PREVALENCE OF CANINE ATOPIC DERMATITIS AND DISTRIBUTION OF CAUSATIVE ALLERGENS IN BANGKOK

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ความชุกของ โรคภูมิแพ้ผิวหนังจากสิ่งแวดล้อมในสุนัข และการกระจายตัวของสารก่อภูมิแพ้ ในกรุงเทพมหานคร

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โสภณ สรสนิท : ความชุกของ โรคภูมิแพ้ผิวหนังจากสิ่งแวดล้อมในสุนัข และการกระจายตัวของสารก่อภูมิแพ้ ในกรุงเทพมหานคร. (PREVALENCE OF CANINE ATOPIC DERMATITIS AND DISTRIBUTION OF CAUSATIVE ALLERGENS IN BANGKOK) อ.ที่ปรึกษาหลัก : รศ. สพ.ญ.ดร.รสมา ภู่สุนทรธรรม

โรคภูมิแพ้ผิวหนังจากสิ่งแวดล้อมในสุนัข (Canine atopic dermatitis) เป็นโรคภูมิแพ้ผิวหนังที่มีสาเหตุจากปฏิกิริยาภูมิไวเกินของร่างกายสุนัขต่อสารก่อภูมิแพ้ที่อยู่ในสิ่งแวดล้อม การวินิจฉัยโรคภูมิแพ้จากสิ่งแวดล้อมนั้นทำได้โดย การนำข้อมูลจาก ประวัติ การตรวจร่างกาย และ การวินิจฉัยแยกโรคผิวหนังอื่นที่ก่อให้เกิดอาการคันออก การตรวจหาสารก่อภูมิแพ้ที่จำเพาะต่ออิมมูโนโกลบูลินอีด้วยวิธีทางวิทยาเซรุ่ม (Allergen-specific IgE serology: ASIS) เป็นวิธีหนึ่งในการตรวจสารก่อภูมิแพ้จากสิ่งแวดล้อมในทางสัตวแพทย์ จุดประสงค์ของการศึกษานี้ เป็นการศึกษาความชุกของโรคภูมิแพ้ผิวหนังจากสิ่งแวดล้อม และ การกระจายตัวของสารก่อภูมิแพ้ ด้วยการตรวจสารก่อภูมิแพ้ที่จำเพาะต่ออิมมูโนโกลบูลินอี ด้วยวิธีทางวิทยาเซรุ่มในเขตกรุงเทพมหานคร การศึกษาแบ่งออกเป็นสองส่วน การศึกษาแรกเป็นการศึกษาในสุนัข 383 ตัวที่ถูกวินิจฉัยว่าเป็นโรคภูมิแพ้จากสิ่งแวดล้อม ด้วยสัตวแพทย์ในคลินิกผิวหนัง โรงพยาบาลสัตว์เล็ก คณะสัตวแพทย์ศาสตร์ จุฬาลงกรณ์มหาวิทยาลัย ระหว่าง ปีคริสตศักราช 2015 ถึง 2019 โดยเป็นการเก็บข้อมูลจาก เพศ พันธุ์ การกระจายตัวของวิการ สิ่งแวดล้อมที่อาศัย และ โรคแทรกซ้อนที่เกี่ยวข้องกับภาวะภูมิแพ้ สำหรับการศึกษาที่สองเป็นการเก็บข้อมูลจากสุนัขที่เป็นโรคภูมิแพ้จากสิ่งแวดล้อม 28 ตัว ที่ได้รับการการตรวจสารก่อภูมิแพ้ที่จำเพาะต่ออิมมูโนโกลบูลินอี ด้วยวิธีทางด้วยวิธีทางวิทยาเซรุ่ม Canine ALLERCEPT® ของบริษัท HESKA โดยสารก่อภูมิแพ้แบ่งออกเป็น 6 กลุ่มได้แก่ ยีสต์ น้ำลายหมัด ไรฝุ่น หญ้า วัชพืช และ ต้นไม้ ผลการศึกษาพบว่า ความชุกของโรคภูมิแพ้ผิวหนังในสุนัขเท่ากับร้อยละ 1.16 และการกระจายตัวของสารก่อภูมิแพ้ในสุนัขที่เป็นโรคภูมิผิวหนังจากสิ่งแวดล้อม ได้แก่ วัชพืช หญ้า ไรฝุ่น หญ้า วัชพืช ต้นไม้ ยีสต์ และ น้ำลายหมัด โดยมีร้อยละ 75.00 62.14 47.45 46.43 39.29 และ 21.43 ตามลำดับ จากการศึกษาอาจแนะนำได้ว่า โรคภูมิแพ้ผิวหนังจากสิ่งแวดล้อมในสุนัข เป็นโรคพื้นฐานและสามารถพบได้ในคลินิกปฏิบัติ การวินิจฉัยโรค มีความซับซ้อน แต่ การกระจายของวิการและการมีโรคแทรกซ้อน คือกุญแจของการวินิจฉัย สำหรับ การตรวจสารก่อภูมิแพ้ที่จำเพาะต่ออิมมูโนโกลบูลินอี ด้วยวิธีทางวิทยาเซรุ่ม มีประโยชน์มากในการระบุชนิดของสารก่อภูมิแพ้ และสามารถประยุกต์ใช้ในการจัดการความคัน และ การทำวัคซีนภูมิแพ้

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Canine atopic dermatitis (CAD) is a pruritic allergic skin disease caused by an overreaction to environmental allergens. The diagnosis of CAD is made on the basis of the patient's history, physical examination, and exclusion of other pruritic skin diseases. Allergen-specific IgE serology (ASIS) is one of the most frequently recommended tests in veterinary dermatology for identifying causative allergens. The purpose of this study was to determine the prevalence of CAD and the types of allergens that cause it in atopic dogs using ASIS in the Bangkok metropolitan area. The study was divided into two sections. Between 2015 and 2019, 383 dogs were diagnosed with CAD by a veterinarian at the Small Animal Hospital, Faculty of Veterinary Science, Chulalongkorn University. We collected data on the sexes, breeds, distribution of lesions, housing environment, and concurrent disease in CAD dogs. Among these dogs, 28 CAD dogs were tested using the HESKA veterinary diagnostic laboratory's Canine ALLERCEPT® (environmental panel). Canine ALLERCEPT® panel results were classified into six allergen groups (Yeast, Flea saliva, House dust mite, Grass, Weed, and Tree). The prevalence of canine atopic dermatitis was found to be 1.16 percent in this study. The allergens responsible for CAD were house dust mite (75.00%), grass (62.14%), tree (46.43%), weed (47.45%), yeast (39.39%) and flea saliva (21.43%). The findings indicate that CAD is the most prevalent skin disease and is frequently encountered in general practice. The diagnosis of canine atopic dermatitis is quite complicated, but lesion distribution and recurrent concurrent disease may provide diagnostic clues. The ASIS is the most effective method for identifying allergens that cause pruritus and plan to do immunotherapy.

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CHAPTER 1 INTRODUCTION

1. Importance and Rationale

Canine atopic dermatitis (CAD) is one of the most prevalent allergic skin diseases. In veterinary practice, there are four types of allergy: flea allergic dermatitis (FAD), contact dermatitis, food allergy, and atopic dermatitis (Hensel et al., 2015; Saridomichelakis and Olivry, 2016). CAD is caused by an immune hypersensitivity reaction triggered by environmental allergens (DeBoer and Marsella, 2001; Santoro et al., 2015). It is primarily triggered by an IgE-antibody reaction. Atopic dogs exhibit clinical signs such as pruritus, biting, licking, and scratching, as well as erythematous dermatitis on the face, ear pinnae, ventrum, axillae, inguinal area, perineal area, and distal extremities (Hensel et al., 2015). Pruritus persists for an extended period of time and is frequently associated with chronic inflammation and secondary infection (Bizikova et al., 2015; Hensel et al., 2015). Inflammation, pruritus, and infection are all part of a vicious cycle that has an effect on dogs, their owners, and veterinarians. It has been reported that CAD has a negative effect on the owner's quality of life, with the primary burden being the high cost of treatment and long-term therapy (Favrot et al., 2010a). CAD is a multifactorial skin disease that is difficult to diagnose and manage effectively in veterinary practice due to its severe pruritus and recurrent infection.

Numerous studies have been conducted to determine the prevalence of canine atopic dermatitis in various regions. In the United States of America, CAD was found in 30% of dogs with skin disease (Hillier and Griffin, 2001). Atopic dogs accounted for 40% of dermatological problem dogs in the United Kingdom (Hill et al., 2006). Kuribayashi's study reported that canine atopic dermatitis accounted for 50% of skin disease's dog in Japan (Kuribayashi et al., 2018). However, based on the results of intradermal skin testing, there is only one report on the prevalence of canine atopic dermatitis in Thailand (Chanthick et al., 2008). The purpose of this study was to determine the prevalence of atopic dermatitis in allergic dogs.

