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# Skin testing

John Oppenheimer, MD,\* and Harold S. Nelson, MD†

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**Objective:** To provide the reader with a relevant review of the literature regarding skin testing in the allergist's office.

**Data Sources:** A PubMed search for the years 1970 through 2005 was performed using the following keywords: *allergy skin testing*, *skin prick testing*, and *intradermal skin testing*.

**Study Selection:** Articles that highlighted aspects of sentinel to clinical allergists' use of skin testing in the office, such as methods of skin testing, intradermal vs skin prick testing, skin test devices, and methods of expressing skin test results, were selected for further review.

**Results:** Skin testing remains the central test to confirm an allergic response. It is minimally invasive and when performed correctly has good reproducibility. Results are easily quantifiable and correlate well with end organ challenge. It is imperative however that technicians who perform the skin tests and the clinicians who order or interpret these tests understand the characteristics of the specific tests they are administering. It is also important that the clinician express skin test results in a manner that allows easy interpretation by another physician.

**Conclusions:** Allergists must consider controllable variables that affect skin test results and their interpretation. When not considered, they may be responsible for some of the inaccuracies associated with allergy skin testing.

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## INTRODUCTION

Credit for the first skin testing device goes to Charles H. Blackley, a Manchester homeopathic physician with allergic rhinitis. In 1865 he abraded a quarter-inch area of his skin with a lancet. He then applied grass pollen grains that had been placed on a piece of wet lint and covered the scarified area with an occlusive bandage. This resulted in intense itching and a large cutaneous response.<sup>1</sup> Even today in the allergist's office, skin testing remains the central test to confirm an allergic response. This is not surprising, because it is minimally invasive and when performed correctly has good reproducibility. In theory, one could apply allergen directly to a mucous membrane such as the eyes, nose, or bronchi, but this is much more time-consuming and has greater inherent risk. Also, skin testing is easily quantifiable and can allow the evaluation of multiple allergens at one session, which is not possible with mucosal challenge. Skin testing has demonstrated good correlation with results of nasal challenge<sup>2</sup> and bronchial challenges (when allowance is made for nonspecific airway responsiveness).<sup>3</sup> It is imperative however that the technician performing the skin tests and the clinician ordering or interpreting these tests be aware of the factors that can affect the results of the tests. These factors include type of skin testing, device used, placement of tests (location and adjacent testing), the particular extracts being used, and the potential confounder of medications that may suppress skin test response. These issues have been reviewed

elsewhere in greater detail.<sup>4–6</sup> It is of paramount importance that the clinician consider the positive and negative predictive value of the tests performed and always rely on testing as an adjunct to history and physical examination when making the diagnosis of allergic disease. Lastly, allergy patients may have to change their allergist for numerous reasons. Thus, it is important that prior allergy testing records be interpretable by the receiving physician.

This article aims to provide the reader with a relevant review of the literature regarding skin testing in the allergist's office. We performed a PubMed search for the years 1970 through 2005 using the following keywords: *allergy skin testing*, *skin prick testing*, and *intradermal skin testing*. Articles that highlighted aspects sentinel to clinical allergists' use of skin testing in the office, such as methods of skin testing, intradermal vs skin prick testing, skin test devices, and methods of expressing skin test results, were selected for further review.

## METHODS OF SKIN TESTING

Skin tests involve the introduction of allergen through a break in the skin. In the modern allergy office, this is achieved via skin prick or puncture testing and intradermal testing. In the past, the scratch method<sup>7</sup> was also used. In the scratch method, a linear scratch was made without drawing blood. The scratch could either be performed first, with the extract then dropped on the abraded skin, or be made through a drop of extract. Scratch testing has largely been abandoned due to greater discomfort, poorer reproducibility, and the possibility of leaving multiple linear depigmented areas for some time afterward.<sup>7</sup>

Intradermal testing is more sensitive than skin prick testing. For equivalent reactions, the extract for skin prick testing must be at least 1,000-fold more concentrated.<sup>7</sup> Direct comparisons indicate that intradermal testing is more reproducible

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\* Department of Internal Medicine, Section of Allergy and Immunology, Morristown Memorial Hospital, Morristown, New Jersey, and Division of Allergy, New Jersey Medical School, Newark, New Jersey.

