Skin prick testing
for the diagnosis of allergic disease

A manual for practitioners

ASCIA skin prick testing working party

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Skin prick testing for diagnosis of allergic disease

This manual has been prepared by a working party of ASCIA and has been endorsed by the ASCIA Council. It is intended for medical and allied health practitioners and it outlines the application, method and interpretation of allergy skin tests.

Contents

**PREAMBLE**

1. **INTRODUCTION**

2. **PRE-TEST CONSIDERATIONS**

   2.1 Conditions for which skin prick testing is considered an appropriate investigation

      2.1.1 SPT disease indications
      2.1.2 SPT not usually indicated
      2.1.3 Indications for intradermal testing

   2.2 Patient selection in skin prick testing

      2.2.1 Patient age
      2.2.2 Contraindications
      2.2.3 Relative contraindications/precautions
      2.2.4 Drugs that interfere with the skin prick test response
      2.2.5 Drugs that may be contraindicated in skin prick testing
      2.2.6 Patient factors leading to variability in skin test results

   2.3 Other tests for specific IgE

      2.3.1 Serum specific IgE

      Table: Comparison of serum specific IgE and skin prick testing

      2.3.2 Intradermal skin testing

3. **METHODS**

   3.1 Allergens for skin prick testing:

      3.1.1 Commercial extracts
      3.1.2 Composition of skin testing extracts
      3.1.3 Cross-reactivity
      3.1.4 Allergen test panels
      3.1.5 Food allergens
      3.1.6 Alternative sources of skin testing reagents
      3.1.7 Maintenance of allergen extracts
      3.1.8 Inappropriate allergens

   3.2 Positive and negative controls

   3.3 Devices used for skin prick testing

   3.4 Skin prick test procedure

      3.4.1 Requirements for skin prick testing procedure
      3.4.2 Site of application
      3.4.3 Method
      3.4.4 Time of reading results
      3.4.5 Measurement of wheal and flare
      3.4.6 Method of recording skin prick test results
      3.4.7 Patient aftercare
      3.4.8 Post-test holding time
3.5 Skin prick test result reporting
   3.5.1 Reporting forms
   3.5.2 Standardised quantitative reporting
   3.5.3 Qualitative reporting
   3.5.4 Qualitative scales

4. INTERPRETATION OF SKIN PRICK TEST RESULTS
   4.1 Meaning of “positive” and “negative” tests
   4.2 Performance characteristics of skin prick testing
   4.3 Challenge tests

5. PERSONNEL
   5.1 Medical staff
   5.2 Paramedical staff
   5.3 Training

6. SAFETY AND RISKS
   6.1 Safety/risks of skin prick testing
       Table: Risk factors for anaphylaxis in skin prick testing
   6.2 Safety measures and safety equipment required
       Table: Suggested minimum standards for available emergency equipment and medications.

7. REGULATORY ISSUES
   7.1 Positive control solutions
       7.1.2 Histamine
       7.1.3 Codeine phosphate
   7.2 Allergen solutions
   7.3 Skin prick test devices
   7.4 Personnel carrying out the test
   7.5 Consent

8. REFERENCES

APPENDIX 1:
   STANDARDS FOR SKIN PRICK TESTING

APPENDIX 2:
   DRUGS WHICH ARE ANTIHISTAMINES OR HAVE ANTIHISTAMINE ACTIVITY WHICH MAY INTERFERE WITH SKIN TESTING

APPENDIX 3:
   IMPORTERS/DISTRIBUTORS OF SKIN PRICK TESTING REAGENTS AND SKIN PRICK TESTING DEVICES IN AUSTRALIA (Current as of August 2008)
PREAMBLE

This manual will focus on skin prick testing in the diagnosis of immediate IgE-mediated allergy. It is intended for all practitioners of skin prick testing. Hitherto there has been no comprehensive and practical guide for this procedure. There have been criticisms that protocols for skin testing have been lacking, and the procedure is often left to nurses and technicians with limited teaching and little attempt at quality control or methodological supervision, and no attempts at standardisation of methodology at different centres. Recent commentaries and surveys have highlighted the variability of the clinical application and technical methodology by different practitioners and also variability in the interpretation and communication of results. In this manual we introduce recommendations for the clinical application of skin testing to allergy diagnosis, standardised methods for the technique of skin prick testing and reporting, and advice on interpretation of the results of skin prick testing.