The diagnosis of CAD is made after a thorough history is taken, a physical examination is performed, and other pruritic skin diseases are ruled out. Before

diagnosing atopic dermatitis as a parasitic infestation, bacterial or fungal skin infection, or other allergic skin diseases, such as food allergy, the veterinarian must rule out other possible causes (Hensel et al., 2015). Subsequently, causative allergen identification may be performed to identify potentially problematic causes, allowing veterinarians to develop an individualized management plan for atopic dogs. In veterinary dermatology, two methods of identification are recommended: intradermal skin testing (IDST) and allergen-specific IgE serology (ASIS) (DeBoer and Marsella, 2001; Hensel et al., 2015). IDST is widely regarded as the gold standard for allergen detection. However, its application has been restricted due to a lack of practicality. Sedation and hair clipping are required in the majority of cases, and the procedure is also quite expensive to maintain. ASIS has grown in popularity over the last decade due to its simplicity and small amount of serum requirement. In theory, the ELISA technique is used to determine the concentration of specific IgE in serum samples. Numerous previous studies concluded that ASIS can be used in practice to diagnose atopic dermatitis as a screening test and allergen identification (Lian and Halliwell, 1998; Tarpataki et al., 2006; Lauber et al., 2012; Kang et al., 2014; Mueller et al., 2016). Several studies on the prevalence of allergens have been conducted in many countries, but only one has been conducted in Thailand since 2008 (Chanthick et al., 2008). The purpose of this study was to use ASIS to determine the distribution of causative allergens in the Bangkok area.

2. Objectives

1. To study the prevalence of canine atopic dermatitis (CAD) of allergic dogs in the Bangkok area.
2. To study types of allergens in atopic dogs by using allergen specific IgE serology testing in the Bangkok area.

3.Hypothesis

The prevalence of atopic dermatitis in canine allergic skin diseases is over 30% with a high variation of causative allergens.

CHAPTER 2 LITERATURE REVIEWS

1. Canine atopic dermatitis

1.1 Prevalence of canine atopic dermatitis

Atopic dermatitis in dogs has been reported to be prevalent in several countries worldwide. Around 30% of skin cases in the United States were CAD (Hillier and Griffin, 2001). In the United Kingdom, 21.4 percent of dogs suffered from dermatological problems, which 30-40 percent of those were diagnosed as atopic dermatitis (Hill et al., 2006). In Asia, a study conducted in Japan discovered that CAD was responsible for 50% of skin disease in dogs (Kuribayashi et al., 2018).

1.2 Clinical manifestations, genetic and breed-specific predisposition

Atopic dermatitis is one of the most frequently encountered allergic skin conditions in veterinary medicine. Pruritus or itching (scathing, rubbing, licking, biting, shaking head, and other signs) are the clinical manifestations of atopic dermatitis (Hensel et al., 2015; Saridomichelakis and Olivry, 2016). Atopic dogs frequently develop erythematous dermatitis, which typically affects the face, ear pinnae, ventrum, axillae, inguinal area, perineal area, and distal extremities (Hensel et al., 2015; Santoro et al., 2015). Secondary skin lesions such as alopecia, lichenification, hyperpigmentation, excoriation, crusting, and seborrhea may manifest as a result of self-trauma, chronic inflammation, and secondary infection in chronic cases. According to a previous study, the average age of onset of canine atopic dermatitis is less than three years. There has also been evidence of breed predisposition to atopic dermatitis in dogs. Boxers, German shepherds, Golden retrievers, Shar-peis, Dalmatians, Labrador retrievers, French bulldogs, West Highland white terriers, and Jack Russell terriers are affected at small proportion (Bizikova et al., 2015). The majority of atopic dogs have been reported to live primarily indoors, where they are more susceptible to exposure to house dust mites. The clinical signs of canine atopic dermatitis reflect a complex interaction between genetic characteristics and the environment. The pruritic lesions and responses to treatment can vary among specifically individual dogs of the same breed. The genomic studies reported that the

lack of filaggrin might be one of the reasons for the phenotype among breeds (Barros Roque et al., 2009; Wood et al., 2009). Canine atopic dermatitis is clearly a heritable disease; however, the genetic studies show complex and varies among breeds and geographic regions (Jaeger et al., 2010).

1.3 Pathogenesis of canine atopic dermatitis

Canine atopic dermatitis is associated with the production of IgE antibody against environmental antigen. Immunity response triggers disease outcome by developing hypersensitivity type 1 (Chanthick et al., 2008; Hensel et al., 2015; Santoro et al., 2015; Nuttall et al., 2019). The pathogenesis of canine atopic dermatitis is shown in figure 1.

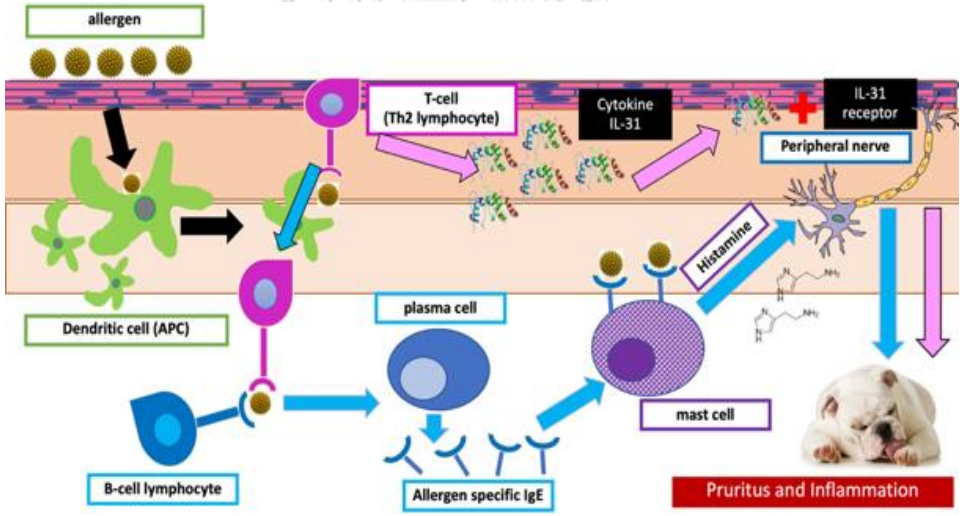

Figure 1 Pathogenesis of canine atopic dermatitis.

When the allergen exposes on the dog's skin. The immunity responses to allergen by activating dendrite cells, macrophages, and B-cell lymphocytes that are antigen presenting cells (APCs). APCs present allergens to T-lymphocytes in particular (T-helper 2 cell lymphocytes), triggering the release of various cytokines and chemokines into the bloodstream, including interleukins (ILs). IL-2,4,6,13, and 31 all play a significant role in canine atopic dermatitis. IL-31 in blood circulation binds its

receptor on peripheral nerve ending and stimuli inflammation and pruritus. Alternatively, Th-2 lymphocytes present allergen to B-cell lymphocytes via other pathways. B cells differentiate into plasma cells. They produce IgE specific to allergens. After allergens re-expose on the skin. Allergens will cross-link with allergen-specific IgE on mast cells, activating mast cell degranulation and histamine secretion. Histamine stimulates itching and inflammation by binding to their receptors on keratinocytes and nerve endings.

2. Allergens

Allergens are immunogens capable of eliciting an immune response. Allergens have a molecular weight of between 10 and 100 kDa (Mueller et al., 2016). Allergens are classified into five categories in veterinary dermatology: house dust mite, pollen and tree pollen, insect, mold, and indoor materials (Lauber et al., 2012; Kang et al., 2014). Exposure of allergens in atopic dogs varies according to their habitat, lifestyle, and geographic location, among other factors. Many countries, including Thailand, have reported on the prevalence of causative allergens in CAD dogs. The positive IDST results indicated that *Dermatophagoides farinae* was the most common allergen in numerous studies (Zur et al., 2002; Tarpataki et al., 2006; Chanthick et al., 2008; Mueller et al., 2016). Lauber et al. (2012) reported a prevalence of D. *farinae* of 78.9 percent , while Tarpataki et al. (2006) reported a prevalence of 52.9 percent. Kang et al. (2014) estimated the prevalence of D. *farinae* to be 61.4 percent using ASIS. Numerous studies indicate that the house dust mite group has the highest positive prevalence. Allergen distribution is determined by geography, climate, and season. By using IDST, the allergens D. *farinae* (74.56 percent), *Dermatophagoides pteronyssinus* (53.51 percent), house dust (26.32 percent), American cockroach (*Periplaneta americana*) (23.68 percent), Para glass (21.93 percent), and mixed ants (20.18 percent) were found to be the most prevalent in Bangkok, Thailand (Chanthick et al., 2008).

