pISSN 1598-298X / eISSN 2384-0749
J Vet Clin 34(1) : 18-22 (2017)
http://dx.doi.org/10.17555/jvc.2017.02.34.1.18

Journal of Veterinary Clinics
www.ksvc.or.kr

Efficacy of Half Dose House Dust Mites-Specific Immunotherapy on Canine Atopic Dermatitis

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(Received: October 16, 2014 / Accepted: January 19, 2017)

Abstract : The aim of this study was to prove that the hypothesis of half dose (HD) allergen-specific immunotherapy (ASIT) in the treatment of canine atopic dermatitis (CAD) would result in a similar success rate compared to the standard dose (SD) ASIT. Clinical signs were evaluated using a third version of the Canine Atopic Dermatitis Extent and Severity Index (CADESI-03) prior to ASIT (day 0), at the end of induction (day 43), and at three month afterwards (day 90). Of the 18 atopic dogs, 12 dogs (SD group: 6; HD group: 6) had a good - excellent response to the house dust mites-specific immunotherapy. The efficacies of ASIT were 66.6% in both groups. The grades of reduction rate CADESI-03 were not different between two groups. Therefore, half dose protocol of house dust mites-specific immunotherapy is an effective and efficient method to treat CAD.

Key words **:** dog, atopic dermatitis, house dust mite, half-dose, immunotherapy.

Introduction

Canine atopic dermatitis (CAD) is a common dermatosis of dogs defined as a genetically - predisposed inflammatory and pruritic skin disease with IgE antibodies, most commonly directed against environmental allergens (9). The true incidence of the disease in the dog population is unknown, but estimates vary from 3-15% (18). The diagnosis of CAD is based on a combination of epidemiological, clinical and diagnostic findings and on ruling out other conditions that may present with similar clinical signs such as flea allergy dermatitis, bacterial pyoderma, food hypersensitivity or *Malassezia* dermatitis (4,23). CAD is commonly managed with glucocorticoids, cyclosporine, antihistamines and essential fatty acid supplementation (14) but the only specific treatment is allergen - specific immunotherapy (ASIT) (4,8). This is the practice of administering gradually increasing quantities of an allergen extract to an allergic subject to ameliorate the symptoms associated with subsequent exposure to the causative allergen (1,8). Numerous open uncontrolled studies suggest that ASIT is efficacious for the treatment of CAD. The reported success rates with ASIT range from 50 to 100% (12,17,18).

Although ASIT is frequently used in CAD, confirmation of the optimal maintenance dose is needed because of the lack of information on the optimal maintenance dose. The optimal maintenance dose range depends on clinical effectiveness and safety. Too low a dose is ineffective, although safe; too high a dose may be effective, but can be associated with a higher incidence of systemic reactions (18). A small number of open studies have also described the use of higher or lower doses of ASIT, either from the beginning of the treatment or in the maintenance stage only, and all protocols seemed to be effective (7,19).

Unlike dogs in the United States and Europe, dogs in South Korea are mostly small breeds. Therefore half dose immunotherapy in the treatment of CAD is thought to be effective as standard dose immunotherapy. The aim of this study was to prove that the hypothesis of half dose ASIT in the treatment of CAD would result in a similar success rate compared to the standard dose ASIT.

Materials and Methods

Patients

Dogs suffering from AD ($n = 18$) were recruited from the Veterinary Medical Teaching Hospital of Chungnam National University. The diagnosis of CAD was made after fulfillment of Favrot's criteria (6) and exclusion of other similar pruritic disorders such as adverse reactions to food, scabies or microbial infections (20).

Allergens

The intradermal skin test (IDST) was examined using two species of house dust mites, *Dermatophagoides farinae* and *Dermatophagoides pteronyssinus*. Commercial allergen preparations were purchased from Greer Laboratoies (Lenoir, USA).