In preparing this manual an extensive literature search was undertaken using keywords “skin, test, allergy, hypersensitivity” and results were culled to focus on papers that primarily examine the skin prick test itself or its broad applicability, and not its use in specific conditions. Papers that exclusively address intradermal skin tests were excluded but those which compared intradermal and prick tests were included. Papers on patch testing were also excluded. Papers were divided into safety, diagnostic accuracy, methodological factors, interfering factors, and reviews. There have been numerous reviews, position papers and practice parameters, many of which were scrutinized (see references). There are relatively few high-quality published studies on the methodology and diagnostic validity of allergy testing. Some optimal studies compare different methodologies with clinically relevant gold standards (NHMRC evidence level II or III-1). This manual is based on expert opinion (ASCIA skin prick testing working party; ASCIA council review and endorsement); published references are cited to support assertions where available.

The manual is intended for use in Australia and New Zealand although some aspects, particularly availability of materials and regulatory issues, do not apply in New Zealand. References are made to some product manufacturers and distributors, to assist with procurement of materials within Australia. The content of this manual is based on the available references at the time of publication.
1. **INTRODUCTION**

There are three types of skin testing used in allergy diagnosis:

1. **Skin prick testing (SPT)** - the primary mode of skin testing for immediate IgE-mediated allergy. It is widely practiced, carries very low (but not negligible) risk of serious side-effects to patients and provides high-quality information when performed optimally and interpreted correctly. (Also called prick skin testing or PST).

2. **Intradermal testing (IDT)** - Relevant to both immediate IgE-mediated allergy and delayed-type hypersensitivity. When used in the diagnosis of immediate allergy, it carries a higher risk of adverse reactions and requires high levels of technical and interpretive expertise.

3. **Patch testing** - relevant to contact hypersensitivity and some other forms of delayed-type hypersensitivity. It is conducted mainly by dermatologists and some immunologists, and is not relevant to immediate or IgE-mediated allergy, and will not be further discussed.

*(NB. “Scratch” testing is not endorsed and should no longer be performed).*

Skin prick testing provides information about the presence of specific IgE to protein and peptide antigens (allergens).

Small amounts of allergen are introduced into the epidermis and non-vascular superficial dermis and interact with specific IgE bound to cutaneous mast cells. Histamine and other mediators are released, leading to a visible "wheal-and-flare" reaction peaking after about 15 minutes.

The value of this test depends on a number of steps, including:

- The relevance of the test allergen to the condition under investigation
- The correct introduction of a sufficient amount of allergen in its native (allergenic) form
- The functional status of cutaneous mast cells
- The interpretation of the reaction in the context of positive and negative controls

Correctly used, the skin prick test has good sensitivity and specificity for the presence of allergen-specific IgE and is in some cases more sensitive than *in-vitro* testing for specific IgE in serum\(^6,7\). The discomfort is small and the risk of systemic reactions is minimal although not negligible.

Ultimately the integration of skin prick test results, knowledge of the biology of the various allergens and the exposures of the patient, and the nature and timing of the symptoms enable the construction of a diagnosis and an appropriate management plan for the patient.

Intradermal skin testing has more specialized applications such as testing for IgE-mediated drug allergy, particularly penicillins, and venom allergy. It carries a higher risk of anaphylaxis and is generally restricted to a hospital or specialist setting. Intradermal testing will be dealt with in more detail in a forthcoming document.
2 PRE-TEST CONSIDERATIONS

2.1 Conditions for which skin prick testing is considered an appropriate investigation:

2.1.1 The following conditions are generally accepted indications for allergy skin prick testing:

- Rhinitis/rhinoconjunctivitis/rhinosinusitis/allergic conjunctivitis
- Asthma
- Atopic dermatitis
- Food reactions such as those manifested by anaphylaxis, immediate acute urticaria, or acute flare of eczema
- Suspected latex allergy
- Conditions in which specific IgE is considered likely to play a pathogenic role (e.g. selected cases of chronic urticaria if the history suggests an exogenous allergic cause)
- Rarer disorders such as allergic bronchopulmonary aspergillosis, eosinophilic oesophagitis or eosinophilic gastroenteritis

(NB the choice of allergens tested will vary according to which of these conditions is being examined and patterns of allergen exposure - see local allergen prevalence charts, appendix 3)

2.1.2 Skin prick testing is not routinely indicated in the investigation of:

- Nonspecific skin rash without allergic/atopic characteristics
- Chronic urticaria in the absence of allergic features on history
- Food intolerance without allergic features (e.g. irritable bowel syndrome)
- Assessment of the effectiveness of allergen immunotherapy
- Chronic fatigue without allergic features
- Migraine headaches/behavioural disorders
- Reactions to respiratory irritants (smoke, fumes, perfumes etc.)

Skin prick testing is not appropriate for the diagnosis of reactivity to low molecular weight substances such as food additives, drugs (with some exceptions - see later), respiratory irritants, and most occupational allergens (with some exceptions - see later).