3. Diagnosis of canine atopic dermatitis

To accurately diagnose canine atopic dermatitis (CAD), it is critical to rule out other pruritic skin diseases. Essentially, CAD is diagnosed after other skin infections and allergic skin diseases are ruled out (Hensel et al., 2015). Pruritus is the primary clinical manifestation of atopic dermatitis and can be caused by a variety of different factors. The initial diagnostic step is to exclude the infection caused by parasites (tick and flea, scabies, demodicosis, cheyletiellosis, and otoacariasis), bacteria (*Staphylococcal* pyoderma), or fungi (dermatophytosis and *Malassezia* dermatitis). The following step is to exclude other allergic skin diseases such as flea allergic dermatitis (FAD), contact dermatitis, and food allergy (Hensel et al., 2015). FAD can be ruled out based on the appearance of dorso-lumbar lesions and intensive flea control. The flea control program may vary according to the severity of the infestation, the environment, the animals' lifestyles, and the veterinarian's decision (Wilkerson et al., 2004). Avoidance and control of the living zone are critical steps in managing FAD because they minimize flea and flea saliva exposure. Contact dermatitis is another type of allergic dermatitis that commonly affects dogs and cats. It is a condition that is brought on by exposure to a chemical toxin or other substances. Contact dermatitis typically manifests itself in hairless areas such as the abdominal and inguinal areas (Hensel et al., 2015). The patch test is a diagnostic tool used to determine the causative substrate of disease. Before diagnosing atopic dermatitis, veterinarians must rule out FAD and contact dermatitis. Food allergy is a type of allergic skin disease that is triggered by food components (Hensel et al., 2015; Gedon and Mueller, 2018). Proteins and carbohydrates in food are frequently reported to be allergens that cause food hypersensitivity reactions (Mueller and Unterer, 2018). Food allergy presents clinically similarly to AD. However, gastrointestinal symptoms such as vomiting and diarrhea may occur in 30% of cases of food allergy. Food elimination diets are the gold standard for diagnosing food allergy. Two types of food are recommended for restriction diet trials to treat and diagnose food allergy: novel and hydrolyzed protein diets. In theory, novel protein refers to a new source of protein that has never been given to dogs, whereas

hydrolyzed protein refers to small molecular protein that has been hydrolyzed into a smaller size via nutritional technique (Mueller and Unterer, 2018). In theory, hydrolyzed proteins are incapable of eliciting an immune response or initiating a hypersensitivity reaction. If the dog improves clinically after eight weeks of restrictive diet testing, it will be diagnosed with food allergy. However, if allergic signs or lesions persist, atopic dermatitis will be diagnosed. Occasionally, veterinarians will use Favrot's criteria to bolster the likelihood of a patient having CAD (Hensel et al., 2015). The sensitivity and specificity of this test are 85.4 and 79.1 percent, respectively. If the dog meets five of the eight criteria, it will be diagnosed in canine atopic dermatitis. Favrot's criteria include age of onset < 3 years, pruritus that is corticosteroid-responsive, chronic or recurrent yeast infections, affected front feet and ear pinnae, and an unaffected dorso-lumbar region. Favrot's criteria are as follows as indicated in table 1. After completing the diagnosis of atopic dermatitis, veterinarians may perform allergen identification using either IDST or ASIS (Hillier and Griffin, 2001).

Table 1 Favrot's criteria

	Use	Reliability
Set 1: 1. Age at onset < 3 years 2. Mostly indoor 3.Corticosteroid-responsive pruritus 4. Chronic or recurrent yeast infections 5. Affected front feet 6. Affected ear pinnae 7. Non-affected ear margins 8.Non-affected dorso-lumbar area	- Use for clinical studies and adapt required criteria based on the goal of the study. - If higher specificity is required, 6 criteria should be fulfilled (e.g. drug trials with potential side effects) - If higher sensitivity is required, 5 criteria should be fulfilled (e.g., epidemiological studies)	5 criteria: Sensitivity: 85.4 % Specificity: 79.1 % 6 criteria: Sensitivity: 58.2 % Specificity: 88.5 %
Set 2: 1. Age at onset < 3 years 2. Mostly indoor 3. "Alesional" pruritus onset 4. Affected front feet 5. Affected ear pinnae 6. Non-affected ear margins 7. Non-affected dorso-lumbar area	- Use to evaluate the probability of the diagnosis of CAD - 5 criteria should be fulfilled - Do not use alone for diagnosis of canine AD, and rule out resembling diseases	5 criteria: Sensitivity: 77.2 % Specificity: 83.0 % 6 criteria: Sensitivity: 42.0% Specificity: 93.7 %

4. Allergen identification

Allergen identification is a technique to determine the allergens that can potentially cause CAD. There are two types of allergen identification techniques currently available: intradermal skin testing (IDST) and allergen-specific IgE serology testing (ASIS) (Lian and Halliwell, 1998; Tarpataki et al., 2006; Lauber et al., 2012; Kang et al., 2014; Mueller et al., 2016). IDST is the gold standard for allergen identification because it detects allergic reactions on the dog's skin caused by mast cell degranulation. It occurs as a result of a direct interaction between allergen and allergen-specific IgE on mast cells. ASIS, on the other hand, is a deceptive technique. It uses an enzyme-linked immunosorbent assay to determine the concentration of allergen-specific IgE in the blood circulation (ELISA).

4.1 Intradermal skin testing (IDST)

The dog must be sedated prior to the IDST procedure. The dogs' lateral thoracic area is clipped and cleaned. The clipped area is approximately 15 cm x 20 cm in size. After that, using a permanent pen, create an injected point on the skin. With 27 gauge needles, approximately 0.05 ml of each allergen and the control are injected intradermally. The negative control is buffered saline, the allergen extracts' diluent. Histamine is used as a positive control (Chanthick et al., 2008). Within 15 minutes of injection, an allergic reaction will manifest. IDST is interpreted by comparing the size of the erythema and wheal to a positive and negative control.

4.2 Allergen specific IgE serology (ASIS)

For ASIS technique, enzyme-linked immunosorbent assay (ELISA) is used to detect concentration of IgE in canine serum (Tarpataki et al., 2006; Lauber et al., 2012; Kang et al., 2014). the popular test of ASIS is (ALLERCEPT^{TM} Detection System) (McCall et al., 2001).

Before performing the IDST and ASIS, dogs must be weaned from antipruritic medication. The optimal antihistamine (oral) and glucocorticoid (short-acting oral, topical, and otic) withdrawal times (days) in IDST are 7 and 14 days, respectively. While in the ASIS technique, only long-acting glucocorticoid withdrawal 28 days prior to blood collection is considered (Olivry and Saridomichelakis, 2013).

The difference between IDST and ASIS is based on techniques, procedures, and practicality in veterinary practice. IDST needs sedation, clipping, experience, and sophisticated process. In ASIS, only blood serum samples are used but samples needed to send to the laboratory. The comparison studies between IDST (gold standard) and ASIS found that sensitivity and specificity of IDST are 77.1% and 93.8% (Lauber et al., 2012). and the sensitivity and specificity of ASIS are 82.4% and 99.3% (Tarpataki et al., 2006). The comparison between IDST and ASIS are shown in table 2. The study showed that ASIS has high sensitivity and specificity and can be recommended for diagnosis of atopy in practice as a screening test and allergen identification for immunotherapy (Lian and Halliwell, 1998; Tarpataki et al., 2006; Lauber et al., 2012; Kang et al., 2014; Mueller et al., 2016).

