

Poster #726

Determination of Allergenic Content in Dog Dander Obtained from Mixed (Mongrel) and Breed-Specific Dogs

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Abstract

Background:

Mammalian epithelial materials are used to produce allergenic extracts. Among these materials, dander isolated from hair contains a large amount of the major dog allergen, Can f 1. Dander also contains a minor allergen, albumin (Can f 3). The purpose of this study was to determine the allergenic content in dog dander derived from mixed (mongrel) and breed-specific dogs.

Methods:

Sixty dog dander lots were obtained over a three year period. Thirty lots (50.0%) were derived from mixed breeds, 28 lots (46.7%) were obtained from five specific breeds, and two lots (3.3%) were combined. Dander was extracted 1:10 (w/v) in Coca's solution for subsequent testing. Total protein content was measured using the Bradford assay, and Can f 1 and albumin contents were measured by ELISA. Non-parametric statistical analysis of the data was performed. The Mann-Whitney test was used to compare the test results obtained for dander derived from mixed and breed-specific dogs. The Spearman-rank correlation coefficient was used to ascertain the association among the parameters tested. A p value ≤ 0.05 was considered significant.

Results:

The 25th, median, and 75th percentiles for Can f 1 levels detected in mixed breed dander were 361.3 mg/g, 589.3 mg/g, and 789.9 mg/g, respectively compared with the respective values of 214.0 mg/g, 352.4 mg/g, and 609.0 mg/g measured in breed-specific dander ($p < 0.02$). The 25th, median, and 75th percentiles for albumin levels detected in mixed breed dander were 195.3 mg/g, 258.1 mg/g, and 361.0 mg/g, respectively compared with the respective values of 92.6 mg/g, 123.0 mg/g, and 169.2 mg/g measured in breed-specific dander ($p < 0.001$). The 25th, median, and 75th percentiles for total protein content did not significantly differ between dander types, and were 6.6 mg/g, 9.0 mg/g, and 11.5 mg/g, respectively in the total dander lots. None of the three parameters tested differed among the specific breeds tested. A low, but significant correlation was detected between Can f 1 and albumin levels in the total dander lots tested ($r = 0.372$, $p = 0.003$).

Conclusions:

Dander obtained from mixed breed (mongrel) dogs may contain greater allergen levels than that derived from breed-specific dogs. Environmental and genetic factors are likely responsible for this observation. This finding should be considered in the manufacture of dog allergenic extracts. Additional data of allergen content in dog specific breeds should be obtained.

Objective

The purpose of this study was to measure and compare the allergen and total protein contents detected in dander derived from mixed-breed (mongrels) and breed-specific dogs.

Materials and Methods

Dog Dander Lots

Total dander lots tested: 60, obtained from February 2006 through December 2009

- Derived from mixed-breed (mongrel) dogs: 30/60 (50.0%)
- Derived from breed-specific dogs (Table I): 28/60 (46.7%)
- Prepared by mixing dander derived from breed-specific dogs: 2/60 (3.3%)

Table I: Breed specific lots tested (N=28)

Breed	# lots (%)
Spaniel	6 (21.4)
Poodle	6 (21.4)
Schnauzer	6 (21.4)
Terrier	6 (21.4)
Alsatian	3 (10.7)
Collie	1 (3.6)

Dander lots according to the collector:

- Provided by local collectors: 32/60 (53.3%)
- Provided by a specialized collector: 34/60 (40.0%)
- Combined: 2/60 (3.3%)
- Not recorded: 2/60 (3.3%) – Those lots were not included in the analysis

Most mixed-breed lots were provided by local collectors, and most breed-specific lots by a specialized collector (Table II).

Table II: Number of lots provided by local collectors and by a specialized collector (N=56)

Collector	Breed	
	Mixed	Specific
Specialized	3	21
Local	25	7

Testing

Can f 1 and albumin (Can f 3) levels were measured by indirect ELISA using alkaline phosphatase as the substrate and the following antibodies and standards:

- Antibodies and standard to measure Can f 1: Indoor Biotechnologies, Charlottesville, VA
- Antibodies to measure albumin: Greer Laboratories, Inc., Lenoir, NC
- Standard to measure albumin: Sigma-Aldrich, Hercules, CA

Total protein content was measured by the Bradford method, using BSA as the standard.