2.1.3 Conditions for which intradermal testing is appropriate:

Intradermal testing may be used in the diagnosis of:

- Insect venom hypersensitivity
- Immediate allergy to beta-lactam drugs, other drugs where validated protocols exist
- Immediate hypersensitivity to some vaccines

(Intradermal testing is recommended for hospital or specialist use only).

Intradermal testing is not indicated for aeroallergens, and is contraindicated in routine practice for food allergy.

Allergy testing has been shown to increase the accuracy of diagnosis when added to history and clinical examination. It differentiates allergic diseases from other mimicking conditions. It may lead to allergen avoidance strategies, improved use of medications, and for some patients, desensitisation treatment (immunotherapy). The strongest indications for skin prick testing are where there is good evidence for the effectiveness of allergen avoidance or immunotherapy.

Skin prick tests are also frequently used for epidemiological purposes or to define atopy in an individual without specific disease diagnosis considerations. A definition of atopy is “the
genetically determined tendency to produce specific IgE to common environmental allergens”. A positive reaction to one or more of a panel of the most prevalent allergens to which the subject or population is likely to be exposed defines the subject as atopic. A lack of atopy, by this definition, does not exclude the possibility of sensitisation to other allergens that were not tested. Certain allergies, for example to insect venom or drugs, are not directly related to atopy.

2.2 Patient selection in skin prick testing

2.2.1 Patient age

There are no strict age limits but skin reactions are often diminished in the very young and the elderly, making interpretation more difficult in both cases. Infants often show larger flares and smaller wheals. Systemic allergic reactions may rarely occur in response to skin testing in infants (as in patients of any age). Because of increased risk and greater complexity of interpretation, skin prick testing below the age of 2 years should be considered a specialist practice.

2.2.2 Contraindications

<table>
<thead>
<tr>
<th>Conditions which contraindicate/preclude skin prick testing</th>
</tr>
</thead>
<tbody>
<tr>
<td>▪ Diffuse dermatological conditions- test must be performed on normal healthy skin</td>
</tr>
<tr>
<td>▪ Severe dermatographism</td>
</tr>
<tr>
<td>▪ Poor subject cooperation</td>
</tr>
<tr>
<td>▪ Subject unable to cease antihistamines/other interfering drugs</td>
</tr>
</tbody>
</table>

2.2.3 Relative contraindications/precautions

<table>
<thead>
<tr>
<th>Contraindicated in non-specialist practices for safety reasons (see section on safety below)</th>
</tr>
</thead>
<tbody>
<tr>
<td>▪ Persistent severe/unstable asthma</td>
</tr>
<tr>
<td>▪ Pregnancy (because of the small risk of anaphylaxis with hypotension and uterine contractions)</td>
</tr>
<tr>
<td>▪ Babies and infants</td>
</tr>
<tr>
<td>▪ Patient on beta-blockers</td>
</tr>
</tbody>
</table>

2.2.4 Drugs that interfere with the skin prick test response

A large range of drugs may reduce skin reactivity and must be withheld before skin testing (see appendix 2). First generation antihistamines usually have a short duration of action whereas second generation act for longer; the duration of suppression of skin test reactivity is variable between different drugs and individuals. Antidepressants such as doxepin, other tricyclics, and tetracyclics have antihistamine activity and may need to be withheld for 1-2 weeks or more. Phenothiazines also have antihistamine activity. Think of OTC cold and flu remedies, “sinus” analgesics, antitussives; also of antiemetics, sedatives, relaxants, migraine prophylactics (cyproheptadine, pizotifen). Oral corticosteroids probably do not significantly diminish the skin test reaction even after prolonged use, but prolonged topical corticosteroids have been shown to reduce skin reactivity. Topical pimecrolimus does not alter skin prick test reactivity. Topical moisturizers do not reduce prick test reactions but may cause extracts to run or disperse which creates a practical difficulty.

2.2.5 Drugs that may be contraindicated in skin prick testing

Beta-blockers are contraindicated in situations in which the risk of systemic anaphylaxis is increased (see “risks of skin testing”). ACE inhibitors may be relatively contraindicated in the same circumstances. These drugs may interfere with the normal compensatory mechanisms
in anaphylaxis and beta-blockers interfere with the effect of adrenaline. In general the risk of systemic anaphylaxis from skin testing is low and the drugs need not be withheld except where certain high-risk features exist (see “risks of skin testing”).

2.2.6 Patient factors leading to variability in skin test results

Dermatographism can cause nonspecific wheal-and-flare results to skin pricking alone; the negative control may show a wheal and this renders the allergens difficult to interpret unless the reaction is markedly larger than the negative control. Mild dermatographism does not preclude skin testing. Some techniques of skin prick testing may be more likely to activate dermatographism.