Intradermal skin test (IDST)

All steroids, sedatives, immunosuppressants, antihistamines and tranquilizers were discontinued for at least 21 days before IDST (11). Each test included two aqueous allergenic extracts diluted with 0.9% normal saline. The positive control was histamine phosphate 0.275 mg/mL (Histatrol; Central

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Laboratories, Port Washington, USA) and negative control was 0.9% normal saline. Hair on the lateral aspect of the thorax was clipped carefully to avoid skin irritation. The injection sites were marked, and 0.05 ml of each allergen or control solution was injected intradermally using a 1 ml syringe with a 26 gauge needle. At 15 min after injection, the diameter of each wheal was measured in millimeters. Reactions were recorded with a score designated as 0, 1, 2, 3, or 4 (0 = a wheal the same size as the negative control, 1 = one - quarter, 2 = one - half, 3 = three - quarter the size of the negative and positive control, and 4 = a wheal the same or larger size of the positive control). Any reaction of 2 or greater was regarded as positive.

Study groups

The dogs were allocated to two treatment groups by block randomization. The control group (standard dose group, SD group) consisted of dogs receiving ASIT following the standard protocol as indicated by the manufacturer. The experimental group (half dose group, HD group) consisted of dogs treated with ASIT but receiving half dose of the allergen at the same standard intervals.

Allergen - specific immunotherapy (ASIT)

The allergens of ASIT vaccines were determined by correlating positive IDST results. All vaccines were prepared with allergen extracts obtained from Greer Laboratories and given subcutaneously, according to the standard protocol (Table 1).

Clinical evaluations

Clinical signs were evaluated using a third version of the Canine Atopic Dermatitis Extent and Severity Index (CADESI-03) prior to ASIT (day 0), at the end of induction (day 43), and at three month afterwards (day 90). Dogs were scored for erythema, lichenification, excoriation and self-induced alopecia on 62 body sites. The lesions were scored on a scale of 0-5 (0: none, 1: mild, 2, 3: moderate, 4, 5: severe).

Table 1. Protocol of ASIT in the SD groups and HD groups

Day	Standard dose		Half dose	
	Volume (ml)	Dose (PNU)	Volume (ml)	Dose (PNU)
1	0.1	10	0.05	5
4	0.2	20	0.1	10
7	0.4	40	0.2	20
10	0.8	80	0.4	40
13	1.0	100	0.5	50
16	0.1	100	0.05	50
19	0.2	200	0.1	100
22	0.4	400	0.2	200
25	0.8	800	0.4	400
28	1.0	1,000	0.5	500
31	0.1	1,000	0.05	500
34	0.2	2,000	0.1	1,000
37	0.4	4,000	0.2	2,000
40	0.8	8,000	0.4	4,000
43	1.0	10,000	0.5	5,000
Every 10 days	1.0	10,000	0.5	5,000

PNU, protein nitrogen units

Calculation of improvement

The CADESI-03 scores before and after treatment were used to grade improvement percentage from the baseline CADESI-03 and transformed to a five-point grade system (0: increase, 1: < 25% reduction in CADESI-03, 2: 25-49% reduction in CADESI-03, 3: 50-74% reduction in CADESI-03, 4: > 75% reduction in CADESI-03). This grade was a five-grade categorical point consisting of the terms and values 'none (0), poor (1), fair (2), good (3) and excellent (4)' (15).

Statistical analysis

Independent *t* - test and Mann - Whitney *U* - test were carried out to investigate whether there were differences between the SD and HD groups at the beginning of the study in age, bodyweight, or interval between the onset of clinical signs suggestive of CAD and the starting of ASIT.

To assess the change in CADESI-03 scores, the end point was defined in this way. Those dogs that were withdrawn were added to the complete set, with their last value prior to withdrawal included as the end point. To evaluate whether the CADESI-03 scores for each dose group separately had changed between the start and the end of the study, Wilcoxon signed rank tests for paired samples (*Wp*) were performed. Mann - Whitney *U* - test was carried out to investigate whether there were differences in grade, between the SD and HD groups. Differences at $p < 0.05$ were considered to be statistically significant.

All the statistical analyses were performed using a computer statistical package (SPSS for windows, Releases 19.0, SPSS Inc., USA).

Results

Patients

Eighteen dogs were enrolled in the study. There were 9 dogs in the SD group, and 9 in the HD group. Data concerning the signalment and the interval between the onset of clinical signs and the beginning of ASIT in the two groups were summarized in Table 2. There were no significant differences in age (SD: mean 4.6 years; HD: mean 4.3 years), weight (SD: mean 8.3 kg; HD: mean 5.6 kg) and interval between the onset of clinical signs and the beginning of ASIT (SD: mean 24.1 months; HD: mean 31.7 months).