Statistical Analysis

- Software:** Prism™ software (Prism Software Co., Irvine, CA).
- Analysis performed:**
 - Descriptive statistics
 - Kolmogorov-Smirnov test: to ascertain the normality of the distributions
 - Mann-Whitney test: to compare the values of the different parameters obtained between mixed and specific breeds
 - Kruskal-Wallis test: to compare the values of the parameters obtained among specific breeds
 - Spearman-rank correlation coefficient: to evaluate the potential association among the different parameters tested

Note: A p value less than 0.05 was considered significant

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Introduction

The prevalence of sensitization to the major allergen derived dogs, Can f 1, in subjects with clinical reactivity to dog allergens is approximately 60-70%. The reported prevalence of sensitization to dog albumin (Can f 3) in these subjects is highly variable, ranging from 6% to 48%, depending on the study reviewed.

Mammalian allergens can occur in a variety of sources, and different raw materials are used for the production of dog allergenic extracts. These materials include dander, hair, epithelium, pelt, skin scrapings, and hides. The dander, or particles from the pelt attached to the hair, is the vector directly responsible for the allergic response. Dog allergen extracts derived from dander isolated from hair typically contain more Can f 1 and lesser albumin than epithelia derived extracts.

Among other raw materials derived from dogs, Greer collects hair to obtain dander for the production of dog allergenic extracts for diagnosis and treatment of dog allergy. Each dander lot is derived from multiple dogs provided by local collectors and by a specialized collector.

Dog allergen production varies among dogs. A number of factors, including breed, hormonal status, and the presence of skin disorders, have been determined to influence Can f 1 levels in individual dogs.