Table 2 Comparison of intradermal skin testing and allergen specific serology (Miller et al., 2013)

Feature	Intradermal skin testing	Allergen specific serology
Detects reaginic antibody	Yes	Yes
Detects presence of reaginic antibody in serum	No	Yes
Detects presence of cutaneous mast cells with reaginic antibody present in them	Yes	No
Determines capability of inducing a cutaneous type 1 hypersensitivity reaction on exposure to antigen	Yes	No
Test results are inhibited by	Yes	Yes

glucocorticoids		
Test results are inhibited by antihistamines	Yes	Not investigated
Sensitivity (%)	70-90	70-100
Specificity (%)	>90	0-90 (depending on laboratory and technology used)
Risk to patient	Rare but possible anaphylaxis or sedative reaction	Only serum sample required
Availability	Limited to certain practices; often requires referral	Excellent
Cost per antigen tested	Inexpensive	Relatively expensive
Clinic overhead	Relatively high	Little to none
Ease and speed of performing test or obtaining sample	Relatively difficult, taking several hours	Easy, taking only minutes
Time to get results	Minutes	Days to weeks

The advantage of allergen identification in CAD is that it enables avoidance of allergens exposure and the administration of allergen-specific immunotherapy (ASIT). Avoidance is the process of preventing atopic dogs from coming into contact with allergens, thereby preventing an allergic reaction. As a result, pruritus severity and recurrent infection with bacteria and yeast can be decreased. ASIT management is a subset of CAD management. Immunotherapy can help to reduce or eliminate allergic symptoms without the use of drugs that may pose a health risk or have adverse effects.

5. Management of canine atopic dermatitis

Atopic dermatitis in dogs is a complicated condition to manage. In some studies, multimodal therapy is used to treat pruritus. The successful combination of an antipruritic or immunomodulator, a skin supplement, and a medical shampoo is critical. Acute and chronic flares are managed differently depending on the duration of therapy. There are three important considerations for managing acute flares of

canine atopic dermatitis (Hensel et al., 2015). First, identification of the underlying cause of the itch, avoidance of flare-up triggers, and management of secondary infection. Environmental aeroallergens (particularly house dust mites and pollens), dietary ingredients, and fleas or other ectoparasites all serve as flare factors. Flares occur when a dog is hypersensitive to any of the aforementioned triggers and the allergen load is high enough to cause flares (Hensel et al., 2015). Bacterial skin disease, Malassezia dermatitis, and otitis externa are common concurrent infections that can cause exacerbations in CAD-affected dogs. Infectious skin diseases, especially bacterial skin diseases, are typically treated with topical and/or systemic antimicrobials. Second, enhancements in skin and coat hygiene and maintenance (Hensel et al., 2015). A shampoo with essential fatty acids and/or antiseptic properties is essential for reducing pruritic symptoms. Bathing with a medical shampoo not only reduces skin inflammation and secondary infection, but also prevents allergens from accumulating on the surface of the skin. Finally, decreasing pruritus and skin lesions with antipruritic and immunomodulator medications are clinical keys of management (Hensel et al., 2015). In acute exacerbations, pharmacological agents include topical or oral glucocorticoids, oclacitinib, and lokivetab.

Glucocorticoids are potent anti-inflammatory agents that alleviate pruritic symptoms. Glucocorticoids have a complex mechanism of action. They exert anti-inflammatory and immunosuppressive effects via genomic and non-genomic mechanisms by inhibiting the synthesis of pro-inflammatory cytokines, chemokines, and leukotrienes (both of which are mediated by increased lipocortin-1 expression); enhancing the clearance of foreign antigens; decreasing the ability of dendritic cells to present antigen and activate T cells; suppressing the cellular immune response via inhibition of IL-12 synthesis, the suppression of IL (Bruet et al., 2022).

Acute CAD flares can be effectively treated with topical glucocorticoid sprays. The topical spray is indicated for the treatment of focal localized skin lesions and is intended to be used in the short term (Hensel et al., 2015). Oral glucocorticoids, such as prednisolone, are typically given at a dose of 0.5 to 1.0 mg/kg every 24 hours, in a single or divided dose, and are likely to improve clinical signs in dogs with severe

atopy (Hensel et al., 2015). Polydipsia (39.2 percent), polyuria (28.4 percent), vomiting (16.2 percent), and diarrhea (14.9 percent) were reported as adverse effects of oral glucocorticoids in dogs in one study (Elkholly et al., 2020). Their severity is proportional to the potency, dosage, and duration of the drug being administered. Long-acting injectable glucocorticoids are not recommended for the treatment of acute CAD flares (Hensel et al., 2015).

Oclacitinib is a targeted therapy that inhibits key pathways responsible for pruritus and inflammatory triggers in allergic dermatitis, including CAD. It inhibits Janus kinase 1 but has a negligible effect on Janus kinase 2 (Cosgrove et al., 2013). The Janus kinase 1 is involved in the signal transduction of pruritogenic cytokines implicated in atopic dermatitis, including interleukin (IL)-2, IL-4, IL-6, and IL-13. Janus kinases play a role in the signaling of IL-31 (Carmi-Levy et al., 2011; Cosgrove et al., 2013). Oclacitinib has been shown to significantly inhibit the function of the IL-31 cytokine in dogs, implying that it may significantly reduce pruritus. Oclacitinib can be prescribed at a dose of 0.4–0.6 mg/kg every 12 hours for up to 14 days in atopic dogs to rapidly reduce skin lesions and pruritus (Cosgrove et al., 2013). In dogs, short-term treatment with oclacitinib appears to be safe (Cosgrove et al., 2013).

Lokivetmab is a canine-specific monoclonal antibody (mAb) that specifically recognizes and neutralizes IL-31 (Souza et al., 2018). IL-31 is a cytokine involved in the pathogenesis of canine atopic dermatitis. It is associated with pruritic behavior in dogs (Carmi-Levy et al., 2011). Lokivetmab is approved in the United States of America for the treatment of canine atopic dermatitis at a dose of 2.0 mg/kg administered subcutaneously with an expected duration of efficacy of at least one month (Souza et al., 2018; Fleck et al., 2021). The advantages of lokivetmab include its rapid onset of action, low frequency of administration, lack of age restriction, and safety.

Similar to acute flare management, chronic flare management for canine atopic dermatitis is divided into three essential factors. First is identifying and avoiding flare-up risk factors. The recommendation includes using IDST or ASIS to

identify allergens that are responsible for allergic reactions. The second is the improvement of the barrier function of the skin. The skin barrier loses its normal structure and function in an atopic dog (Cornegliani et al., 2011). Because stratum corneum intercellular lipids lack ultrastructure, transepidermal water loss occurs and secondary infection is easily acquired (Cornegliani et al., 2011). Oral or topical supplementation with essential fatty acids can improve the structure and function of the skin. A lipid complex containing ceramides, cholesterol, and essential fatty acids applied topically mimics the intercellular lipids (Hensel et al., 2015). Reduced itching and skin lesions as a result of antipruritic medication is the third step in managing chronic flares. As an adjunctive therapy for inflammatory disorders such as atopic dermatitis, dermatologists typically recommend 50 mg/kg/day of eicosapentanoic acid (EPA) and 35 mg/kg/day of docosapentanoic acid (DHA) (Mueller et al., 2005; Koch et al., 2012). The dose is then determined based on the amount of EPA and DHA in a specific commercial product. Clinical improvement may require eight to twelve weeks (Mueller et al., 2005). Additional antipruritic drugs include glucocorticoids, antihistamines, and calcineurin inhibitors.

Antihistamines of type 1 (H1 histamine receptor antagonists) can help reduce allergic reactions in patients with coronary artery disease. The H 1 receptor, which is found in blood vessels, smooth muscle of the airway and gastrointestinal tract, the heart, and the central nervous system, is primarily associated with the pruritus, pain, and increased vascular permeability associated with histamine (Koch et al., 2012). Antihistamines work by inhibiting the release of inflammatory mediators by mast cells and basophils; decreasing the migration, accumulation, or activation of inflammatory cells; and decreasing the expression of adhesion molecules (Saridomichelakis and Olivry, 2016; Bruet et al., 2022).

Cyclosporin, a calcineurin inhibitor, is one of the immunomodulators used to treat canine atopic dermatitis. Cyclosporin exerts its action by inhibiting cytoplasmic calcineurin phosphatase activity (Allenspach et al., 2006). It inhibits the T-cell signal transduction pathway by binding to a receptor on calcineurin. As a result, cyclosporin inhibits T-cell cytokine gene transcription, particularly IL-2, which is required for T-cell proliferation (Koch et al., 2012). Cyclosporine is a potent inhibitor of cell-mediated

immunity but has a lesser effect on humoral immunity. It inhibits the proliferation of cytotoxic T cells, antibody production by helper T cell-dependent B cells, activated T cell proliferation, and the activation of mononuclear phagocytes and helper T cells selectively (Koch et al., 2012). Oral cyclosporin at a dose of 5 mg/kg every 24 hours should be given until pruritic symptoms are completely controlled, which should take 4 to 6 weeks (Koch et al., 2012; Hensel et al., 2015). Following that, the dose required to maintain remission should be tapered (e.g., from daily to every other day, then twice weekly, or by reducing the daily dose) (Hensel et al., 2015).