The following factors may lead to some variability but this is not usually significant in result interpretation: menstrual phase, race, circadian rhythm, seasonal variation, atopic dermatitis (elsewhere on body).

The following conditions can reduce skin test reactivity: chronic renal failure, CVA, cancer (some cases), spinal cord injury, diabetic neuropathy, recent anaphylaxis. Skin prick testing should not be carried out on limbs affected by lymphoedema, paralysis or neurogenic abnormalities.

A very recent report demonstrates that individuals infected with RSV show increased histamine wheal size and false positive allergen skin test wheals. This study remains to be confirmed and broadened but suggests the possibility that skin tests carried out in the presence of acute viral infection may need to be interpreted with caution.³

2.3 Other tests for specific IgE

2.3.1 Serum specific IgE

Serum allergen specific IgE testing (formerly known as the RAST* test) is an automated test performed on blood samples by a pathology laboratory. As the name suggests it detects free antigen-specific IgE in serum as opposed to antigen-specific IgE bound to mast cells in the skin.

Whilst the results of skin prick testing and serum specific IgE tests are usually concordant, there are some exceptions to this and in the past it was considered that the skin prick test is more sensitive.

Newer methods may have improved the sensitivity of serum testing compared to skin testing however in some cases this remains limited (eg. latex testing).

The sensitivity and specificity of both tests depend on the cutoff of the serum IgE level or the skin test wheal size.

This is not intended as a detailed review of the comparative diagnostic utility of both of these tests but a comparison of the main features of skin prick testing and serum specific IgE testing is shown in the table on the following page.

*RAST is an abbreviation for ‘Radio Allergo Sorbent Test’, which was the original technology used for serum specific IgE testing.
### Serum specific IgE test

<table>
<thead>
<tr>
<th><strong>Favour serum IgE testing:</strong></th>
<th><strong>Skin test</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td>Widely available in any medical setting</td>
<td>Available only where equipment, reagents and trained staff are on hand</td>
</tr>
<tr>
<td>minor pain- venesection</td>
<td>minor discomfort, itching</td>
</tr>
<tr>
<td>Little patient effort or cooperation required</td>
<td>Requires patient cooperation</td>
</tr>
<tr>
<td>No risk to patient; may be first line with certain high-risk allergens</td>
<td>Slight risk of systemic allergic reaction, more so in some situations</td>
</tr>
<tr>
<td>Can be done where there is extensive skin disease</td>
<td>Require areas of normal skin for testing</td>
</tr>
<tr>
<td>Can be done where the patient has taken antihistamines or is unable to stop certain medications which might interfere with SPT</td>
<td>Must stop antihistamines and some antidepressants and other drugs several days before test (see appendix 2)</td>
</tr>
<tr>
<td>Many allergens available, including some which are not available for skin testing or not routinely carried in skin test settings. Some laboratories may send away samples for rarer allergens</td>
<td>Many allergens available, but some low-demand allergens will not be carried by individual practices</td>
</tr>
<tr>
<td>Laboratory test, subject to quality control and standardization</td>
<td>Methodology and result quality variable, no standardization or formal quality control at the current time</td>
</tr>
</tbody>
</table>

### Favour skin prick testing:

| **Venesection may be painful or anxiety-provoking particularly in children** | Minor scratch, itch if positive |
| Results may take days or weeks | Results in half an hour |
| Results are not directly meaningful to patients | Results are visible and compelling to patients; may have value in ensuring compliance with allergen avoidance measures |
| Reasonably good sensitivity | In most cases, shown to have better sensitivity for clinically valid allergies |
| Some food allergens, drugs, rarer pollens not available for testing | Can extemporaneously prepare allergens (with appropriate considerations; specialist practice) |
| Some allergens particularly foods may show low sensitivity in certain clinical situations | Freshly prepared food allergens may show more sensitivity in certain circumstances (caution- risk of anaphylaxis) |
| False positives possible with high total IgE levels | No interference from high total IgE |
| Numerical results obtained on different types of equipment are not directly comparable | Numerical measurements may vary by different operators |

Both serum allergen specific IgE tests and skin prick tests require skill and knowledge for the interpretation of results and application to the clinical problem of the patient.
2.3.2 Intradermal skin testing.

Allergens are injected intradermally to produce a small bleb, and the outcome measure is an increase in the size of the wheal at 20 minutes. Allergens need to be diluted (100-1,000 fold) from the concentrations used for skin prick testing. Skill is required to inject correctly and interpret the result. Most importantly, there is a small but significant (much higher that skin prick testing) risk of systemic reactions including anaphylaxis. A number of deaths have been reported from intradermal skin testing, but only one from prick testing\textsuperscript{14}.