† Division of Allergy, National Jewish Medical and Research Center, Denver, Colorado.

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Table 1. Size of Wheals That Are Larger Than 99% of the Wheals With Saline Using the Same Device on the Patient's Back Performed by the Same Operator\*

| Device                                               | Wheal size, mm |
|------------------------------------------------------|----------------|
| Devices for which a 3-mm wheal would be significant  |                |
| Quintest (HS) puncture                               | 0              |
| Smallpox needle (HS) prick                           | 0              |
| DuoTip (Lincoln) prick                               | 1.5            |
| Lancet (HS) puncture                                 | 2.0            |
| Lancet (ALK) puncture                                | 3.0            |
| DermaPICK II (Biomedix)                              | 0              |
| Devices for which a >3-mm wheal would be significant |                |
| DuoTip (Lincoln) twist                               | 3.5            |
| Bifurcated needle (ALO) prick                        | 4.0            |
| MultiTest (Lincoln) puncture                         | 4.0            |
| Bifurcated needle (ALO) puncture                     | 4.5            |
| Quick Test (Pantrex)                                 | 4.0            |
| Greer Track (Greer)                                  | 3.5            |

Abbreviations: HS, Hollister Steir; Greer, Greer Laboratories; Lincoln, Lincoln Diagnostics; ALK, ALK America; ALO, Allergy Labs of Ohio.

\* Data are from Nelson et al.<sup>12-14</sup>

than percutaneous testing.<sup>7</sup> Despite this, many factors favor the percutaneous test as the routine for allergy testing. These factors include economy of time, patient comfort, and safety. Percutaneous testing has a steeper dose response curve and therefore is more useful in titrated skin testing. Also, percutaneous testing allows use of extract in 50% glycerin and thus greater extract stability. Intracutaneous testing cannot use this diluent, because it results in an irritant or false-positive response.<sup>8,9</sup> Of paramount importance is that studies have demonstrated that percutaneous response correlates much better with clinical allergy.

## CLINICAL UTILITY OF INTRACUTANEOUS VS PERCUTANEOUS METHOD

Although the intracutaneous test, at the strength customarily performed, is more sensitive, it is questionable whether this increased sensitivity is clinically necessary or simply increases the chance of a false-positive response. The prick test, performed with potent extracts, produces a positive result in many patients who do not have a personal or even a family history of allergy.<sup>10</sup> A number of studies have addressed the clinical usefulness of intracutaneous testing and deserve review.<sup>8,10,11</sup>

Two recent studies examined the intracutaneous test as a predictor of symptoms with natural exposure to the allergen.<sup>10,11</sup> In the first study, 4 groups were compared: 3 of the groups had a history of seasonal allergic rhinitis during grass season, 1 with positive skin prick test results to timothy, 1 with a negative skin prick test result but a positive intradermal test result to timothy, and 1 with both skin prick and intracutaneous test results negative for timothy. The fourth group was a nonallergic control.<sup>10</sup> On the basis of nasal challenge with timothy grass pollen, allergic reactions were present in 68% of those with positive skin prick test results to timothy and none of the nonallergic controls. In both the

groups with negative skin prick test results to timothy, 11% had positive results whether intradermal skin test results to timothy were positive or negative. Patients were then followed up though the grass pollen season. Their symptom scores, recorded in a diary, were examined for a correlation with grass pollen counts. A positive correlation was present in 64% of those with positive skin prick test results and none of the nonallergic controls. In the seasonally symptomatic group of patients with negative skin prick test results, positive correlation of symptoms and pollen count was present in 22% of those with a positive intracutaneous test result and 21% of those with a negative intracutaneous test result to timothy. Both criteria for allergy to timothy, a positive nasal challenge result and a correlation between symptoms and grass pollen counts, were met in 46% of those with positive skin prick test results, but in none of the other 3 groups. Thus, under the conditions of this study, the presence of a positive intradermal skin test response to timothy in the presence of a negative skin prick test response to timothy did not indicate the presence of clinically significant sensitivity to timothy grass.<sup>10</sup>