Clinical evaluations

CADESI-03 scores and reduction rates in the SD and HD groups were summarized in Table 3 and Table 4. The median CADESI-03 scores at the beginning of the study were 214.1 in the SD group and 175.8 in the HD group. At the end of the study, the medians were 96.2 and 103.1 in the SD and HD group respectively. There were no significant differences in the change of CADESI-03 score from the beginning to the end of the study in the SD group. In contrast, there

Table 2. Signalment, intervals between the onset of clinical signs and the beginning of ASIT and skin test results in the two groups

Standard dose group						
	Breed	Age (years)	Sex	Weight (kg)	Interval (months)	Allergen
1	Shih Tzu	3	MC	6.6	30	Df, Dp
2	Yorkshire Terrier	3.5	MC	3.8	1	Df
3	Cocker Spaniel	5	M	10.0	36	Df, Dp
4	Cocker Spaniel	5	F	10.0	48	Df, Dp
5	Cocker Spaniel	6.5	F	11.3	6	Df, Dp
6	Shar-Pei	7.5	FS	14.5	66	Df, Dp
7	Boston Terrier	4	FS	8.7	6	Df, Dp
8	Maltese	3	MC	3.9	12	Df, Dp
9	Shih Tzu	4	MC	6.3	12	Df, Dp
Half dose group						
	Breed	Age (years)	Sex	Weight (kg)	Interval (months)	Allergen
1	Shih Tzu	2	F	5.5	1	Df
2	Shih Tzu	6	MC	5.4	70	Df
3	Maltese	3	MC	6.0	12	Df, Dp
4	Chihuahua	3	F	2.8	12	Df, Dp
5	Shih Tzu	6	M	5.4	36	Df
6	Shih Tzu	6	M	4.5	60	Df, Dp
7	Shih Tzu	8	MC	9.7	60	Df
8	Shih Tzu	2	M	5.8	22	Df, Dp
9	Miniature Pinscher	3	MC	5.4	12	Df, Dp

F, female; FS, female spayed; M, male; MC, male castrated
Df, *D. farinae*; Dp, *D. pteronyssinus*

Table 3. CADESI-03 scores and reduction rates in the standard dose group

	Day 0	Day 43	Day 90	Reduction rate (%)
1	307	118	118	61.6
2	162	118	74	54.3
3	509	141	64	87.4
4	178	194	194	–9.0
5	97	51	98	–1.0
6	217	141	99	60.8
7	84	129	85	–1.2
8	251	210	87	65.3
9	122	61	61	50.0

Table 4. CADESI-03 scores and reduction rates in the half dose group

	Day 0	Day 43	Day 90	Reduction rate (%)
1	212	98	62	70.8
2	143	108	69	51.7
3	63	109	166	–1.8
4	182	80	80	56.0
5	149	72	72	51.7
6	158	86	77	51.3
7	298	148	201	32.6
8	165	167	167	–1.2
9	112	102	34	69.6

were significant differences in the HD group. The rates of improvement in CADESI-03 scores did not differ between the SD and HD group ($p = 0.825$).

Efficacy of ASIT

Both SD group and HD group had similar responses to ASIT, which showed decreased CADESI-03 scores throughout the study. Out of these 18 cases, 1 from the SD group had an excellent response, 11 (SD group: 5; HD group: 6) had a good response, 1 from the HD group had a fair response, and 5 (SD group: 3; HD group: 2) had no response. The immunotherapy was therefore considered to have been helpful in 12 (66.6%) patients, and there were no significant differences between SD group and HD group ($p = 0.879$).

Discussion

Responses to house dust mites-specific immunotherapy in this study were graded as good to excellent in 66.6% of the SD group and 66.6% of the HD group. These results were comparable with some other studies, in which 61.3% (12), 63.0% (17), 50-80% (3) and 50-100% (18) of the cases

responded to ASIT.

The efficacy of ASIT in CAD is associated with several factors that include the dose of allergen, the number of allergens to which the patient is hypersensitive, body weight, age and interval between the onset of clinical signs and the beginning of ASIT (2).