Intradermal testing is considered a specialist practice and is generally performed in a hospital (or equivalent specialist) setting.

Intradermal testing is usually contraindicated for food allergy and is considered inappropriate for the vast majority of cases of suspected inhalant allergy because of lack of specificity\textsuperscript{6,15,16}. Skin prick testing has been shown to correlate better with symptoms than intradermal testing\textsuperscript{17,18}.

Intradermal testing has an established place in testing for penicillin allergy and may be considered appropriate for cephalosporin allergy, although validated protocols are lacking. The test is also used for diagnosis of allergy to a number of other drugs such as insulin, opiates, anaesthetic agents, muscle relaxants, and enzymes. It can be used for bee venom allergy testing although the clinical predictive value of the test is open to question\textsuperscript{19}.

Intradermal skin testing has been widely practiced in the US for routine allergy diagnosis but is gradually falling out of favour for the reasons stated above. Interpretation of the allergy literature needs to take into account the type of tests that have been carried out.

Detailed description of the technique and interpretation of intradermal skin testing is outside the current scope of this manual.
3. METHODS

3.1 Allergens for skin prick testing

3.1.1 Commercial extracts
Allergen extracts are manufactured specifically for the purpose of skin prick testing. These are aqueous solutions of proteins extracted from the relevant materials, combined with 50% glycerol which acts as a preservative. The solutions are therefore quite viscous. They are supplied in multi-use dropper bottles.

Extracts are not currently manufactured in Australia, there are a small number of manufacturers and suppliers internationally. Allergens potentially available in Australia at the current time are manufactured by Hollister-Stier (USA), Stallergenes (Europe) or ALK-Abello (Europe and USA).

Skin testing reagents must be registered by the TGA before they can be distributed in Australia.

However there have been a number of supply, distribution and regulatory issues with regard to these products which affect their availability to clinicians (see section 7.2, appendix 3).

3.1.2 Composition of skin testing extracts
Allergy extracts should contain all allergenically relevant proteins of the labeled substance, and should be free of cross-contamination with allergenic proteins of other substances, eg. an allergen extract of one type of plant pollen should not be contaminated with another pollen. Some extracts contain defined mixtures of related allergenic substances (eg. a mixture of weed pollens or related tree pollens, or several species of alternaria mould). Some extracts are standardized for allergenic potency, whereas others are prepared according to weight of allergenic material used for elution of allergens.

Allergen extracts are complex mixtures and contain a range of allergenic proteins which can be separated by electrophoresis and visualized by immunoblotting. Different manufacturer’s preparations (and different batches from the same manufacturer) of the same allergen may vary in their content and proportion of the major allergenic proteins. This might be due to differences in the source material or its preparation (eg. fungal species from different sources, cultured under different conditions and harvested at different stages of life cycle) and the techniques of allergen preparation (eg. the use of pyridine in extraction of dust mite allergens reduces the proportion of Der P1).

Although only one range of extracts is predominantly used in Australia, these differences may explain variability of results, unexpected positive or negative results, and some differences between skin prick testing and serum specific IgE tests. Interpretation of published studies must take into account the possibility of results being affected by the source of extract. Standardisation of extracts is a major issue to which attention is being directed by allergy authorities and manufacturers.

Allergenic substances invariably contain hundreds of different proteins, each with a unique sequence; only a subset of these proteins is potentially allergenic. However, different individuals form IgE to different proteins within this mixture. If the particular protein(s) to which IgE is directed in a particular individual is not represented within the allergen extract (due to manufacturing processes or protein lability), this may lead to a negative allergy test, even though the individual is allergic to the substance when encountered in nature. This is a potential cause for false negative skin prick tests.
3.1.3 Cross reactivity

Cross-reactivity is an important concept in choice of extracts for skin testing and interpretation of results. Cross-reactivity describes the phenomenon whereby IgE reactive to a particular allergen also reacts to other similar allergens; the patient may never have been exposed to the second allergen. Cross-reactivity of pollen and other allergens often relates to phylogeny but there are sometimes patterns of cross-reactivity that would not have been predicted by biological relatedness, due to proteins that have conserved structures across diverse species. Where two allergens are fully cross-reactive it may not be necessary to include both in testing panels if economy is important. For example many grass pollens are fully cross-reactive with rye grass, so it may not be necessary to test separately for orchard grass etc. On the other hand there are reports that timothy grass, which is in the same family as rye grass, is usually cross-reactive but has some unique allergenic proteins, so in areas where it is prevalent it should be included in the panel.

