skin lesions changed to a lower score compared with the initial visit) varied between all of the dogs. The range was 4 to 10 weeks, with a median of 6 weeks of therapy. Laboratory tests did not differ between the first and last sampling times (month 3). Pentoxifylline and M1 and M5 concentrations (ng mL^{-1}) for the 11 dogs varied between dog and across time for each dog. In general, concentrations for all three compounds were detectable only with peak (3 h) samples. Concentrations for M5 were highest for all dogs at each sampling time, and appeared to be the most consistent and characterized by the least variability. Concentrations were not detectable in one dog for pentoxo or M1 at any time, but were detectable for M5. Data for this dog were not included. Based on the response to therapy in this study, a dose 25 mg kg^{-1} of pentoxifylline administered every 12 h appears to be an effective beginning dose. Because of variability in disposition, including metabolite formation, monitoring of pentoxifylline may not offer a therapeutic advantage.

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Comparison of the average percentage of thyroid colloid and selected serum thyroid analyses in healthy euthyroid and severely ill dogs

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The diagnosis of canine hypothyroidism is not straightforward because the results of thyroid tests can be influenced by severe illness and medications. The need to find diagnostic tests that are not affected by these factors is urgent. The objectives of this investigation can be summarized as follows: (1) to establish a reference range for percentage colloid from healthy euthyroid dogs; (2) to compare the mean percentage colloid of the healthy dogs with that of severely ill dogs; (3) to define a group of ill-euthyroid dogs based on the percentage colloid and evaluate their serum concentrations of total thyroxine (TT4), free thyroxine by equilibrium dialyses (fT4d) and endogenous TSH (eTSH); and (4) to determine if the mean percentage colloid correlates with the serum concentrations of TT4, fT4d and eTSH. Sixty-one healthy-euthyroid dogs and 66 severely ill dogs were included on the study. Thyroid glands were removed immediately after death. Morphometric analyses were performed on each thyroid lobe to determine the percentage colloid. Serum hormone concentrations were analyzed using commercially available tests validated at our laboratory. Descriptive statistics were used to report the results of the average percentage colloid and hormone concentrations. Unpaired Student's *t*-test was applied to compare the percentage colloid between the group of healthy-euthyroid dogs and the groups of sick dogs. Spearman's rank correlation coefficient method was used to determine the correlation between each of the measured thyroid hormone values and the percentage colloid. The statistical analyses were carried out using Statistical Analysis System (SAS) software. A *P*-value of < 0.05 was considered statistically significant. No statistically significant difference was observed in mean percentage colloid between the groups of severely ill and healthy-euthyroid dogs. Serum TT4 and fT4d concentrations were below the reference range in 57.6 and 31.8% of severely ill dogs, respectively. Only 6.1% of severely ill dogs had increased eTSH concentrations. No clinically significant correlation was found between serum thyroid hormones and percentage colloid. In conclusion, serum TT4 and fT4d concentrations were more frequently altered by severe illness than eTSH in dogs with percentage thyroid colloid equivalent to normal dogs. Serum thyroid hormones did not have a direct correlation with percentage colloid.

Pharmacodynamics of clemastine in healthy horses

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The aim of this study was to evaluate the efficacy of the H₁-receptor inhibitor clemastine (a compound belonging to the first generation of antihistamines) in equine skin after a single intravenous and oral dose. Histamine binds to H₁-receptors involved in pruritus, increased vascular permeability, release of inflammatory mediators and recruitment of inflammatory cells. To counteract these effects antihistamines have classically been used for treatment of allergic hypersensitivity disorders and urticaria in several species including dogs, cats and horses. Six horses received clemastine orally, 200 µg kg⁻¹, by stomach tube and 50 mg kg⁻¹ as an intravenous infusion in a crossover study with crossover design. The ability to reduce wheal formation after intradermal injection of histamine was evaluated. Intradermal injections of 0.07 mL histamine dihydrochloride (0.1 mg mL⁻¹) were given before and at seven time periods ranging from 0.5 to 24 h after clemastine administration. Wheal area was measured 20 min after the injection. Sterile saline (0.07 mL) served as a negative control. There was a marked reduction in wheal size for approximately 6 h after the intravenous administration, indicating that clemastine is distributed to, and effective in inhibiting H₁-receptors in, equine skin. After oral administration the effect was almost negligible, indicating low oral bioavailability of clemastine in the horse.