Results

Can f 1

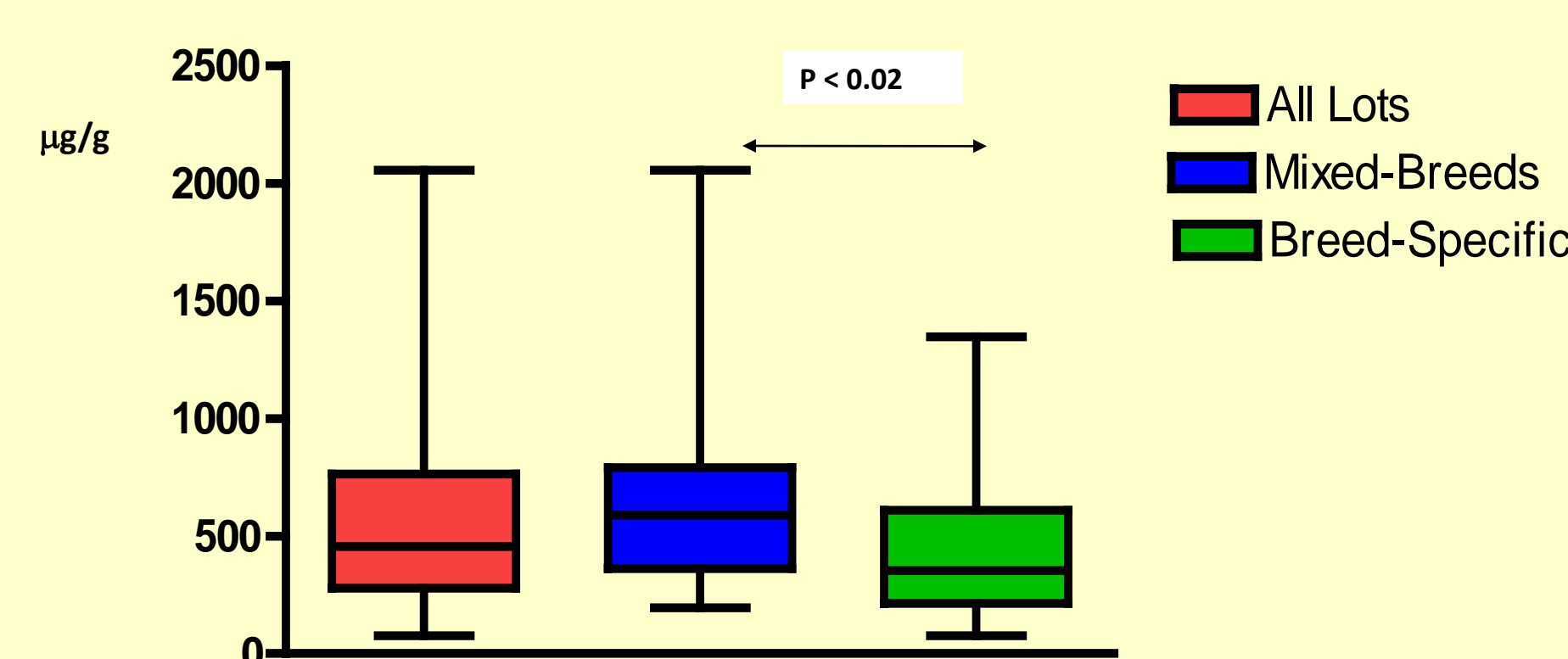
Can f 1 level distribution

- Range of values detected: 75.4 mg/gram (obtained for an Alsatian breed) to 2,056.0 mg/gram (for dander derived from a mixed breed)
- Median: 455.4 mg/gram

Can f 1 according to dog breeds

- The Can f 1 level in dander derived from mixed-breed (mongrel) dogs was significantly greater than that detected in dander derived from breed-specific dogs, $p < 0.02$, (Figure 1).
- The Can f 1 levels detected in dander derived from breed-specific dogs did not significantly differ among them

Figure 1: Can f 1 level distributions in the dander lots tested



Note: The horizontal lines delimiting the boxes correspond to the 75th, 50th, and 25th percentiles (from top to bottom)

Albumin (Can f 3)

