

Use of histamine phosphate for the interpretation of intradermal skin tests in dogs

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INTRADERMAL skin tests have been used in veterinary and human medicine for many years to identify IgE-mediated hypersensitivity to allergens (Hillier and DeBoer 2001). Positive and negative control reactions are used for the interpretation of these tests. A skin test reaction is defined as positive when its diameter (the mean of the maximum diameter and the diameter perpendicular to it) is equal to or larger than the mean diameter of the positive and negative controls (Reedy and others 1997, Hillier and DeBoer 2001). Histamine is frequently used as a positive control for the interpretation of skin tests. However, exogenous histamine presents some disadvantages: histamine is not the only mediator of allergic reactions (Casale and others 1984), it elicits a maximum skin response within a time period different from that of the allergens (Voorhorst 1980, Reedy and others 1997), it can provoke widely variable responses, especially at low concentrations (Taudorf and others 1985), it does not show a good correlation with allergens when the dose of both is increased (Malling 1984), and the size of the skin reaction depends on whether the patient is normal, weakly allergic or severely allergic (Malling 1987).

The positive control solution used most commonly is 0.01 or 0.001 per cent histamine phosphate (Reedy and others 1997, Scott and others 2001). Hensel and others (2004) compared these two concentrations in 30 normal dogs and concluded that the optimal concentration for use in intradermal skin testing in dogs is 0.01 per cent, or 1:10,000 (w/v). However, the authors have observed that the response to 0.01 per cent histamine phosphate tends to vary with the bodyweight and degree of sensitisation of the dogs. Therefore, the aim of this study was to determine whether the reactions to 0.01 per cent histamine in dogs are influenced by these two variables.

Thirty-two healthy dogs without a history or clinical signs of any type of allergic dermatitis were divided into two groups according to their bodyweight and then tested. Group 1 consisted of 16 dogs, with a mean age of three years and mean (sd) bodyweight of 9.5 (3.24) kg (range 5 to 15 kg). Group 2 consisted of 16 dogs, with a mean age of 3.4 years and bodyweight of 33 (4.31) kg (range 25 to 40 kg). In addition, a third group of 16 dogs was studied, with a mean age of 3.9 years and bodyweight of 9.68 (4.75) kg (range 4.5 to 19 kg); all the dogs in group

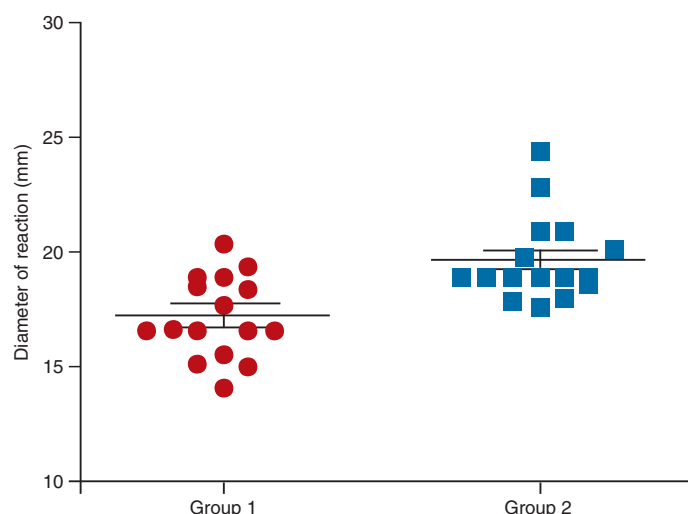


FIG 1: Skin reactions to injections of 0.01 per cent histamine phosphate in dogs with small (group 1) or large (group 2) bodyweight

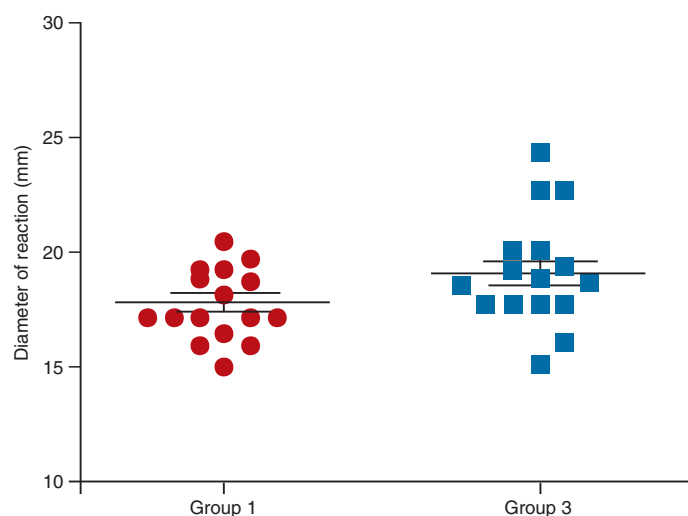


FIG 2: Skin reactions to 0.01 per cent histamine phosphate in a group of clinically healthy dogs (group 1) and a group of dogs of similar bodyweight diagnosed with atopic dermatitis (group 3)

3 had a clinical diagnosis of atopic dermatitis according to the criteria suggested by Prélard and others (1998), and other skin diseases characterised by pruritus had been excluded.