The study was funded by Linnea and Axel Ericsson's Grant fund at the Veterinary Faculty, Swedish University of Agricultural Sciences.

A pilot trial on the effect of Cyclosporin A on intradermal skin test reactions in dogs with atopic dermatitis

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The purpose of this study was to determine if Cyclosporin A, given at a dose of 5 mg kg⁻¹ per day for 42 days, negatively influenced intradermal skin test results in dogs with atopic dermatitis. Six mixed-breed dogs of varying ages and sexes with a clinical diagnosis of atopic dermatitis were included in this study. All dogs were intradermally skin tested (IDT) with 61 allergens from varying groups, grass pollens, weed pollens, tree pollens, moulds, insects and dust mites. IDT reactions were recorded as absent, weak, moderate or strong reactions. All dogs showed several strong reactions in the initial IDT. On days 0 to 42, the dogs received 5 mg kg⁻¹ per day of Cyclosporin A. On day 42, all dogs showed a marked reduction in pruritus. A second IDT was performed and the results of both tests compared. The χ^2 -test was used to statistically analyse changes between pre- and post-treatment reactions for each of the allergen groups. There were no mould reactions recorded and this group was discarded from analysis. When all reactions were used in analysis, Cyclosporin A tended to significantly reduce the number of IDT reactions. However, when only moderate and strong reactions were considered (weak reactions excluded) there were no significant differences between pre- and post-treatment results for the other allergen groups. Cyclosporin A at a dose of 5 mg kg⁻¹ per day for 42 days controlled pruritus, but did not significantly alter moderate and strong intradermal skin test reactions in the six dogs tested. This suggests that Cyclosporin A, at this dose, did not prevent mast cell degranulation.

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Concurrent hyperadrenocorticism in a miniature schnauzer with severe *Trichophyton mentagrophytes* infection

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A 4-year-old intact female schnauzer dog was presented with a history greater than 1 year of alopecia, thick crusting and secondary seborrhea with mild pruritus. The dog was diagnosed pituitary-dependant hyperadrenocorticism (PDH) concurrent with severe *Trichophyton mentagrophytes* infection. The dog showed generalized alopecia, enlarged pendulous abdomen, mild polydipsia, polyuria and polyphagia. Liver enzymes alanine aminotransferase (ALT) and alkaline phosphatase (ALKP) were normal. ACTH stimulation and low-dose dexamethasone suppression tests were performed prior to therapy. The concentrations of serum cortisol were determined by radioimmunoassay (RIA) and direct enzyme immunoassay (EIA) methods. The results of both assay systems indicated PDH. Fungal cultures showed *T. mentagrophytes* infection in 4 days. Histological examination of a skin biopsy specimen revealed severe irregular acanthosis accompanied by compact hyperkeratosis, and numerous dermatophytic spores and hyphae heavily colonized in the keratin layer. After 3 months of oral terbinafine (25 mg kg⁻¹, once daily) and mitotane therapy, the hair regrew and the result of an ACTH stimulation test were normal. Three sets of biweekly fungal cultures were negative after 6 months. In this case, terbinafine was given concurrently with mitotane without detectable adverse effects (no abnormal clinical signs were observed and liver enzymes remained within normal ranges). Thus, the combination of terbinafine and mitotane appears to be a safe therapeutic protocol in treatment of PDH with dermatophytosis.

This study was supported by The Association of Veterinary Dermatology, Taipei.