CHAPTER 3 MATERIALS AND METHODS

This was a cross-sectional study conducted from 2015 to 2019. This study consisted of two phases. The study's initial phase examined evaluated the prevalence of canine atopic dermatitis in allergic dogs in the Bangkok area. The purpose of phase two was to characterize allergens in atopic dogs in the Bangkok area using allergen-specific IgE serology (ASIS).

Phase 1: Prevalence of canine atopic dermatitis

Sample size calculation

A sample size of this study was calculated by using unknown population size to estimate population proportion (Cochran and Carroll, 1953). The formula and results were shown in table 3.

Table 3 Sample size calculation

$n = [Z^2 P(1-P)]/e^2$	n = Sample size
$n = [1.96^2 (0.5)(0.5)]/0.05^2$	Z = Confidence level (95%) = 1.96
n = 384	P = Proportion (unknown) = 0.5
	e = Error (95%CI) = 0.05

The study population was 33,111 visiting dogs at the Small Animal Hospital, Faculty of Veterinary Science, Chulalongkorn University between 2015-2019. All cases were divided into two categories: those involving dogs with skin diseases and those involving dogs with other diseases. This study found skin diseases in 3,299 dogs. Then, skin diseases were classified as either allergic skin disease (946 dogs) or other skin diseases (2,353 dogs). To ensure a reliable and systematic diagnosis, allergic dermatitis was diagnosed by veterinary dermatologists. Initially, infections or infestations of the skin, such as parasites, bacteria, or fungi, were ruled out. Intensive parasitic control was used to rule out flea allergic dermatitis. As a result, all dogs were put on a food elimination diet to rule out food allergy. Food trials lasted

between 8 and 12 weeks, depending on clinical signs, disease severity, and owner compliance. CAD was diagnosed in dogs with persistent pruritus following a successful food trial. Finally, patient data on sex, breed, lesion distribution, housing environment, and concurrent disease were entered into the computerized program for CAD dogs.

The data of atopic dogs was collected form dermatology clinic at the Small Animal Hospital, Faculty of Veterinary Science, Chulalongkorn University. 383 atopic dogs were included from 33,111 dog patients at the Faculty of Veterinary Science's Small Animal Hospital. All dogs were privately owned and had been diagnosed with CAD based on clinical history, signs, and diagnostic criteria by dermatologists.

Inclusion criteria

Dogs visited only at dermatology clinic at the Small Animal Hospital, Faculty of Veterinary Science, Chulalongkorn University between 2015-2019 with the pruritic sign were included. All cases were diagnosed with allergic skin diseases and canine atopic dermatitis.

Exclusion criteria

Dogs with other pruritic skin diseases including infectious skin’s disease (Parasite, Bacteria, Fungi) and other allergic skin diseases (Flea allergic dermatitis, Food allergy, contact dermatitis) were excluded.

Phase 2: Distribution of causative allergens

The study collected data of 28 CAD dogs that were performed allergen identification to identify causative allergens. The data of these dogs were the results from Allergen specific IgE serology testing (ASIS): Canine ALLERCEPT® (environmental panel) from the veterinary diagnostic laboratory (HESKA). Results from Canine ALLERCEPT®panels were divided into six allergen groups (yeast, flea saliva, house dust mite, grass, weed, and tree). The result of them were separated in four levels consist of negative, level 1 (+), level 2 (++), and level 3 (+++) that are shown in table 4.

Table 4 HESKA laboratory results

Level	Result	HERBU	Interpretation
	negative	≤ 10	No allergen-specific IgE detected
Level 1	+	10 to 25	Allergen-specific IgE detected. Allergens in this category should be considered for immunotherapy if exposure is consistent with patient history.
Level 2	++	26 to 50	High levels of allergen-specific IgE detected. Allergens in this category should be considered for immunotherapy if exposure is consistent with patient history.
Level 3	+++	51 to >600	Very high levels of Allergen-specific IgE detected. Allergens in this category should be considered for immunotherapy if exposure is consistent with patient history.

Blood collection

Blood was drawn from cephalic vein, allowed to clot then centrifuged at 2,000 g for 15 minutes at room temperature. Serum samples were harvested and frozen at -20 °C for further diagnosis. Serum samples from 28 dogs were sent to distributor center of HESKA laboratory Bangkok, Thailand.

Statistical analysis

The data were evaluated using SPSS program version 22.0. Descriptive analysis of prevalence of canine atopic dermatitis, sex, breeds, distribution of lesion, housing environment, concurrent disease, and type of allergens were reported as percentage.

CHAPTER 4 RESULTS

Phase 1: Prevalence of canine atopic dermatitis

From 33,111 dogs visited the Small Animal Hospital, Faculty of Veterinary Science, Chulalongkorn University 2015-2019. The result revealed that 3,299 canines had skin diseases. In 3,299 skin disease's dogs were diagnosed with allergic skin disease 946 dogs (28.68 percent). Three hundred and eighty-three dogs with allergic skin disease were diagnosed with CAD. During 2015-2019, 1.16 percent of all visiting dogs at this hospital were diagnosed with CAD. In contrast, the prevalence of CAD in dogs with skin disease was 11.61 percent and in dogs with allergies it was 40.48 percent.

1. Breed

The top ten of breeds in visiting dogs and skin disease dogs were presented in table 5.

Table 5 Breeds of visiting dogs and skin disease dogs

Breed	Number of visiting dogs	Percentage	Breed	Number of skin diseases's dogs	Percentage
Mixed breed	8,490	25.64	Mixed breed	613	18.58
Pomeranian	3,721	11.24	Shih Tzu	450	13.64
Chihuahua	3,438	10.38	Pomeranian	314	9.52
Shih Tzu	3,113	9.40	Chihuahua	252	7.64
Poodle	2,512	7.59	Poodle	247	7.49
Golden Retriever	829	2.50	Beagle	153	4.64
Beagle	748	2.26	French Bulldog	133	4.03
French Bulldog	743	2.24	Golden Retriever	92	2.79
Siberian Husky	733	2.21	Pug	87	2.64

Labrador Retriever	709	2.14	Labrador Retriever	85	2.58
Other	8075	24.39	Other	873	26.46
Total	33,111	100.00	total	3299	100.00

Breeds of allergic dogs represented in this study included Shih Tzu (209/946; 22.09%), Mixed breed (160/946; 16.91%), Beagle (75/946; 7.93%), Poodle (74/946; 7.82%), Chihuahua (57/946; 6.03%), French Bulldog (47/946; 4.97%), Golden Retriever (36/946; 3.81%), Labrador Retriever (33/946; 3.49%), Pug (31/946; 3.28%), Yorkshire terrier (24/946; 2.54%) and Others.

Breeds of CAD dogs represented in this study included Shih Tzu (95/383; 24.80%), Mixed breed (67/383; 17.59%), Beagle (39/383; 10.18%), Poodle (30/383; 7.83%), French Bulldog (21/383; 5.48%), Chihuahua (19/383; 4.96%), Golden Retriever (14/383; 3.66%), Labrador Retriever (13/383; 3.39%), Pug (12/383; 3.13%), West Highland White Terrier (8/383; 2.09%) York Shire Terrier (8/383; 2.09%), Maltese (7/383; 1.83%), Pomeranian (6/383; 1.57%), German Shepherd (4/383; 1.04%), Chow Chow (3/383; 0.78%), Cocker Spaniel 3/383; 0.78%), Miniature Pinscher (3/383; 0.78%), Terrier (3/383; 0.78%), Thai Bangkaew (3/383; 0.78%), Welsh Corgi (3/383; 0.78%), American Pit Bull Terrier (2/383; 0.52%), Bulldog (2/383; 0.52%), Cavalier King Charles Spaniel (2/383; 0.52%), Schnauzer (2/383; 0.52%), Shiba Inu (2/383; 0.52%), Thai Ridgeback (2/383; 0.52%), Akita (1/383; 0.26%), Boston Terrier (1/383; 0.26%), Boxer (1/383; 0.26%), Bull Terrier (1/383; 0.26%), Jack Russell Terrier(1/383; 0.26%), Pekingese(1/383; 0.26%), Rottweiler (1/383; 0.26%), Samoyed (1/383; 0.26%), Shar-Pei (1/383; 0.26%), and Siberian Husky (1/383; 0.26%) (Table 6).