3.1.4 Allergen test panels

It is important that the allergens tested for should be relevant to the patient’s clinical condition and to exposure. In general the smallest number of allergens required to establish a diagnosis and adequately manage the patient should be used. Relatively small allergen panels (eg. 8-12 inhalant allergens) would usually be considered adequate for testing by general practitioners or respiratory laboratories. For allergy specialists, more detailed information may be required, particularly when planning immunotherapy, and to identify less common allergies. Panels should also vary with the locality depending on differences in flora and fauna (see appendix 3). However practice varies widely, and panels of between 6 and 60 allergens for one test are advocated by different authorities. If a practice does not perform large numbers of tests it is usually not economical to maintain a large panel.

3.1.5 Food allergens

Testing for IgE-mediated food allergy by skin prick testing is valid but interpretation is complex. Positive tests often occur without clinical allergy and negative tests in the presence of clinical reactivity may occur, for many reasons. There is a greater risk of anaphylaxis than skin prick testing with aeroallergens, and intradermal testing is almost never appropriate for foods. Commercial allergen extracts are available but are non-standardised. In some cases it is more effective to carry out skin prick testing using freshly prepared food extracts or the food itself. Food allergy testing is not appropriate for non-specialist practices, general practitioners and respiratory laboratories because of risks, carrying and managing reagents, and complexity of interpretation of results and counseling of patients.

3.1.6 Alternative sources of skin testing reagents

As mentioned, fresh foods can be used for skin testing but the procedure and interpretation are specialized, and risk of anaphylaxis is increased. There may be variability in the allergenic quality of the food or its relevance for testing. Pollen extracts may theoretically be prepared but this is difficult and best left to experienced practitioners. Pollen collection must be optimal as should extraction of allergens into solution. Most of the allergenically relevant pollens (or closely related, cross-reactive species) are available as extracts.

Other sources of skin testing reagents include other commercial companies and laboratory-prepared protein extracts, generally for research applications. When any sources of allergen other than the standard commercially available ones are used it is strongly recommended that this should be recorded as such on the report form.
3.1.7 Maintenance of allergen extracts
Allergen extract bottles should be clearly labeled. They are usually supplied as a dropper bottle with a rubber teat and glass dropper. They should be stored in a temperature-monitored refrigerator and left out for as short a time as necessary to conduct the test. The expiry dates should be checked since the potency of the extracts may vary with time. Allergen extracts often remain active long after the expiry date but the reliability of this cannot be guaranteed. Potency and longevity are also compromised by dilution and high temperatures. Precautions must be used to prevent bacterial contamination and cross-contamination between allergens.

The following practical measures are recommended:
- Label the test solution bottles with numbers and place them in order in a rack.
- Only open one bottle at a time; if a stopper is put onto the wrong bottle, this results in contamination with the other allergen, and bottle and stopper must be discarded.
- Clean the patient’s skin prior to testing to prevent contamination of the tip of the dropper; use only on intact skin.
- When depositing the allergen solution drop on the patient’s skin, it is acceptable to touch the drop against the skin but not the glass tip of the dropper.

3.1.8 Inappropriate allergens
Allergens acting through IgE or type-1 mechanisms can be appropriately tested by the skin prick method. However, airborne substances may produce allergy-like symptoms through other mechanisms such as respiratory irritation. For example it is not appropriate to test for cigarette smoke or tobacco by skin prick testing, since it acts as a respiratory irritant rather than an allergen. Plants may be strongly scented or produce volatile irritant compounds which can cause allergy-like symptoms, yet this is distinct from allergy to plant pollen. Patients may complain of symptoms from (for example) jasmine vine or roses, yet this is not due to pollen allergy, and skin prick testing is not an appropriate investigation. Pollens from many flowers are entomophilous (designed to be spread by insects) therefore sticky and heavy and likely to fall to the ground rather than be inhaled.

Foods often produce symptoms through non-IgE mechanisms, for example, negative skin prick testing for wheat does not exclude coeliac disease, and negative testing for milk does not exclude lactose intolerance or delayed immune reactions to dairy products. If these disorders are suspected based on the nature of the symptoms then skin prick testing is not the appropriate investigation.

3.2 Positive and negative controls
These are essential for the following reasons:
Some patients display dermatographism or develop a small flare or wheal from the pinprick alone. This leads to an apparent reaction to extracts to which the patient is not actually sensitised. The negative control would be expected to show a similar reaction. If this occurs then either the test must be rejected as uninterpretable (if there is insufficient distinction between the reaction to the negative control and the positive control), or interpreted by comparison with reaction to the negative control (eg. if the negative control produces a wheal of 3mm, only wheals of >6mm will be considered positive). Caution is required since the dermatographic response is often inconsistent at different skin sites, and may produce different reactions for a range of extracts to which the patient is not allergic. Wheals of >3mm to the negative control indicate severe dermatographism and would require rejection of the test. Careful technique can minimize nonspecific reaction in dermatographic patients.
The positive control should produce a wheal of approximately 6mm, and if there is no wheal or only a tiny one, this may indicate either that the patient has taken an antihistamine or a drug with antihistamine activity (see appendix 2) or that they have non-reactive skin, in which case SPT will not be possible. It is recommended that a wheal of \( \geq 4 \text{mm} \) to the positive control is acceptable (or 4mm greater than the negative control) and if it is <4mm the test should be considered uninterpretable.