Systemic candidiasis due to *Candida albicans* in a dog

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In this case report, we describe pyogranulomatous lymphadenitis due to *Candida albicans* that progressed to a systemic infection in a 10-kg, 1-year-old spayed female mixed-breed dog. The dog had bilaterally enlarged submandibular lymph nodes with peripheral oedema. Fine-needle aspirate of fluid from a lymph node and associated area was collected for fungal culture; a skin biopsy was also performed at the same time. Growth of budding yeast was observed on mycobiologic agar subcultured via inoculation in human serum. The characteristic germ tube of *C. albicans* was observed. Histopathology revealed chronic pyogranulomatous lymphadenitis with fungal elements. Lymphadenitis due to *C. albicans* was diagnosed tentatively. The dog was treated initially with itraconazole (30 mg kg⁻¹ orally once daily) without an adequate response; treatment was changed to fluconazole (150 mg every other day). Leucocytosis, lymphocytosis, elevated alanine aminotransferase (ALT) and alkaline phosphatase (ALKP) were observed to wane and wax during 4 months of therapy. The skin lesions did not respond to fluconazole therapy and progressed to involve the subcutaneous scapular and ocular regions, and the dog developed generalized lymphadenopathy. Finally, the dog appeared to seizure and the owner requested euthanasia. At necropsy, gross lesions showed disseminated white spots on the cerebrum, pancreas, liver, kidney, lung, heart and all the lymph nodes, especially the abdominal lymph system. *Candida albicans* was also cultured from the spinal fluid. No obvious invasion site was noted and the dog did not have a previous history of long-term usage of corticosteroids and/or antibiotics.

This study was supported by The Association of Veterinary Dermatology, Taipei.

***Hepatozoon canis*, a fortuitous or pathogenic agent in canine dermatology: a review of three cases**

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The role played by *Hepatozoon canis* is subject to considerable discussion, as its presence does not consistently appear to be pathogenic. Should the pathology be severe, blood samples or medullary specimens enable the parasite to be isolated, either on its own or associated with other parasitic agents such as *Ehrlichia canis*. To identify it in a dermatological context is a much rarer occurrence. A description is provided of three cases in which the search for an underlying cause of disease revealed the presence of the parasite; the role of the parasite in the process was unclear. A 9-year-old Beauceron dog did not recover from a case of oral papillomatosis. A loss of weight, generalized adenomegaly and splenomegaly completed the clinical picture. The myelogram showed that *H. canis* gametocytes were present. Therapy with imidocarb and toltrazuril neither improved the dog's general condition nor did it have a clinical impact on the oral papillomatosis. An 11-year-old Beauceron bitch was examined for pyoderma on the foreleg and a hind limb digit. Antibiotic treatment did not produce a clinical cure. Two weeks later, cytological examination of exudates revealed the presence of two *H. canis* gametocytes. The patient's condition was improved by the administration of imidocarb and doxycycline. A 10-year-old poodle dog exhibited numerous ulcers, and on histological examination of tissue biopsies and the cutaneous slides, *Leishmania canis* and *H. canis* were identified. There was no clinical response to treatment. *H. canis* should be considered where there are unusual manifestations of skin diseases and lack of response to appropriate therapy.

Four examples of metastatic canine cutaneous nodules

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The causes of single or multiple cutaneous nodules are many. It is important to perform fine-needle aspirations for cytological examination and to perform skin biopsies. In certain cases a surgical excision of the entire nodule, with a subsequent histological examination, is a simpler approach. When the nodule represents a cutaneous metastasis of a primary neoplasia and is the only sign of the tumour, the aetiology of the nodule can sometimes be rather uncommon. The diagnosis may be made more difficult when the lesion is isolated, appears benign, is in an unusual location (cranial aspect of foreleg, dorsal trunk or scapular region) or is difficult to excise due to haemostasis and local tissue infiltration. The histological interpretation can sometimes prove to be a complicated matter, requiring the use of specific markers to identify anaplastic tumour, but nonetheless is necessary in order to be able to characterize the primary tumour. Using these four examples, a description is provided of the distant localization of an ovarian dysgerminoma in an Afghan hound bitch, a mammary adenocarcinoma in a Labrador bitch, and an extraskeletal mammary osteosarcoma in a doberman bitch and a vesicular adenocarcinoma in a Briard dog. In the last two cases cited, the Alamartine–Ball–Cadiot syndrome was in its final stage of development.