Table 6 Breed of allergic dogs, CAD dogs and prevalence of CAD dogs in allergic dogs

Breed	Number of CAD dogs (n_1)	Percentage	Number of allergic dogs (n_2)	Prevalence of CAD dogs in allergic dogs each breed (n_1/ n_2*100)
Shih Tzu	95	24.80	209	45.45
Mixed breed	67	17.49	160	41.88
Beagle	39	10.18	75	52.00
Poodle	30	7.83	74	40.54
French Bulldog	21	5.48	47	44.68
Chihuahua	19	4.96	57	33.33
Golden Retriever	14	3.66	36	38.89
Labrador Retriever	13	3.39	33	39.39
Pug	12	3.13	31	38.71
Yorkshire Terrier	8	2.09	24	33.33
West Highland White Terrier	8	2.09	17	47.06
Maltese	7	1.83	14	50.00
Pomeranian	6	1.57	21	28.57
German Shepherd	4	1.04	9	44.44
Chow Chow	3	0.78	4	75.00
Cocker Spaniel	3	0.78	12	25.00
Miniature Pinscher	3	0.78	6	50.00
Terrier	3	0.78	4	75.00
Thai Bangkaew	3	0.78	15	20.00
Welsh Corgi	3	0.78	9	33.33
American Pit Bull Terrier	2	0.52	8	25.00
Bulldog	2	0.52	8	25.00
Cavalier King Charles Spaniel	2	0.52	4	50.00
Schnauzer	2	0.52	8	25.00
Shiba Inu	2	0.52	3	66.67
Thai Ridgeback	2	0.52	7	28.57
Akita	1	0.26	1	100.00
Boston Terrier	1	0.26	4	25.00
Boxer	1	0.26	3	33.33
Bull Terrier	1	0.26	7	14.29
Jack Russell Terrier	1	0.26	12	8.33
Pekingese	1	0.26	2	50.00

Rottweiler	1	0.26	1	100.00
Samoyed	1	0.26	2	50.00
Shar-Pei	1	0.26	1	100.00
Siberian Husky	1	0.26	1	100.00
Total	383		929	

n_1 = Number of CAD dogs in each breed

n_2 = Number of allergic dogs in each breed

The Bangkok metropolitan consists of fifty districts, The distribution of 383 CAD dogs in Bangkok's districts and surrounding areas was show in table 7.

Table 7 Distribution of 383 CAD dogs (n) in Bangkok and surrounding areas

Distribution of 383 CAD dogs (n) in Bangkok and surrounding areas					
	Name of districts in Bangkok	Number of dogs (n)		Name of districts in Bangkok	Number of dogs (n)
1	Phra Nakhon district	11	26	Din Daeng district	5
2	Dusit district	7	27	Bueng kum district	2
3	Nong Chok district	0	28	Sathon district	6
4	Bang Rak district	9	29	Bang Sue district	2
5	Bang Khen district	2	30	Chatuchak district	6
6	Bang Kapi district	3	31	Bang Kho laem district	13
7	Pathum Wan district	12	32	Prawet district	11
8	Pom Prap Sattru Phai district	8	33	Kholong Toei district	16
9	Phra Khanong district	12	34	Suan Luang district	5
10	Min Buri district	1	35	Chom Thong district	15
11	Lat Krabang district	4	36	Don Mueang district	5
12	Yan Nawa district	12	37	Ratchathewi district	8
13	Samphathawong district	1	38	Lat Phrao district	2
14	Phaya Thai district	10	39	Watthana district	15
15	Thon Buri district	9	40	Bang Khae district	6
16	Bangkok Yai district	7	41	Lak Si district	1
17	Huai Khwang district	8	42	Sai Mai district	3
18	Khlong San district	9	43	Khan Na Yao district	2
19	Taling Chan district	8	44	Saphan Sung district	3

20	Bangkok Noi district	3	45	Wang Thonglang district	6
21	Bang Khun Thian district	7	46	Khlong Sam Wa district	0
22	Phasi Charoen district district	16	47	Bang Na district	6
23	Nong Khaem district	2	48	Thawi Watthana district	0
24	Rat Burana district	8	49	Thoung Khru district	11
25	Bang Phlat district	5	50	Bang Bon district	0
Surrounding areas					
1	Nonthaburi province	22	5	Ayutthaya province	2
2	Samut Prakan province	21	6	Nakhon Pathom province	2
3	Pathum Thani province	7	7	Samut Sakhon province	2
4	Pathum Thani province	4			

2. Sex

Form 383 dogs, CAD dogs were found in 209 males (54.57%) and 174 females (45.43%)

3. Distribution of lesions

The distribution of skin lesions was reported as the percentage in figure 2.

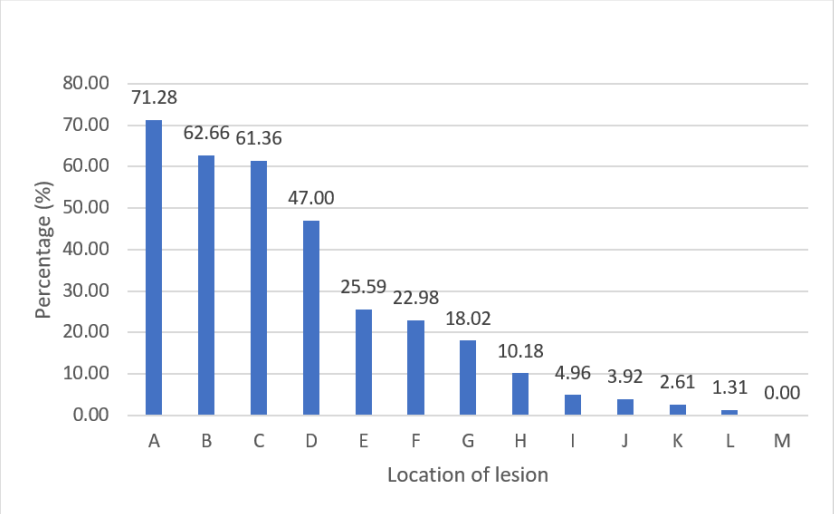

Figure 2 Percentage of distribution of skin lesions observed in 383 CAD dogs.
A: front feet, B: ear pinnae, C: hind feet, D: abdomen, E: lips, F: axillae, G: chest, H: genitalia or ventral tail, I: elbow, J: lumbosacral area, K: lateral flanks, L: eyelids, M: ear margin.

4. Housing environment

The housing environment of CAD dogs were indoor dogs (281/383; 73.37%) and outdoor dogs (102/383; 26.63%).

5. Concurrent disease

Concurrent diseases included Malassezia dermatitis, otitis externa, pododermatitis, and pyoderma. The prevalence of concurrent diseases is present in figure 3.

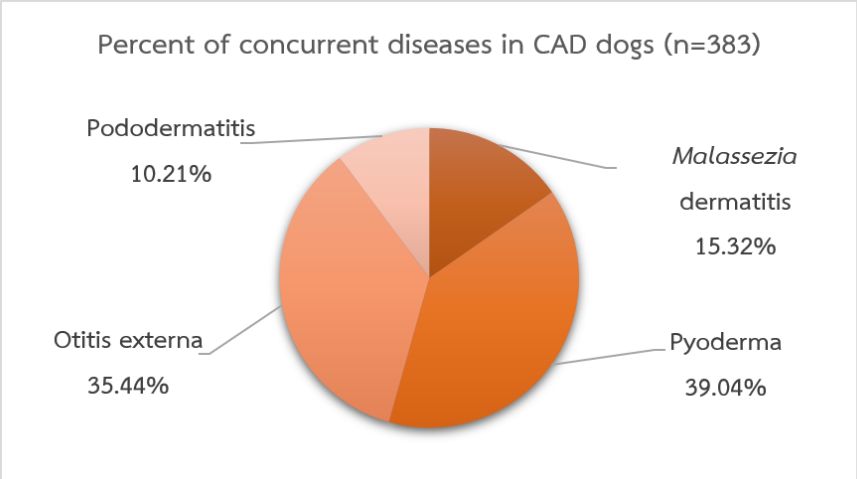

Figure 3 Percentage of concurrent diseases observed in 383 CAD dogs.

Pyoderma was the most frequently elicited a concurrent disease, followed by otitis externa, Malassezia dermatitis, and pododermatitis, respectively.

Phase 2: Distribution of causative allergens

Twenty-eight CAD dogs were completely identified the causative allergens by Canine ALLERCEPT® panels from HESKA. The percentage positive results in each group of causative allergens were as follows: house dust mite (75.00%), grass (62.14%), weed (47.45%), tree (46.43%), yeast (39.29%), and flea saliva (21.43%) (Table 8).