The negative control is the same solution as the allergens are made up in, eg. saline buffer/50% glycerol, without any allergen. It is also available commercially. The positive control can be a solution of histamine (usually histamine phosphate 10mg/ml) (directly induces cutaneous wheal and flare response) or codeine (usually 9% solution) (degranulates cutaneous mast cells, indirectly causing wheal and flare). Availability of positive control solutions is problematic (see section 7.1).

### 3.3 Devices used for skin testing

Sharp lancets are used to prick through the drop into the epidermis and superficial dermis. Some devices consist of a point on a flat stopper, so that the device can be “jabbed” onto the patients skin entering the epidermis and upper dermis, without penetrating too deeply. A sharp pointed device such as a prick lancet can be used with an oblique “prick and lift” technique, without inserting the needle too deeply. The prick should not be deep enough to draw blood, although in the elderly with thin skin this may be unavoidable. Hypodermic needles were formerly used but are not recommended since they are difficult to control sufficiently so as not to prick too deeply, and they are quite expensive and increase the hazards of the test to both patient and practitioner (see below).

Some practitioners advocate using the same device for several pricks, wiping on gauze or an alcohol swab between each one to reduce the chance of carry-over of allergen. Clearly this is more economical. However studies have demonstrated that the risk of carry over of allergens remains, and may vary between different allergens\(^{20,21}\). Another drawback of this method is the risk of injury to the practitioner during the wiping procedure, which could result in blood contact and needlestick injury. For these reasons multiple use of the same lancet device is not recommended. In particular, multiple use of a hypodermic needle is contraindicated because of the increased likelihood of carry-over and the greater risk of needlestick injury. Many other devices (eg plastic multitest devices, duotip devices) have been developed in other countries but are not registered for use or generally available in Australia.

### 3.4 Skin prick test procedure

#### 3.4.1 Requirements for skin prick testing procedure:

- Allergen extracts
- Positive and negative control solutions
- Sterile lancets for skin pricking
- Sharps container for disposal of lancets
- Marker pen for the skin
- Ruler for measuring reactions
- Tissues for wiping solutions
- Recording sheets
- Gloves (latex and latex-free)
The subject needs to be in a comfortable position, with the forearms or back at a convenient height for the practitioner to do the test. The procedure should be explained to the patient (an information sheet can be provided), reassurance provided if necessary, and an enquiry should be made about medications that the patient is taking. Patients must have avoided antihistamines and other interfering drugs as well as skin moisturizers prior to the procedure (see section 2.2.4 above and appendix 2). The area to be tested should be exposed with no risk of clothing brushing across the test area and wiping the test solutions (especially wiping the solution onto another prick location). The room should be private and at a comfortable temperature especially if the patient needs to disrobe. It is advisable to provide the patient with a magazine or something to occupy themselves for the 15 minutes or so that is required for the test to develop (and to distract them from any discomfort).

3.4.2 Site of application
Generally the most convenient and frequently used sites are either the volar surface of the forearm or outer upper arm, and the back. Reactions to allergen (but not histamine) are larger on average on the back than the arm, larger on the lower than the upper back, and on the upper forearm compared to the wrist. In the presence of appropriate controls these differences should not be clinically significant but because some small reactions can be close to the threshold for positivity, one study showed a slightly larger number of positive reactions on the back. However the clinical significance of these was not investigated. Generally it is advisable to site tests more than 5cm from the wrist and 3cm from the antecubital fossa.

3.4.3 Method
It is desirable but not essential to clean the skin site with alcohol prior to skin prick testing (this may be contraindicated in cases of extreme dry skin and eczematous tendency). Positions for skin pricks should be marked by numbers on the skin to identify the allergen, and pricks should be made immediately adjacent to the numbers to avoid confusion between allergens. Prick tests should be at least 2cm apart to avoid overlapping reactions and false-positive results. Allergen will be applied from the dropper bottle prior to pricking the skin. The drop on the tip of the dropper can be touched on the skin to transfer the liquid but the actual tip of the dropper should not touch the skin. In cooperative patients or if a small number of allergens are used, all drops can be deposited before commencing pricking. In other cases it may be preferable to deposit a group of drops and prick them, then another group. In some cases, for example children with poor cooperation, it may be more practical to deposit each drop and prick each drop straight away. It is important not to allow the extract to run onto the next prick site. In patients with eczema who use moisturizers the drop may flatten or run more easily on the skin. Where many allergens are used it may be necessary to take into account the time that the first pricks are done compared with the last ones, when deciding the appropriate time to read the results. Many practitioners leave the drops on the skin until the test is ready to read but this is probably not necessary; the test solution can be blotted from the skin after 1 minute without compromising the eventual result.