Determination of strain variability of *Microsporum canis* to disinfectants

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The isolated infected hair model is a commonly used technique to test the fungicidal efficacy of topical therapies and/or disinfectants against *Microsporum canis*. Results from various laboratories have differed and it is unclear if they are due to methodology and/or strain variability. The objective of this study was to look for strain variability of *M. canis* to disinfectants. Ten strains of *M. canis* were used in this study. Infected hairs and crusts were soaked and processed in order to remove and isolate ectothrix spores. Eight dilutions of the following disinfectants were tested: Virkon® S, (Antec International Ltd, Suffolk, UK), chlorhexidine, lime sulfur, enilconazole and bleach. Sterile distilled water was used as a control. An equal volume of spore suspension and disinfectant solution were incubated for 5 min and then plated onto Mycosel fungal culture plates (Mycosel Agar, Becton Dickinson, Cockeysville, MD, USA). Fungal cultures were counted at days 7 to 10. Fungal culture data was evaluated using an end-point dilution at which there was 100% fungicidal activity, i.e. no growth on the plates. The 10 samples showed identical results. Chlorhexidine and Virkon® S were ineffective even when used at 4× the manufacturer's recommended dilution. Lime sulfur (1:33), enilconazole (20 µL mL⁻¹), and bleach (1:10) were consistently effective when used at the recommended dilution. In addition, lime sulfur and enilconazole were 100% fungicidal even when the recommended concentration was diluted 1:4 or 4× as dilute as recommended. In conclusion, using isolated infective spores of *M. canis* we were unable to detect strain variability to commonly used disinfectants.

This study was funded by the Winn Foundation and the SVM Companion Animal Research Fund.

Prevention of *Microsporum canis* infection in a cat challenge model

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Oral lufenuron is reportedly an effective treatment for some cats with existing dermatophytosis. We sought to determine if lufenuron, when used as a pre-treatment *prior* to challenge exposure and infection, would prospectively protect against development of infection by direct application of *Microsporum canis* spores. Three groups ($n = 6$ per group) of juvenile cats were treated with either monthly oral lufenuron (30 or 133 mg kg⁻¹) or placebo. After 2 months of treatment, kittens were challenged using 10⁵ *Microsporum canis* spores applied to the skin under occlusion. Cats were observed weekly, including: Wood's lamp examination; scoring for scale/crust, erythema, induration; lesion size; and development of satellite lesions. Fungal cultures were performed biweekly. All cats became infected; the infections progressed, and then regressed, in a similar fashion in all groups. There were no consistent statistically significant differences in weekly infection scores throughout the study. Treated cats did not recover faster than untreated cats. We concluded that oral lufenuron at the dosing schedule and conditions used in this study did not prevent dermatophyte infection by direct topical challenge. Further studies are warranted to evaluate alternate dosing schedules, less severe challenge exposures, varying strains of *M. canis*, and other factors that may be present under field conditions.

This study was funded by Novartis Animal Health.