In the second study, patients were challenged with cat exposure for 1 hour.<sup>11</sup> Both positive skin prick test results and radioallergosorbent test results to cat were highly predictive of the development of symptoms on exposure to the cat room. Patients with a negative skin prick test result were just as likely to have a positive challenge result if they had a negative (31%) compared with a positive (24%) intracutaneous skin test result. The authors concluded that, at least with regard to cat allergy, these results strongly suggest that major therapeutic decisions, such as environmental control or immunotherapy, should never be based on a positive intracutaneous skin test result alone.<sup>11</sup>

Both of these studies were performed in adults and both relied on skin testing with potent allergens (timothy grass and cat). Application of these results to other less potent allergens

Table 2. Semiquantitative Reporting of Skin Test Results\*

| Criteria to read test results                 | Results                                                                       |
|-----------------------------------------------|-------------------------------------------------------------------------------|
| Skin prick or puncture tests                  |                                                                               |
| Negative                                      | No reaction or no different from control                                      |
| 1+                                            | Erythema less than a nickel in diameter                                       |
| 2+                                            | Erythema greater than a nickel in diameter                                    |
| 3+                                            | Wheal with surrounding erythema                                               |
| 4+                                            | Wheal with pseudopods and surrounding erythema.                               |
| Intracutaneous tests when control $\geq 2$ mm |                                                                               |
| Negative                                      | No different from control                                                     |
| 1+                                            | Wheal 1 to 2 times control or definite erythema greater than a nickel in size |
| 2+                                            | Wheal 2-3 times control                                                       |
| 3+                                            | Wheal $>3$ times control                                                      |
| 4+                                            | Wheal with pseudopods                                                         |
| Intracutaneous tests when control $<2$ mm     |                                                                               |
| Negative                                      | No difference from control                                                    |
| 1+                                            | 3- to 4-mm wheal with erythema or erythema greater than a nickel in size      |
| 2+                                            | 4- to 8-mm wheal without pseudopods                                           |
| 3+                                            | $>8$ -mm wheal without pseudopods                                             |
| 4+                                            | Wheal with pseudopods and erythema                                            |

\* Data are from Vanselow.<sup>38</sup>

(eg, dog) and to younger patients (especially infants) requires clinical judgment regarding what action should be taken as a result of the information gleaned.

## SKIN TESTING DEVICES

Although intracutaneous skin tests are only performed using a hypodermic syringe and needle, percutaneous tests may be performed with a variety of devices.<sup>12-14</sup> These devices differ in whether they are used to prick or puncture (some being used both ways). Some have a single stylus with a single or several points and are used either to prick or puncture through a drop of extract or to carry a drop of extract from the extract bottle so that application of extract and the puncture occur in one step. Devices with multiple heads allow up to 8 tests to be performed with one application. Generally, the multiple-headed skin test devices are designed to first be dipped into the extract bottles, then applied to the skin in one step. The devices for percutaneous testing vary in the degree of trauma that they impart to the skin. Therefore they differ in the size of positive reactions and also in the likelihood of producing a reaction at the site of the negative control. Thus, they require different criteria for what constitutes a positive reaction (Table 1). Comparisons of percutaneous devices have been reviewed elsewhere in greater detail.<sup>6,9,12-19</sup>

Allergy skin testing has recently come under the scrutiny of the Occupational Safety and Health Administration (OSHA) of the US Department of Labor. In 1995, OSHA alerted its field personnel to the possible safety and health risk that may arise with the practice of using one device per person and wiping the device between tests.<sup>20</sup> OSHA considered this practice to have the potential for a blood-borne

pathogen exposure incident should the technician unintentionally prick himself or herself with the device in wiping it. The implications of this notice have led many allergists to abandon the use of solid-bore needles for percutaneous testing, resulting in greater use of the newer devices that are disposed of after each application of a test.

