Common Antigens in Elimination Diets (Parts 1 & 2)

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The term: 'food allergy' and 'food intolerance' have been confused and used interchangeably to describe adverse food reactions but in fact reactions to a food can be delineated into either an immunologic or non-immunologic reaction.



A food allergy is an abnormal response to ingested food or food additive which is often cited by owners but rarely documented to be true. Such an allergenic reaction may manifest with dermatologic signs of non-seasonal pruritic dermatoses that may include bilateral otitis and /or gastrointestinal signs of gastritis, enteritis and/or colitis. It can be difficult to distinguish between food intolerance vs. food allergy in a patient with only GI signs. However, a patient with dermatological signs can have a food allergy (Gaschen, et al., 2011)

Currently, the definitive diagnosis of food allergy in dogs is based on results of an elimination trial followed by a food challenge test. After a successful dietary elimination trial with resolution of skin and/or gastrointestinal signs a food challenge is performed. The patient is fed the previous diet, which should elicit the original symptoms generally within 2-14 days. Then re-feeding the successful food trial diet should resolve the signs if the patient has a true food allergy (Scott et al., 2001).

It is hypothesized that immunological food reactions develop when a food specific IgE antibody on a mast cell binds with a food antigen. This binding initiates mast cell release of potent inflammatory mediators and cytokines (Sampson, 2004). It is believed that most food allergies are Type I or IgE mediated but there is some evidence to suggest Type III and IV hypersensitivity reactions may be involved (Verlinden et al., 2006).

This immunologic response to a food borne antigen must be a protein or a glycoprotein in order to initiate an antigen/antibody complex and the resulting immune cascade that causes inflammation in the skin or gastrointestinal tract (Roudebush et al., 2000). The meat proteins commonly used in pet foods are beef, poultry, egg, fish and pork. The most common grain proteins are soybean, rice, corn and wheat (Cowell et al., 2000). Dogs have been reported to have food allergies to beef, dairy products, wheat, lamb, egg, chicken and soy (Roudebush et al., 2000). Food allergies to rice and corn protein have been reported (Scott et al., 2001, Jackson 2002) but not to a starch or pure carbohydrate.

When a pet develops a food allergy it is most often after chronic exposure to one of these common dietary proteins. Hence, diets used in diagnostic elimination trials must contain only novel protein sources, i.e. food proteins never before consumed by the patient. The diet also cannot contain any amount of the more commonly used protein ingredients. Known and unknown consumption of common allergen proteins during the food trial nullify or confound the results of such a trial and preclude an accurate diagnosis. (Roudebush et al., 2000). Intentional or known consumption of a suspected allergen occurs when the owner gives an unapproved treat during a food trial; whereas an unknown or unintentional consumption of an allergen would occur when a dog food contains ingredient residues from other products, i.e. ingredient cross contamination during manufacturing.

General practitioners, gastroenterologists and dermatologists use dietary elimination trials to diagnose food allergies in dogs. These trials are lengthy (4–13 weeks) and require a client's strict adherence to the prescribed food. There has been an increase in pet owner interest in using over the counter (OTC) commercial diets sold through retail outlets for a diagnostic dietary elimination trial because of price and convenience (Scott et al., 2001). General practitioners and some veterinary specialists have acquiesced to such client requests, not fully understanding the difference and value in product quality control between veterinary therapeutic and OTC commercial lines of pet foods. The pet owner may select OTC diets based on the name of the product, e.g. venison and sweet potato, which ensures nothing more than the product will contain at least 3% of those ingredients (Cowell et al., 2000). Pet owners and veterinarians also make the assumption that if food proteins or isolates of a food protein are not named in the product ingredient list, then the product does not contain those food proteins and therefore, is a suitable diet to be used in a diagnostic elimination trial (Scott et al., 2001). We conducted two studies ELISA testing for common food proteins in various diets to determine the suitability of such diets in a food elimination trial.

Additionally, we recognized there are many common possible confounders of dietary elimination trials in treats, snacks and table foods. It is a general recommendation that flavored over the counter (OTC) products (Pet-Tabs, Pill Pockets) and flavored medications (heartworm and arthritis preventatives) should be avoided during dietary elimination trials because these products may contain common proteins that would confound the results of the trial although this has yet not been well documented to date. We performed a third set of ELISA tests on products commonly used during a food elimination trial in attempt to document the food proteins they may contain.