A retrospective study regarding the treatment of idiopathic onychomadesis (lupoid onychodystrophy) in 30 dogs

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The purpose of this retrospective study was to evaluate the efficacy of various therapeutic options in dogs with lupoid onychodystrophy. The treatment records of 30 dogs with lupoid onychodystrophy diagnosed by clinical and histological changes at three institutions (Animal Skin and Allergy Clinic: Melbourne, Colorado State University and Denver Veterinary Specialists) were evaluated retrospectively. The response to various treatments was graded as excellent (normal claws), good (claws still dystrophic, but no associated clinical signs or onychomadesis), partial (still occasional onychomadesis) or poor (no change with treatment). Dogs were treated with either fatty acid supplementation ($n = 17$), doxycycline and niacinamide ($n = 13$), tetracycline and niacinamide ($n = 10$), pentoxifylline ($n = 5$), prednisolone ($n = 4$), azathioprine ($n = 1$) or clofazimine ($n = 1$) or with combinations thereof. Most dogs initially were treated with antibiotics typically effective against cutaneous pathogens, and in most dogs this led to partial improvement only. An excellent or good response was seen in approximately half of the patients treated with tetracycline or doxycycline in combination with niacinamide. Six of the dogs were maintained successfully on fatty acid supplementation, two on pentoxifylline. An excellent response was seen in two of the three dogs treated exclusively with antibiotics. Spontaneous remissions and recurrences made evaluation of success rates difficult. A variety of medications may be useful in the treatment of lupoid onychodystrophy, although the high success rate of fatty acids previously reported could not be confirmed. The study emphasized the varied and often unclear aetiology and natural course of the syndrome.

This study was supported financially by Colorado State University.

Immunohistochemical evaluation of mononuclear infiltrates in canine lupoid onychodystrophy

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The purpose of this study was to characterize the immunophenotype of the inflammatory infiltrate in canine lupoid onychodystrophy and to determine if decalcification interferes with immunoreactivity. Claw biopsies of 14 dogs with lupoid onychodystrophy were stained with CD3, BLA36 and HM57 (CD97 α), MAC 387, lysozyme and MHC class II using an immunoperoxidase and avidin/biotin technique. Cells infiltrating the claw matrix were counted in two high-power fields; numbers were expressed as a percentage of the cellular infiltrate. The inflammatory infiltrate consisted predominantly of B and T cells; macrophages were typically only present in small numbers. There was no correlation between the predominant inflammatory cell and clinical parameters. CD3, BLA36, lysozyme and MHC II preserved significant antigenicity during formalin fixation and short decalcification, while staining for CD79A and particularly MAC 387 was less reliable. Prolonged decalcification of 7–14 days abolished immunostaining with all antibodies. Short decalcification periods allow immunostaining of samples with some antibodies (CD3, BLA36, lysozyme, MHC II), but not others (CD79A, MAC387), while prolonged decalcification prohibits immunostaining. The nature of the mononuclear infiltrate in lupoid onychodystrophy is not helpful clinically.

This study was funded by the American Academy of Veterinary Dermatology.

***Putrescentiae* and *Lepidoglyphus destructor* in normal dogs and dogs with atopic dermatitis**

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The purpose of this study was to evaluate the threshold concentration for the storage mites *Tyrophagus putrescentiae* and *Lepidoglyphus destructor* in intradermal testing of healthy and atopic dogs, and to compare the incidence of positive reactions in the two groups. Twenty-one healthy dogs and twenty-six atopic dogs were tested intradermally with *T. putrescentiae* and *L. destructor* extracts at 1000, 500, 250, 125, 63, 32 and 16 PNU mL⁻¹. Reactions were evaluated objectively and subjectively. A Mann–Whitney *U*-test was used to determine differences in grade of reaction to storage mites between healthy dogs and dogs with atopic dermatitis. Positive reactions to storage mite extract were most common at 1000 PNU mL⁻¹, with approximately one-third of normal and atopic dogs showing a positive reaction to *T. putrescentiae* and an even higher number to *L. destructor*. No positive reactions were seen in normal dogs at a concentration of 63 PNU mL⁻¹ of *L. destructor* extract and 32 PNU mL⁻¹ of *T. putrescentiae* extract. There was no significant difference in the incidence of positive reactions between normal versus atopic dogs for any of the *Tyrophagus* or *Lepidoglyphus* extract concentrations. Based on the findings in this study, the preliminary recommended concentrations for intradermal testing with *T. putrescentiae* and *L. destructor* extract are 16 and 32 PNU mL⁻¹, respectively.