In all the dogs, a volume of 0.05 ml 0.01 per cent histamine phosphate (FDA-Allergenic) was inoculated intradermally using an insulin syringe and a 8 mm, 0.3 mm internal diameter needle. The lateral region of the chest was chosen as the test site and shaved with a number 40 shear blade. The dogs in group 3 required sedation with 0.5 mg/kg xylazine; these dogs were subjected to intradermal skin testing using extracts of house dust mites and storage mites. The dogs in groups 1 and 2 did not require sedation and were restrained physically during the procedure.

The *t* test was used to evaluate significant differences in the mean skin reaction values obtained with 0.01 per cent histamine phosphate between groups 1 and 2, and between groups 1 and 3. All analyses were performed using GraphPad PRISM for Windows v 5.01. *P* < 0.05 was considered significant.

The mean skin reaction to histamine phosphate in the dogs in group 1 was 17.16 mm in diameter (range 14.0 to 20.5 mm), in group 2 it was 19.72 mm (range 17.5 to 24.5 mm), and in group 3 it was 18.78 mm

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(range 14.0 to 25.0 mm). The difference between groups 1 and 2 was significant ($P=0.005$) (Fig 1). The difference between group 1 and group 3 was not significant ($P=0.07$) (Fig 2).

The difference between the diameters of the skin reactions in the dogs in group 1 and group 2 indicate that bodyweight does influence the reaction to 0.01 per cent histamine phosphate. Thus, when 0.01 per cent histamine phosphate is used as the control for interpreting skin reactions to allergenic extracts, veterinarians should consider the possibility that very heavy dogs may require more concentrated allergenic extracts to show responses that are judged to be positive. However, it is not clear whether heavier dogs also react more intensely to allergenic extracts, and whether any variations in reaction are proportional to the variation observed for the reaction to histamine phosphate. This should be tested by comparing the diameters of reactions to both allergenic extracts and histamine in dogs of different bodyweights.

No significant difference in the reaction diameters was observed between group 1 and group 3, although the mean reaction to histamine was higher in group 3. It is possible that the number of dogs in both groups was insufficient to identify an association between the degree of sensitisation and the diameter of the skin reactions in dogs, as demonstrated by Malling (1987) in human beings. Due to the urban and cultural characteristics of the region where the dermatological tests were carried out, the number of large, heavy dogs available for the study was very small. In addition, most of these dogs were kept outside, and therefore rarely exposed to indoor allergens such as house dust mites. The most prevalent allergic disease in dogs kept outdoors in Rio de Janeiro is flea allergy dermatitis. Over the past two years the authors saw only nine atopic dogs weighing between 25 and 40 kg; these dogs underwent skin

tests using extracts of house dust mites (the authors test only for house dust mite and storage mite allergy). The mean diameter of the reactions provoked by 0.01 per cent histamine in these nine dogs was 20.22 mm (range 18.0 to 25.0 mm). Further studies involving large, heavy dogs are necessary to test this hypothesis.

References

- CASALE, T. B., SHELHAMER, J. H., PARRILLO, J. E. & KALINER, M. A. (1984) Dopamine inhibition of histamine-mediated cutaneous responses. *Journal of Allergy and Clinical Immunology* **73**, 837-841
- HENSEL, P., AUSTEL, M., MEDLEAU, L., ZHAO, Y. & VIDYASHANKAR, A. (2004) Determination of threshold concentrations of allergens and evaluation of two different histamine concentrations in canine intradermal testing. *Veterinary Dermatology* **15**, 304-308
- HILLIER, A. & DEBOER, D. J. (2001) The ACVD task force on canine atopic dermatitis (XVII): intradermal testing. *Veterinary Immunology and Immunopathology* **81**, 289-304
- MALLING, H.-J. (1984) Skin prick testing and the use of histamine references. *Allergy* **39**, 596-601
- MALLING, H.-J. (1987) Quantitative skin prick testing. Dose-response of histamine- and allergen-induced wheal reactions. *Allergy* **42**, 196-204
- PRÉLAUD, P., GUAGUÈRE, E., ALHAIDARI, Z., FAIVE, N., HÉRIPRET, D. & GAYERIE, A. (1998) Réévaluation des critères diagnostiques de la dermite atopique. *Revue de Médecine Vétérinaire* **149**, 1057-1064
- REEDY, L. M., MILLER, W. H. & WILLENME, T. (1997) Allergy testing. In *Allergic Skin Diseases of Dogs and Cats*. 2nd edn. W. B. Saunders. pp 83-115
- SCOTT, D. W., MILLER, W. H. & GRIFFIN, C. E. (2001) Skin immune system and allergic skin diseases. In *Muller and Kirk's Small Animal Dermatology*. 6th edn. W. B. Saunders. pp 543-666
- TAUDORF, E., MALLING, H.-J., LAURSEN, L. C., LANNER, A. & WEEKE, B. (1985) Reproducibility of histamine skin prick test. Inter- and intravariation using histamine dihydrochloride 1, 5, and 10 mg/ml. *Allergy* **40**, 344-349
- VOORHORST, R. (1980) Perfection of skin testing technique. A review. *Allergy* **35**, 247-250