- 1. ELISA Testing for Common Food Antigens in Dry Dog Foods
 - a. Objective: to test whether novel or exotic protein named (e.g., venison) OTC diets contained common food proteins, specifically soy, poultry, or beef, that were not identified on the label.
 - b. Method: An amplified, double sandwich ELISA test of soy, poultry and beef proteins were performed by an independent accredited food and feed laboratory. ELISA testing for soy, poultry, and beef protein was completed on 100 gram sample of each diet four control diets: three with at least one of these proteins named in the ingredient list and one veterinary therapeutic diet with none listed on the label. Additionally, four OTC venison dry dog foods with no soy or beef named in the ingredient list were similarly tested for soy, poultry and beef proteins. Only two OTC venison diets with no poultry in the ingredient list could be located; hence two of the four OTC venison products did list a poultry ingredient on the label.
 - c. Results: ELISA assay for soy and beef protein on the four control diets performed as expected; however, the poultry protein assay was unreliable due to false negatives. Three of the four OTC venison canine dry foods with no soy products named in the ingredient list were ELISA positive for soy. One OTC venison diet tested positive for beef protein which was also not listed as an ingredient on the label. None of the four OTC venison diets tested were suitable for a diagnostic elimination trial as they all contained common pet food proteins, some of which were readily identifiable on the label and some (beef and soy) that were only detected by ELISA.
- 1. ELISA testing for soy antigens in dry dog foods used in dietary elimination trials
 - a. Objective: to determine if: 1) four over-the-counter (OTC) dry dog foods carrying a 'made with no soy' claim and 2) seven veterinary therapeutic dry dog foods designed for food elimination trials were suitable for a canine soybean elimination trial.
 - b. Method: A 100 gram sample of each test diet plus a soy positive and negative control diet were submitted for ELISA testing to an outside independent food laboratory. The ELISA test is quantitative for soy flour protein concentrations between 2.5 and 25 ppm. The positive control diet contained >25 ppm soy protein antigens and the negative control < 2.5 ppm soy.</p>
 - c. Results: Three of the four OTC 'no soy' claiming diets were positive for soy antigen; two contained >25 ppm. Four veterinary therapeutic diets had less than the lower detectable limit of soy protein. Two veterinary therapeutic hydrolyzed soy diets were positive (>2.5 ppm): one diet had a natural flavor (with a soy carrier) contained >25 ppm soy and the other diet contained 4.6 ppm with no soy ingredients listed.
- 2. ELISA Testing for Soy, Pork and Beef Antigens in Common Confounders of Dietary Elimination Trials
 - a. Objective: to determine if there are protein antigens (soy, pork and beef) in common OTC products and/or flavored medications, i.e. confounders fed to dogs during an elimination diet trial.
 - b. Method: 100 grams of seven different products (three OTC products and four veterinary medications) tested for the presence of soy, pork and beef antigens in addition to positive and negative controls were submitted for ELISA testing to an outside independent food laboratory.
 - c. Results: All OTC test products produced ELISA results in agreement with their ingredient list. The results of ELISA testing of veterinary therapeutic products did not agree with the ingredient lists/product inserts. After directly contacting the manufacturers of the veterinary medications, they were forthcoming with the proprietary protein ingredients but these were surprisingly not always in agreement with the ELISA test results.

Conclusions

- 1. If the four OTC venison products selected in the first study are representative of OTC products, then OTC venison dry dog foods should not be used for elimination trials in suspected food allergy patients.
- 2. OTC dog food diets that claim to contain 'no soy' may actually contain high concentrations of soy protein antigen and therefore should not be considered for soy elimination trials.
- 3. The veterinary therapeutic diets chosen for a soy elimination diet trial needs to be carefully selected to ensure no soy protein present in the diet.
- 4. Continue to recommendation that flavored over the counter (OTC) products (Pet-Tabs, Pill Pockets) and flavored medications (heartworm and arthritis preventatives) be avoided during dietary elimination trials because these products do contain common proteins that will confound the results of the trial.

- 5. Gelatin capsules may contain both beef and/or pork proteins and therefore gelatin capsules of any product should not be fed during a dietary elimination trial.
- 6. Veterinarians should contact manufacturers of oral veterinary therapeutics prior to prescribing them during a dietary elimination trial to determine the proprietary protein ingredients which need not be listed on the package ingredient list.

References

Cowell, C. S.; Stout, N. P.; Brinkmann, M. F.; Moser, E. A.; Crane, S. W., 2000: Making of commercial pet foods. In: M. S.Hand, C. D.Thatcher, R. L.Remillard, P.Roudebush (eds), Small Animal Clinical Nutrition, 4th edn. Walsworth Publishing Company, Marceline, USA, pp. 127–146.

Gaschen, F.P., Merchant, S.R., 2011: Adverse food reactions in dogs and cats. Vet Clin Small Anim 41;361-379.

Jackson, H.A.; Hammerberg B. 2002: Evaluation of a spontaneous canine model of immunoglobulin E-mediated food hypersensitivity: dynamic changes in serum and fecal allergen-specific immunoglobulin E values relative to dietary change. Comp Med 52;318-324.

Roudebush, P.; Guilford, W. G.; Shanley, K. J., 2000: Adverse reactions to food. In: M. S.Hand, C. D.Thatcher, R. L.Remillard, P.Roudebush (eds), Small Animal Clinical Nutrition, 4th edn. Walsworth Publishing Company, Marceline, USA, pp. 432–446.

Sampson, H. A., 2004: Update on food allergy. Journal of Allergy Clinical Immunology 113, 805-819.

Scott, D. W.; Miller, W. H.; Griffin, C. E. eds. 2001: Canine food hypersensitivity. In: Muller and Kirk's Small Animal Dermatology, 6th edn. WB Saunders Company, Philadelphia, PA, USA, pp. 615–627.

Verlinden, A.; Hesta, M.; Millet, S.; Janssens, G. P., 2006: Food allergy in dogs and cats: a review. Clinical Reviews in Food Science and Nutrition. 46, 259–273.