Table 8 Type and percentage of allergens found in 28 CAD dogs using Canine ALLERCEPT® panels from HESKA

type	Group (%)	Allergens	Number of dogs	Percentage
Indoor allergens	House dust mite (75.00%)	*Dermatophagoides farinae*	22	78.57
		Acarus siro	21	75.00
		Tyrophagus putrescentiae	21	75.00
		Blomia tropicalis	20	71.43
Outdoor allergens	Grass (62.14%)	Bahia Grass (*Paspalum notatum*)	19	67.86
		Johnson Grass (*Sorghum halepense*)	18	64.29
		Bermuda Grass (*Cynodon dactylon*)	17	60.71
		Meadow Fescue (*Festuca pratensis*)	17	60.71
		Timothy (*Phleum pratense*)	16	57.14
	Tree (46.43%)	Palm (*Syagrus Romanzoffianum,Elaeis guineensis*)	18	64.29
		English Plantain (Plantago lanceolata)	14	50.00
		Mugwort (*Artemisia vulgaris*)	14	50.00
		Melaleuca (*Maleleuca quinquenervia*)	13	46.43
		Mulberry (*Morus alba*)	12	42.86
		Oak (*Quercus alba*)	7	25.00
	Weed (47.45%)	Careless Weed (*Xanthium strumarium*)	17	60.71
		Cocklebur (*Cyclachaena xanthifolia*)	14	50.00
		Marsh Elder (*Ambrosia artemisifolia*)	13	46.43
		Red Sorrel (*Chenopodium*)	13	46.43
		Russian Thistle (*Amaranthus hybridus*)	13	46.43
		Shadscale (*Salsolakali*)	13	46.43
		Ragweed mix (*Plantago lanceolata*)	10	35.71
Other	Yeast	*Malassezia pachydermatis*	11	39.29
	Flea saliva	Flea Saliva	6	21.43

CHAPTER 5 DISCUSSION

Prevalence of canine atopic dermatitis in Bangkok

This research has attempted to assess prevalence of canine atopic dermatitis in Bangkok area. In the first phase of study, prevalence of CAD from visiting dogs was 1.16 percent and percentage of CAD dogs from skin disease's dogs and allergic dermatitis's dogs were 11.61% and 40.48% respectively. The results were lower than previous reports (Hillier and Griffin, 2001; Hill et al., 2006; Kuribayashi et al., 2018).

Breed

The study included 36 different breeds of dogs as atopic dogs. Shih Tzu (95/24.80 percent), Mixed breed (67/17.49 percent), Beagle (39/10.18 percent), Poodle (30/7.83 percent), and French Bulldog (21/5.48 percent) were the top five most atopic breeds. The prevalence of breeds with atopic dermatitis varies according to other surveys. The West Highland White Terrier, Golden Retriever, and Labrador Retriever were breeds with a history of CAD (DeBoer and Marsella, 2001; Nuttall et al., 2006; Marsella and De Benedetto, 2017).In our study, we discovered that mixed breed dogs constituted the majority of visiting dogs (8490/25.64%). It is possible that the mixed breed is the largest population in the Bangkok area. As a result, our study discovered a high prevalence of CAD in mixed breed dogs. Furthermore, CAD was also found in Thai Bangkaew and Thai Ridgeback dogs in this study. Our finding discovered a new report on canine atopic dermatitis in mixed breed and Thai breeds dogs.

Distribution of lesions

All of the atopic dogs exhibited a range of lesion distributions. Our study was interested in these clinical signs and the distribution of lesions because they are frequently regarded as a primary feature of canine atopic dermatitis. This predilection has been attributed to the percutaneous absorption of allergens (Zur et al., 2002). The front feet, ear pinnae, hind feet, abdomen, lips, axillae, chest, genitalia or ventral tail, elbow, lumbosacral area, lateral flanks, and eyelids all had skin lesions, consistent with previous research (Chanthick et al., 2008; Hensel et al., 2015). One of the clinically significant features of canine atopic dermatitis was the distribution of lesions. Occasionally, veterinarians will use Favrot's criteria to bolster the likelihood of a patient having CAD (Favrot et al., 2010b; Hensel et al., 2015). Favrot's criteria include an onset age of three years, pruritus that is corticosteroid responsive, chronic, or recurrent yeast infections, affected front feet and ear pinnae, and an unaffected dorso-lumbar region. As a result, it may aid veterinarians in diagnosing CAD.

Housing environment

Similarly to the previous report, 73.37 percent of atopic dogs in this study were housed indoors (Favrot et al., 2010b; Hensel et al., 2015). As a result, it appears as though the majority of CAD dogs lived in an indoor environment.

Concurrent diseases

Concurrent diseases are frequently seen in dogs with atopic dermatitis. Concurrent diseases were classified into four categories in this study: pyoderma, otitis externa, Malassezia dermatitis, and pododermatitis. Pyoderma was the most frequently occurring concurrent disease in this study (39.04 percent). It is estimated

that approximately one third of atopic dogs suffer from pyoderma. Pyoderma was a significant issue not only in the treatment of pruritus and allergy, but also in the development of multidrug resistance (Zur et al., 2002). Otitis externa was the second concurrent disease in this study (35.44 percent). Otitis externa was an inflammatory condition that affected the ear pinnae and outer ear canal. CAD was a contributing factor to the development of otitis externa (Saridomichelakis and Olivry, 2016; Olivry and Banovic, 2019). Thus, when veterinary medicine is able to control the allergic reaction to CAD, it is also able to reduce and prevent the occurrence of Otitis externa (Saridomichelakis and Olivry, 2016; Olivry and Banovic, 2019). *Malassezia* dermatitis was a frequent occurrence in this study, affecting 15.32 percent of CAD dogs. It is caused by an overgrowth of *Malassezia patchydermatis* on the dog's skin's lipid layer (Zur et al., 2002). *Malassezia* populations have been found to be significantly higher in atopic dogs than in non-atopic dogs (Zur et al., 2002). *Malassezia* dermatitis is characterized by odor, inflammation, and, most significantly, itching. When, veterinarians control yeast infections, which can reduce pruritus and inflammation (Zur et al., 2002). Another concurrent disease in our study was pododermatitis (10.21 percent). Pododermatitis was a term used to describe inflammation of the foot skin (DeBoer and Marsella, 2001; Zur et al., 2002; Favrot et al., 2010b). It frequently results in swollen, red, and itchy feet in dogs. The underlying cause of pododermatitis, particularly pedal furunculosis, was canine atopic dermatitis. Pododermatitis is clinically significant in canine atopic dermatitis because the interdigital space of the four paws is an area of the body susceptible to allergen accumulation(Favrot et al., 2010b; Hensel et al., 2015; Santoro et al., 2015).

Distribution of causative allergens

In the second phase of the study, ALLERCEPT® panels from HESKA (Asia panel) were used to detect aeroallergens specific to the Asia region. This test identified 24 allergens responsible for allergic reactions in six groups (house dust mite, grass, weed, tree, yeast, and flea saliva) (Mueller et al., 2016; Saridomichelakis and Olivry, 2016). Allergens are immunogens capable of eliciting an immune response. Aerollergens in veterinary dermatology range in size from 10 to 100 kDa (Maeda et al., 2009; Mueller et al., 2016). The allergens that atopic dogs are exposed to vary according to their habitat, lifestyle, geographical location, and seasonality (Bizikova et al., 2015). A WHO/International Union of Immunological Societies committee assigns standardized names to allergens from any organism based on their order of discovery and sequence homology (Maeda et al., 2009). House dust mites (HDM) and their products are major causes of atopic sensitization and allergic disease worldwide (Sánchez-Borges et al., 2017; Huang et al., 2018). They are eight-legged arachnids. Egg, larval, protonymph, tritonymph, and adult stages comprise their approximately three-month of lifespan. The adults are measured between 30-40 microns. *Dermatophagoides farinae, Dermatophagoides pteronyssinus, Tyrophagus putrescentiae, Acarus siro,* and *Blomia tropicalis* were the most allergenic house dust mite species (Chanthick et al., 2008). Other studies have identified HDM as the most common allergen associated with CAD (Masuda et al., 2000; Zur et al., 2002; Chanthick et al., 2008; Kang et al., 2014; Chermprapai et al., 2020). *D. pteronyssinus* was the most abundant species in previous surveys of house dust mites in Thailand, followed by *D. farinae* and other species (Chanthick et al., 2008). On the other hand, our study discovered that *D. farinae* had the highest percentage of positive results (78.57%), followed by *Acarus siro* (75.00%), T. *putrescentiae* (75.00%), and *Blomia tropicalis* (71.43%). These percentages of positive results were higher than those for the other allergens examined in this study. Table 9 compares the prevalence of causative allergens of our study to other studies.