## PLACEMENT OF ADJACENT SKIN TESTS

There are 2 reports, both describing intracutaneous testing with Hymenoptera venom, that indicate that false-positive test results can result from an adjacent positive reaction.<sup>21,22</sup> In light of this, the influence of large positive reactions to histamine or to allergen on adjacent skin prick tests was prospectively studied with the skin prick test method.<sup>23</sup> These results were different from previous reports, because there was no evidence of false-positive skin prick test results attributable to the large adjacent reactions, even when the test sites were only separated by 2 cm. Thus, it appears that the augmentation that has been observed may be limited to intracutaneous testing or perhaps to the active constituents in the Hymenoptera venom.

## EXPRESSING AND SCORING OF RESULTS OF SKIN TESTING

The results of both percutaneous and intracutaneous skin tests are often reported in only semiquantitative terms. Results may be recorded only as positive or negative or in terms of 0 to 4+ without any indication of what size reactions these numbers represent.<sup>24</sup> Allergy patients may have to change their allergist for numerous reasons. Thus, it is important that prior allergy testing records be interpretable by the receiving

Figure 1. Skin test form developed by the Immunotherapy Committee of the American Academy of Allergy.

physician. At the very least, a record of skin testing should indicate sufficient information to allow another physician to interpret the results and avoid the need to repeat skin testing. In addition to the concentration of extract used (consider including manufacturer), the form should indicate whether the tests are percutaneous or intracutaneous and, if the former, which device was used for testing, whether testing was performed on the back or the arm, and the size of the positive and negative control reactions. Finally, if an arbitrary grading system is used, the range of reaction for each grade should be clearly indicated on the form (Table 2). A skin test form has been developed by the Immunotherapy Committee of the American Academy of Allergy (Fig 1).

A superior method of expressing results is to measure the reaction and enter the measurement on the form. This need not be excessively time-consuming. Although areas of the wheal and erythema are the most accurate, measurements of the product of the orthogonal diameters, the sum of the orthogonal diameters, and even the longest diameter correlate very well with area, with *r* values greater than 0.9.<sup>25</sup> The importance of performing such measurements are exemplified by a recent study by McCann and Ownby<sup>26</sup> in which allergists were asked to interpret photographic copies of skin test reactions. They demonstrated a great degree of variability in scoring and interpreting of the skin test results. The authors thus conclude that the most reliable method to report a skin test reaction was to measure and record the reaction size.

The Scandinavian Society of Allergology recommended that skin test results be standardized in relation to the size of the reaction to histamine, using 0.1 mg/mL of histamine for intradermal testing and 1 mg/mL mg of histamine for skin prick testing.<sup>27</sup> The suggested method was to calculate the mean diameter. If the reaction to allergen was the same size as the histamine reaction, the grade was 3+; if half that size, 2+; and if twice as large, 4+. A subsequent study suggested that the histamine control should be 10 mg/mL because of the small reactions with high coefficient of variation with the 1-mg/mL histamine skin prick test.<sup>28</sup> Even the 20% to 30% coefficient of variation for reactions to 10 mg/mL raises questions regarding the desirability of standardizing to a histamine control, which if used for this purpose should be performed at least in duplicate.<sup>29</sup>

The reliability of different means of interpreting results of skin prick testing was compared in patients sensitive to dogs.<sup>30</sup> A determination of sensitivity to dog was made in 202 children based on a composite score from history, radioallergen sorbent testing, and bronchial or conjunctival allergen

challenges. The results of 3 common means of expressing test results are given in Table 3. Although the overall efficacy of the histamine reference was greatest in this study, most allergists would prefer to have the maximum sensitivity so not to miss any truly sensitive patients. Other methods, or clinical judgment, should then be used to distinguish between those only sensitized and those clinically allergic.<sup>27</sup>