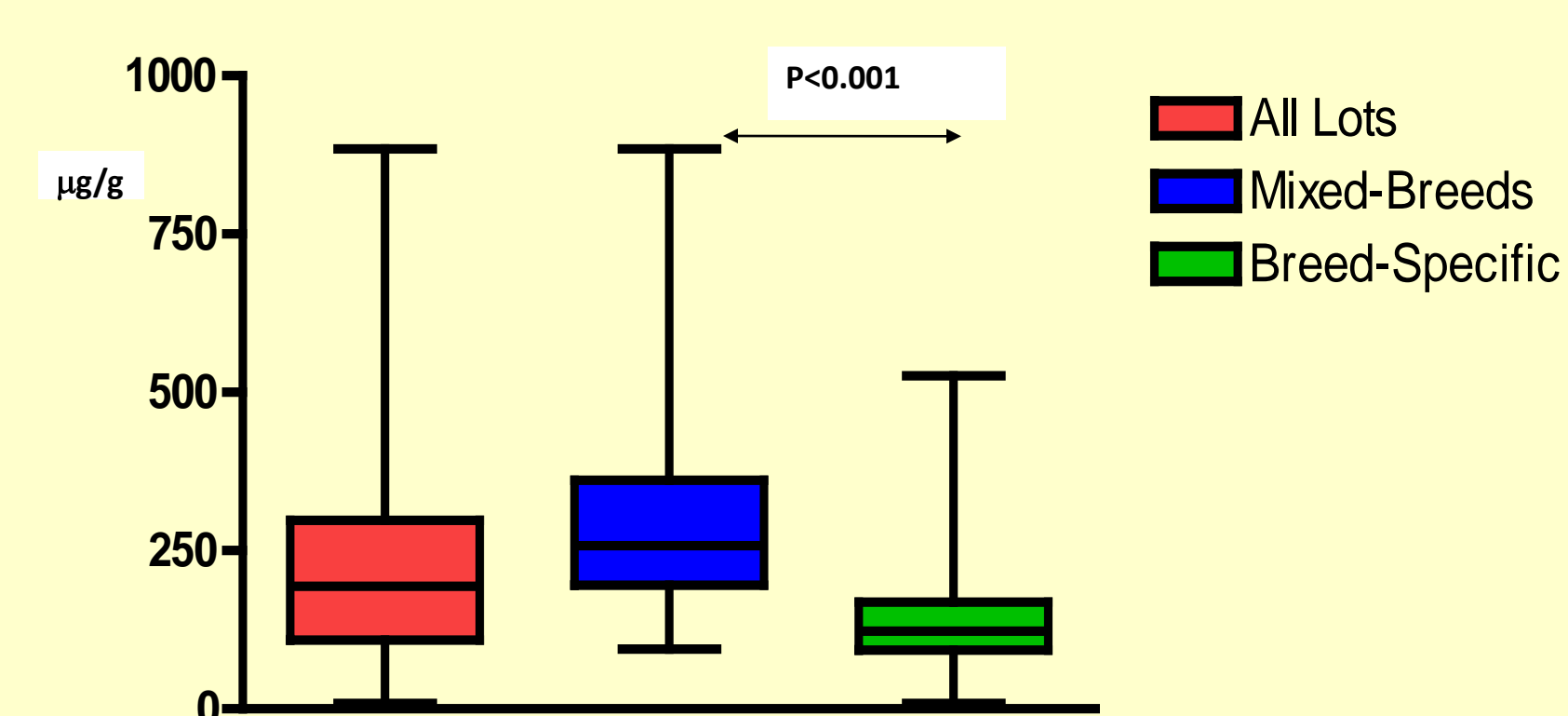
Albumin level distribution

- Range of values detected: 9.1 mg/gram (obtained for the Alsatian breed with the lowest levels of Can f 1) to 884.5 mg/gram (for dander derived from a mixed breed)
- Median: 193.4 mg/gram (for dander derived from a mixed breed)

Albumin levels according to dog breeds

- As for Can f 1, the albumin level in dander derived from mixed breed dogs was significantly greater than that detected in dander derived from breed-specific dogs, $p < 0.001$, (Figure 2).
- Albumin levels detected in dander derived from breed-specific did not significantly differ among them.

Figure 2: Albumin level distributions in the dander lots tested



Note: The horizontal lines delimiting the boxes correspond to the 75th, 50th, and 25th percentiles (from top to bottom)

Bradford Protein Content

Bradford protein content distribution

- Range of values detected: 2.90 mg/gram (obtained for a poodle breed) to 15.31 mg/gram (obtained for another poodle breed).
- Median: 9.01 mg/gram

Bradford protein content according to dog breeds

- No significant differences between the Bradford protein content measured in dander derived from mixed-breed and breed-specific lots, or among specific breeds

Correlation among The Parameters Tested

Not significant for the total lots tested or according to the breed type

- Can f 1 and albumin – total protein content

Significant ($r=0.372$, $p=0.003$)

- Can f 1 – albumin in the total lots tested

Test Results according to The Collector whom Provided the Dog Hair

- Can f 1 and albumin content in dander obtained by local collectors was significantly greater than the levels detected in dander provided by the specialized collector ($p < 0.001$).
- The fact that mixed-breed dogs contain significantly greater levels of the two allergens is likely responsible for the different concentrations detected between the two dander sources.

Conclusions

- A large variation in allergen and protein contents were observed among dander lots.
- Dander lots derived from mixed breed dogs contain greater allergen levels than those derived from breed-specific dogs, supporting the role of genetic and/or environmental components in dog allergen production.
- Mixed breed dogs may be more suitable than breed-specific dogs for dander production.
- While dog breed has been associated with allergen production, no significant differences among dog-specific breeds were observed in this study, perhaps because of sample size limitations.
- Additional data should be obtained for further analysis. This information could be relevant in manufacturing and in the selection of specific breeds for dander production, and could also provide a better understanding of the factors involved in dog allergen production.