Table 9 Comparison of the prevalence of causative allergens associated with canine atopic dermatitis (IDST and ASIS)

Allergens	Present study (n=28)	Chermprapai *et al.*, 2020 (n=23)	Kang *et al.*, 2014 (n=101)	Masuda *et al.*, 2000 (n=42)	Zur *et al.*, 2002 (n=266)	Chanthick *et al.*, 2008 (n=114)
Counties	Thailand	Thailand	South Korea	Japan	USA	Thailand
technique	ASIS			IDST		
House dust mites (HDM)	75.00	69.57	61.4	69.0	71.0	-
D. *farinae*	78.57	56.52	-	-	-	74.56
D. *pteronyssinus*	-	-	-	-	-	53.51
Grass	62.14	34.78	-	31.4	35.3	31.3*
Weeds	47.45	43.38	-	17.1	45.5	29.7*
Trees	46.43	30.43	-	76.2	53.4	-
Yeast and molds	39.29	30.43	21.9	16.7	60.0	
Flea saliva and insect	21.43	0.00	7.9	25.7	46.0	-

*Allergen: wild grass and weed pollens (Chanthick et al., 2008)

Outdoor allergens such as grasses, weeds, and trees were discovered to produce positive responses in dogs living in Bangkok. Bangkok is situated in a tropical region that is extremely humid and hot. Pollens come in a variety of forms (Chanthick et al., 2008). In a previous survey of aeroallergens in Bangkok, it was discovered that the most prevalent pollens were wild grasses (31.3 percent) and weeds (29.7 percent) (Chanthick et al., 2008). Bermuda grass, Para grass, and Bahia grass were all common wild grasses in Bangkok, while careless weed, Shadscale (Salsolakali), and Red Sorrel were all common weeds. Palm and other tree pollens account for less than 5% of the pollen found in the atmosphere (Chanthick et al., 2008). The grasses, weeds, and trees that performed the best in our study were Bahia grass (67.86 percent), palm (64.29 percent), and Careless weed (60.71 percent), respectively. The results were higher than in the previous report (Chermprapai et al., 2020). It may be related to the owners' and dogs' lifestyles in Bangkok, where dogs are frequently housed indoors rather than outdoors, and receive limited outdoor exposure.

CHAPTER 6 CONCLUSION

The current study established the prevalence of canine atopic dermatitis and elucidated the allergens responsible for the disease in atopic dogs using allergen-specific IgE serology. Our findings indicate that 1.16% of visiting dogs may have CAD. Breed predisposition, lesion distribution, and concurrent disease are all tools that can assist veterinarians in remaining current on this disease. The diagnosis of CAD was made to rule out the possibility of another type of skin disease. ASIS is simple to implement in practice and causes less stress to patients. Our results found 78.57% of positive IgE to *Dermatophagoides farinae.* It's highest percentage of causative allergens in this study. The serology results may be beneficial for pruritus control, allergen avoidance, and allergen-specific immunotherapy.

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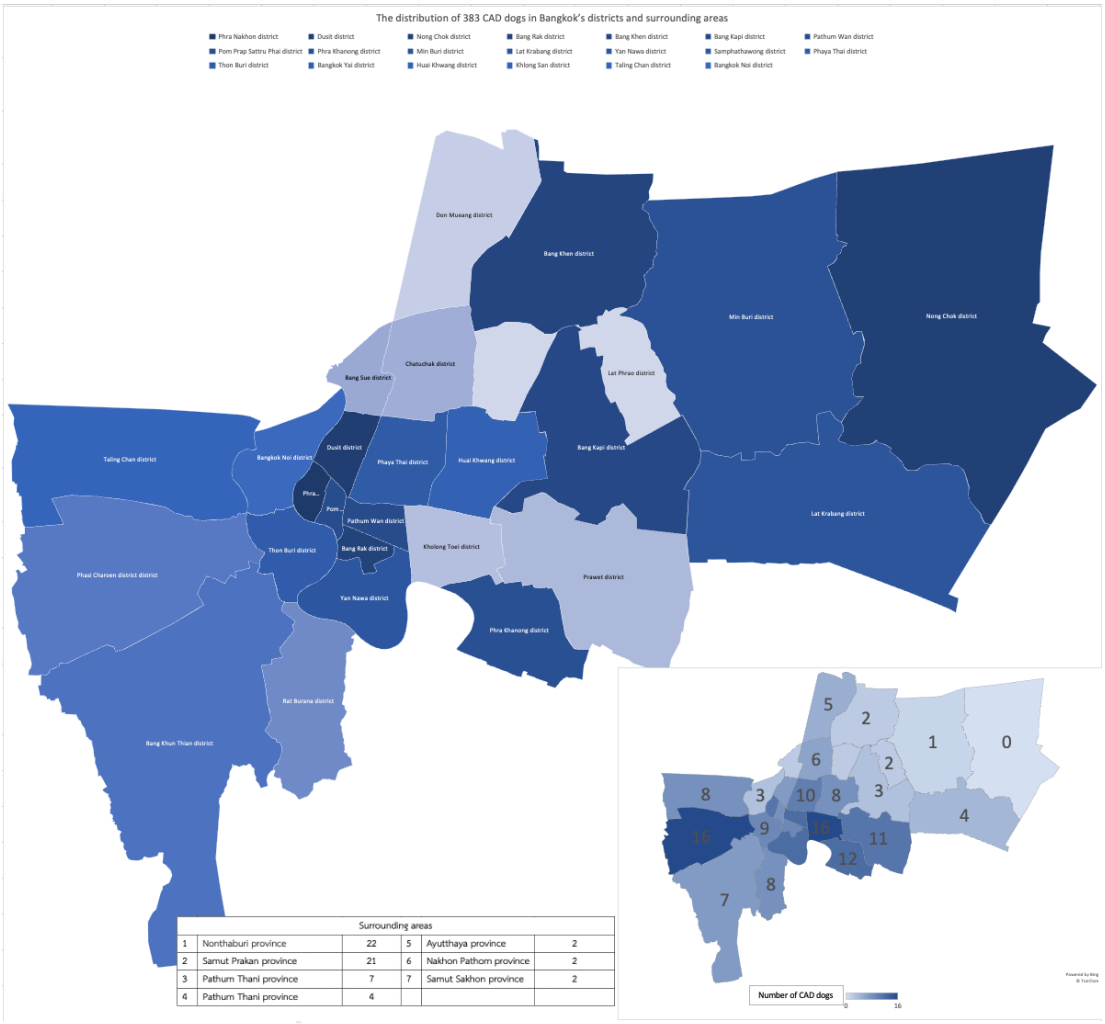
Appendix



Surrounding areas					
1	Nonthaburi province	22	5	Ayutthaya province	2
2	Samut Prakan province	21	6	Nakhon Pathom province	2
3	Pathum Thani province	7	7	Samut Sakhon province	2
4	Pathum Thani province	4			

The distribution of 383 CAD dogs in Bangkok's districts and surrounding areas

Type of allergens

Group	Picture	Allergens	Thai name
House dust mite		*Dermatophagoides farinae*	-
		*Acarus siro*	-
		*Tyrophagus putrescentiae*	-
		*Blomia tropicalis*	-
Grass		Bahia Grass (*Paspalum notatum*)	หญ้าบาเฮีย
		Johnson Grass (*Sorghum halepense*)	หญ้าพง
		Bermuda Grass (*Cynodon dactylon*)	หญ้าแพรก

Group	Picture	Allergens	Thai name
Grass		Timothy (*Phleum pratense*)	หญ้าทิโมธี
Tree		Palm (*Syagrus Romanzoffianum)*	พืชตระกูล ปาล์ม
		English Plantain (Plantago lanceolata)	เทียนเกล็ดหอย
		Mugwort (*Artemisia vulgaris*)	โกฐจุฬาลัมพาไทย
		Melaleuca (*Maleleuca quinquenervia*)	เสม็ดขาว
		Mulberry (*Morus alba*)	ม่อน
		Oak (*Quercus alba*)	โอ๊คขาว

Group	Picture	Allergens	Thai name
Weed		Careless Weed (*Xanthium strumarium*)	ผักกระชับ
		Cocklebur (*Cyclachaena xanthifolia*)	ดอกสร้อยทอง
		Marsh Elder (*Ambrosia artemisifolia*)	วัชพืช กลุ่มผักโขม
		Red Sorrel (*Chenopodium*)	ผักโขมหัด
		Russian Thistle (*Amaranthus hybridus*)	หญ้ากาลี
		Shadscale (*Salsolakali*)	อับทานตะวัน
		Ragweed mix (*Plantago lanceolata*)	ผักกระชับ

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