Immediate skin reactions to histamine typically peak at 8 minutes, whereas those to allergen peak at 15 minutes. Isolated, delayed reactions to allergy skin testing have been described.<sup>31-33</sup> Furthermore, when looked for, they appear fairly commonly to follow intracutaneous testing.<sup>31,32</sup> Delayed reactions have been demonstrated to be more than twice as common at sites of positive immediate compared with negative immediate skin reactions. Those occurring at the site of negative immediate skin test results have displayed the histologic features of a delayed-type hypersensitive reaction.<sup>32</sup>

## PROFICIENCY TESTING

Like all other laboratory tests, it is imperative that quality assurance standards be met to ensure that accurate testing technique is being performed. To confirm such standards, it is recommended that all technicians who perform skin testing undergo evaluation of their technique.<sup>34</sup> The National Committee for Clinical Laboratory Standards recommends such quality control procedures for daily performance of *in vitro* allergy testing, with a recommended coefficient of variation of less than or equal to 15%.<sup>35</sup> Certainly, it would be comforting to know that some degree of consistency in skin test performance is achieved by skin test technicians. Although there are no formal criteria available regarding skin test proficiency testing, several publications have provided suggested values. European publications have suggested a coefficient of variation of less than 20% following repeated skin test control applications,<sup>36</sup> and the recent Childhood Asthma Management Program study required a coefficient of variation of less than 30% be attained to confirm proficiency in skin testing. A suggested protocol for quality assurance testing and proficiency testing for skin testing technicians is shown in Figure 2.

## CONCLUSION

As written by Dr Walzer in 1947, "the fact that skin testing has not turned out to be a simple and completely reliable technics [sic] does not detract from the fact that when it is intelligently and skillfully performed, it remains the most

Table 3. Comparison of Criteria for a Positive Skin Prick Test Result to Dog\*

| Criteria               | Sensitivity | Specificity | Overall efficacy |
|------------------------|-------------|-------------|------------------|
| ≥3-mm wheal diameter   | 0.98        | 0.82        | 0.89             |
| ≥5-mm wheal diameter   | 0.66        | 0.98        | 0.84             |
| 3+ histamine reference | 0.92        | 0.93        | 0.93             |

\* Data are from Vanto.<sup>30</sup>

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**Suggested Proficiency Testing and Quality Assurance Technique for Skin Prick Testing:**

- Using desired skin test device, perform skin testing with positive (histamine 1-10) and negative controls (saline 1-10) in an alternate pattern on a subject's back
- Record histamine results at 8 minutes by outlining wheals with a felt-tip pen and transferring results with transparent tape to a blank sheet of paper
- Record saline results at 15 minutes by outlining wheal and flares with a felt-tip pen and transferring results with transparent tape to a blank sheet of paper
- Calculate the mean diameter  $X=(D+d)/2$ ; D=largest diameter and d=perpendicular diameter at midpoint of D
- **Histamine**  
Calculate the mean and standard deviations of each mean wheal diameter  
Determine coefficient of variation = standard deviation/mean  
Quality standard should be less than 30%
- **Saline**

All negative controls should be <3-mm wheals and <10-mm flares

Figure 2. Suggested proficiency testing and quality assurance technique for skin prick testing.

effective diagnostic procedure in reaginic allergic disorders.”<sup>37</sup> To improve on the predictive values of allergy skin testing, there are several controllable variables that when addressed can result in more reliable skin test results. Allergists should also consider quality standards with proficiency testing and proper documentation of results so that others can easily interpret results. In so doing, we can improve our most effective diagnostic tool available for allergic disease.

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- Requests for reprints should be addressed to:*  
*John Oppenheimer, MD*  
*c/o Pulmonary and Allergy Association*  
*8 Saddle Rd*  
*Cedar Knolls, NJ 07927*
-