In the dose of allergen, it is convention to use a maintenance vial with a total dose of 10,000 to 20,000 PNU/ml of allergen (18). However, there are no studies that confirm these doses to be any better or worse than any other dose of allergens. An unpublished study compared high dose ASIT (maintenance dose 40,000 PNU/ml) to low dose ASIT (maintenance dose 10,000 PNU/ml), and found that the high dose ASIT was more effective than the low dose ASIT (7). In a study, low dose ASIT using 100 to 1,000 fold less than the dose used for IDST was effective in 71.6% of the dogs with atopic dermatitis (7). An open study that compared the response rates in two groups of dogs receiving ASIT found a significant difference with the high dose group (maintenance dose 20,000 PNU/ml) exhibiting 77% good to excellent response while the low dose group (maintenance dose 10,000 PNU/ml) exhibited a 96% good to excellent response. The low dose group included dogs with either persistent worsening of the initial problem of pruritus or an unacceptable adverse reaction on the high dose ASIT. Low dose ASIT showed high responses because the doses were evaluated and modified to minimize adverse effects (19). In our study, we could also find favorable treatment results in HD group (maintenance dose 5,000 PNU/ml) compared with SD group (maintenance dose 10,000 PNU/ml).

In human medicine, optimal immunotherapy doses for house dust mite - induced allergy were established to be approximately 7 μg (13) and 12 μg (5) for *Dermatophagoides pteronyssinus* and 10 μg (16) for *Dermatophagoides farinae*. However, optimal doses of house dust mites have not been studied for immunotherapy in veterinary medicine. The present investigation showed the response rates to maintenance dose of *Dermatophagoides farinae* were 100% (1/1) at 10,000 PNU/ml, 60% (8/12) at 5,000 PNU/ml and 60% (3/5) at 2,500 PNU/ml.

In our study, only two indoor allergens, *Dermatophagoides farinae* and *Dermatophagoides pteronyssinus,* were used in IDST and ASIT. Only two allergens were used because these allergens appeared to be the most important allergens among atopic dogs in Korea (21,25). These results were different to the results of studies in USA, which concluded that both house dust mites and pollen are important allergens (10).

A recent randomized placebo controlled trial that included dogs allergic to house dust mites and at least one additional allergen found that house dust mites restricted immunotherapy was insufficient to control the disease (24). In contrast, in this study, the ASIT using house dust mites was effective. One possible explanation is that there are differences in the number of causative allergens with respect to the geographical region. The number of causative allergens was 6.5 ± 5.4 in a European study (24) while the number was 2.5 ± 0.9 in a Korean study (21). These results were similar to the result of a previous study which concluded that an improved efficacy is associated with fewer numbers of causative allergens (8).

The other factors for each dog herein, such as body weight, age and interval between the onset of clinical signs and the beginning of ASIT were investigated. In this study, the success rates of immunotherapy were 100% (3/3) in dogs with a bodyweight less than 5 kg, 60% (6/10) in 5-10 kg and 50% (2/4) in dogs that weighed more than 10 kg. The response rates to ASIT in regard to age were 70% (7/10) in dogs under 5 years and 63% (5/8) in dogs above 5 years. There were no significant differences in intervals, 66.6% (6/9) in both less than 12 months and more than 12 months.

In summary, this study shows that ASIT using half dose protocol in comparison to standard dose protocol had similar therapeutic effect but also had reduced costs and lower incidence rates of adverse effects. Therefore, half dose ASIT is considered to be a useful protocol to treat CAD.