Storage mite extracts were provided by Heska Corporation, Fort Collins, CO.

Importance of psychogenic dermatoses in dogs with pruritic behavior

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Canine psychodermatopathies are appreciated, but our knowledge of this field is extremely limited. We suspect that psychodermatopathies could be expanded to include dermatoses featuring pruritic behavior such as licking, scratching and biting. The purpose of this study was to evaluate these clinical findings of a series of suspect dogs. Seven dogs with psychogenic dermatoses featuring pruritic behavior are described. Differential diagnoses including infectious disease, allergy, congenital disorder, metabolic and neurological disturbances, and previous trauma at the lesions were all ruled out. All dogs had concurrent behavior problems or inductive life events, and both skin and behavior problems were improved with behavior modification with or without psychopharmacological therapy. The type and distribution of the skin lesions and incidental context of pruritic behavior were evaluated. Histopathological examination of the skin lesion was performed in five cases. In the seven dogs, characteristic skin lesions included: broken hairs ($n = 7$); and erythema and/or excoriation ($n = 3$); broken hairs located on the lateral thigh ($n = 4$), medial forearm ($n = 4$), flank ($n = 3$), and perioral area ($n = 3$); and erythema and/or excoriations located on the cheek ($n = 2$), perioral area ($n = 2$), periocular area ($n = 1$) and nails ($n = 1$). In all cases, the onset of pruritic behavior was associated with emotionally unstable situations, and did not appear during sleep or during periods of focused attention. Histopathological findings were unremarkable in all cases. In conclusion, psychogenic disorders should be considered as a cause of dermatoses in dogs with pruritic behavior, and this disorder can be identified with clinical evaluation of skin lesions and the relationship between the behavior and its context.

Immunofluorescent determination of the isotype of serum antikeratinocyte autoantibodies in dogs with *Pemphigus foliaceus*

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In humans with pemphigus foliaceus (PF), pathogenic autoantibodies are suspected to be of IgG4 subclass. The goals of this study were to determine the isotypes of circulating autoantibodies in dogs with PF, and to assess whether serum autoantibody titers decreased during successful treatment outcome. Using an indirect immunofluorescence assay performed on neonatal mouse skin sections, circulating IgG autoantibodies were detected in 36 out of 44 dogs with PF (82%). Serum autoantibodies belonged predominantly to IgG4 (35/44; 80%) and IgG1 (30/44; 68%) subclasses. Circulating IgA and IgE autoantibodies were not found. Serum IgG autoantibodies targeted autoantigens in both superficial and/or basal epidermal layers. Remarkably, antikeratinocyte IgG autoantibodies were detected in 16 out of 20 normal dogs (80%), and these autoantibodies were of IgG1 (16/20, 80%) but rarely of IgG4 (2/20; 10%) isotypes. Antikeratinocyte IgG, IgG1 and IgG4 autoantibody serum titers were followed in six dogs with PF while the severity of their clinical signs decreased with immunosuppression. IgG, IgG1 and IgG4 autoantibody serum titers decreased during remission in four (67%), one (17%) and four (67%) dogs, respectively. Conversely, serum IgG, IgG1 and IgG4 autoantibody titers increased during remission in one (17%), one (17%) and no dogs, respectively. These observations suggest that, in dogs with PF, antikeratinocyte IgG4 autoantibodies could be relevant to the pathogenesis of the disease and that monitoring of IgG4 autoantibody titers could be useful for assessment of clinical outcome. Remarkably, the detection of circulating IgG1 antikeratinocyte autoantibodies in normal dogs is similar to the situation observed in geographical areas where human PF is endemic.