References

1. Bousquet J, Lockey R, Malling HJ. Allergen immunotherapy: therapeutic vaccines for allergic diseases. A WHO position paper. J Allergy Clin Immunol 1998; 102: 558-562.
2. Colombo S, Hill PB, Shaw DJ, Thoday KL. Effectiveness of low dose immunotherapy in the treatment of canine atopic dermatitis: a prospective, double - blinded, clinical study. Vet Dermatol 2005; 16: 162-170.
3. Day MJ. Immune-mediated skin disease. In: Immunology of the dog and cat, 2nd ed. London: Manson. 2008: 122-135.
4. DeBoer DJ, Hillier A. The ACVD task force on canine atopic dermatitis (XV): fundamental concepts in clinical diagnosis. Vet Immunol Immunopathol 2001; 81: 271-276.
5. Ewan PW, Alexander MM, Snape C, Ind PW, Agrell B, Dreborg S. Effective hyposensitization in allergic rhinitis using a potent partially purified extract of house dust mite. Clin Allergy 1988; 18: 501-508.
6. Favrot C, Steffan J, Seewald W, Picco F. A prospective study on the clinical features of chronic canine atopic dermatitis and its diagnosis. Vet Dermatol 2010; 21: 23-31.
7. Garfield RA. Injection immunotherapy in the treatment of canine atopic dermatitis: comparison 3 protocols. Proceedings of the 9th Annual AAVD and ACVD Meeting. Montreal, Canada. 1992: 7-8.
8. Griffin CE, Hillier A. The ACVD task force on canine atopic dermatitis (XXIV): allergen-specific immunotherapy. Vet Immunol Immunopathol 2001; 81: 363-383.
9. Halliwell R. Revised nomenclature for veterinary allergy. Vet Immunol Immunopathol 2006; 114: 207-208.
10. Hill PB, DeBoer DJ. The ACVD task force on canine atopic dermatitis (IV): environmental allergens. Vet Immunol Immunopathol 2001; 81: 169-186.
11. Hillier A, DeBoer DJ. The ACVD task force on canine atopic dermatitis (XVII): intradermal testing. Vet Immunol Immunopathol 2001; 81: 208-304.
12. Nuttall TJ, Thoday KL, van den Broek AH, Jackson HA, Sture GH, Halliwell RE. Retrospective survey of allergen immunotherapy in canine atopy. Vet Rec 1998; 143: 139-142.
13. Olaguíbel JM, Tabar AI, García BE, Martín S, Rico P. Long-term immunotherapy with an optimal maintenance dose of a standardized Dermatophagoides pteronyssinus extract in asthmatic patients. J Investig Allergol Clin Immunol 1999; 9: 110-116.
14. Olivry T, Mueller RS. Evidence-based veterinary dermatology: a systemic review of the pharmaco-therapy of canine atopic

dermatitis. Vet Dermatol 2003; 14: 121-146.
15. Olivry T, Marsella R, Iwasaki T, Mueller R. Validation of CADESI-03, severity scales for clinical trials enrolling dogs with atopic dermatitis. Vet Dermatol 2007; 18: 78-86.
16. Olsen OT, Larsen KR, Jacobsen L, Svendsen UG. A 1 - year, placebo - controlled, double - blind house - dust - mite immunotherapy study in asthmatic adults. Allergy 1997; 52: 853-859.
17. Park S, Ohya F, Yamashita K, Nishifuji K, Iwasaki T. Comparison of response to immunotherapy by intradermal skin test and antigen - specific IgE in canine atopy. J Vet Med Sci 2000; 62: 983-988.
18. Reedy LM, Miller WH, Willemse T. Immunotherapy. In: Allergic Skin Diseases of Dogs and Cats, 2nd ed. Philadelphia: WB Saunders. 1997: 116-149.
19. Rosser EJ. Aqueous hyposensitization in the treatment of canine atopic dermatitis: a retrospective study of 100 cases. In: Advances in Veterinary Dermatology, Vol. 3. Butterworths 1998; 169-176.
20. Scott DW, Miller WH, Griffin CE. Skin immune system and allergic skin disease. In: Muller and Kirk's Small Animal Dermatology, 6th ed. Philadelphia: WB Saunders. 2001: 543-601.
21. Song KH, Lee JY, Liu J, Lee SE, Park SJ and Kim DH. Prevalence of causative allergens on canine atopic dermatitis in Daejeon area. J Vet Clin 2005; 22: 26-30.
22. Wagner R. A retrospective survey of hyposensitisation therapy using low concentrations of alum - precipitated allergen. Proceedings of the 15th ESVD SCVD Congress. Maastricht, the Netherlands 1998: 167.
23. Willemse T. Atopic skin disease: a review and a reconsideration of diagnostic criteria. J Small Anim Pract 1986; 27: 771-778.
24. Willemse T, Bardagi M, Carlotti DN, Ferrer L, Fondati A, Fontaine J, Leistra M, Noli C, Ordeix L, Scarampella F, Schleifer S, Sinke J, Roosje P. Dermatophagoides farinae - specific immunotherapy in atopic dogs with hypersensitivity to multiple allergen: A randomized, double blind, placebo - controlled study. Vet J 2009; 180: 337-342.
25. Youn HY, Kang HS, Bhang DH, Kim MK and Hwang CY. Allergens causing atopic diseases in canine. J Vet Sci 2002; 3: 335-341.