Immediate intradermal flea-antigen reactivity in dogs in a flea-scarce environment

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The aim of this study was to determine the prevalence of intradermal test (IDT) reactions to flea antigen in dogs in a flea-scarce environment. Seventy-three client-owned dogs without present or prior exposure to fleas were used. Forty dogs had a history of, and clinical signs compatible with, atopic dermatitis (AD), after exclusion of other causes of pruritus, and 33 dogs were healthy or had some disease other than AD. A questionnaire was used to collect information about previous ectoparasitic infestations. An aqueous extract of whole bodies of *Ctenocephalides* spp. (Nelco Laboratories Inc., NY, USA) was used for IDT at a concentration of 1:1000 w/v. None of the dogs had been treated with corticosteroids or antihistamines within the previous 3 months. A positive reaction was defined as a wheal that was equal to or larger in diameter than the average of the saline and histamine controls. Two of the dogs with clinically diagnosed AD and none of the nonatopic dogs had immediate skin-test reactions to flea antigen. The prevalence (2.7%) for positive IDT against flea extract found in this study is low compared with studies performed in a flea-rich environment. Immediate skin-test reactivity against flea antigen in clinically normal dogs in flea-endemic areas could represent subclinical hypersensitivity or true false-positive reactions (due to skin irritation). Thus, these findings indicate that false-positive reactions against whole-body flea antigen from Nelco in dogs not exposed to fleas are rare.

Canine epidermal nevus treated with cryotherapy

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Epidermal nevus is a hamartomatous disorder characterized by hyperplasia of epidermal structures in a circumscribed area of the skin. This condition has been well recognized in humans, but rarely reported in dogs. Surgical excision is the most reliable treatment, and systemic retinoids has been reported to be effective in dogs. We describe a dog with epidermal nevus treated with liquid nitrogen cryotherapy. A 5-month-old male Shetland sheepdog presented with symmetrical linear and verrucous papules and plaques with mild pruritus on the axillae, ventral trunk and perianal area. Skin lesions were noticed at 2 months of age, and the colour of lesions gradually changed from normal to brownish. The general condition of the dog was otherwise good. Complete blood counts and biochemical analysis showed no abnormalities, and skin biopsies revealed orthokeratotic hyperkeratosis and papillomatosis. Based on these findings, the dog was diagnosed with epidermal nevus and treated with liquid nitrogen cryotherapy applied with a cotton applicator stick. Five days after the treatment, the lesions began slough with mild pigmentation developing at the sites. There was no recurrence of the lesions 3 months after treatment. It appears that liquid nitrogen cryotherapy could be considered a useful therapeutic option to control this disease in dogs.

Evaluation of canine ACTH stimulation test with direct enzyme immunoassay for serum cortisol in Taiwan

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In this study, we evaluated cortisol concentrations from 53 healthy dogs in response to ACTH stimulation test. The concentrations of serum cortisol were directly determined by a competitive enzyme immunoassay (EIA) with a 10^4 -fold diluted self-made monoclonal antibody. The sensitivity of EIA was 0.1 ng mL^{-1} with intra-assay and interassay coefficients of variation of 8.28 ± 0.09 and $14.60 \pm 0.28\%$, respectively. The recovery rates for 1, 5 and 10 ng mL^{-1} of cortisol in dog serum were 77.8–128.7% with direct detection. The parallelism test revealed its reliability to detect directly the cortisol concentration in canine serum without extraction. In general, this EIA showed low crossreactivities with a variety of steroids. In the ACTH stimulation tests, we gave each dog $250 \mu\text{g}$ ACTH (Cortrosyn® Organon) intramuscularly and collected blood samples before and 1 h after administration of ACTH. The cortisol concentrations from 53 healthy dogs were 0.85 – 14.63 ng mL^{-1} pre- and 3.06 – 37.22 ng mL^{-1} post-test, respectively. Sex and age did not significantly affect the cortisol concentrations in response to ACTH stimulation, but the large breeds (body weight $> 25 \text{ kg}$; most of them were German shepherd dogs) showed lower cortisol responses than the smaller breeds. Thus, the results of this test suggested that body weight and breed might play an important role in adrenal dynamic tests.