3.4.4 Time of reading results
The reaction to the histamine positive control is at its maximum size at approximately 10 minutes whereas the allergen reaction reaches its maximum at around 15 minutes. In practice the histamine wheal is usually still showing at 15 minutes and this is recommended as the optimal time for reading skin test results. Occasionally allergen responses continue to enlarge up to about 20 minutes. Overall, the histamine result should be read at 10-15 minutes after the skin prick, and the allergens at 15-20 minutes. If the test is left for longer than 20 minutes
the histamine and allergen response may diminish or be lost, and if not measured on time due to some delay, the test may need to be repeated.

3.4.5 Measurement of wheal and flare

The drops must always be carefully blotted from each test site prior to taking measurements; care should be taken not to cross-contaminate allergen test sites with the blotting tissue or the ruler used to measure the results. The standard and accepted method for quantifying the skin prick reaction is to measure the mean diameter of the wheal, using a ruler marked in mm (a transparent ruler is often most convenient; calipers are also available for this purpose). If the result is a circular wheal, one measurement of the diameter (in mm) is sufficient; if ovoid or irregular, it should be measured on the longest and shortest perpendicular axis and the numbers are added and divided by 2 (mean diameter). The flare may also be recorded by the same method. If flares are overlapping then only the width of the flare in the non-overlapping region need be recorded. The result should be recorded as a single figure each for wheal and flare in mm. Some would argue that only the wheal should be recorded since flares show greater variability of measurement by observers. Pseudopods (irregular linear extensions of the wheal) are not included in the measurement, but may be marked separately; however their significance is unknown.

Some practitioners advocate measuring the longest diameter; others use planimetry to produce a measurement in mm$^2$; however mean diameter is easily measured and should be considered the standard.

If the test has been carried out by a nurse or technician, it is important that the skin reactions should be inspected by the medical practitioner who ordered the test, to confirm the measurements and aid in interpretation, to monitor the quality of the test, and to determine whether any of the tests need to be repeated. For example, where there is an apparent discordance of results between allergens which are usually cross-reactive (eg. d.pteronyssinus and d.farinae, rye grass and timothy grass), there may be a false negative due to incorrect pricking of the extract and the discordant allergens should be repeated.

3.4.6 Method of recording skin prick test results

A chart should be kept and the wheal (and flare) size in mm recorded next to each allergen name. It is now considered an essential part of good clinical practice to record at least the wheal diameter in numerical form and to not use a qualitative marking (eg. +, ++) as the primary reported result (see later, “skin prick test reporting”.

3.4.7 Patient aftercare

Some patients experience considerable discomfort as a result of the itching of the skin test. Numbers should be removed from the skin, usually by cleaning with an alcohol solution (unless contraindicated by dry skin or a skin condition). Usually itching from skin prick testing subsides within 15 minutes or so. Some measures may be taken to reduce discomfort, including topical creams to reduce itching such as urea creams (Urex), or an ice-pack. Topical corticosteroids have been shown not to be useful$^{35}$. Some practitioners recommend an oral antihistamine. There is no evidence for the relative effectiveness of these approaches.

Patients should be warned that there is a possibility of a late-phase reaction (LPR), although this is relatively uncommon with prick tests (more common with intradermal testing). The significance of the presence of absence of the LPR in skin prick testing is unknown.
It is essential that the patient should receive counselling regarding the significance of the test results from the medical practitioner who ordered the test and receive information on any implications of the test, for example allergen avoidance etc.

3.4.8 Post-test holding time
Because of the small risk of a systemic reaction occurring after the test has been completed, it is recommended that some patients should remain in the medical rooms for a period afterwards. It is unnecessary to hold patients after a negative test, or where there have been only moderate skin prick test reactions to aeroallergens in a patient with no history of asthma. In the general setting, where there have been multiple positive results and there is a history of asthma or anaphylaxis, the patient should remain under observation for 40 minutes after the commencement of the test (~20 minutes after completion of the test). Where additional risk factors exist such as severe asthma, use of beta blockers, pregnancy, testing with foods, latex, or drugs, or intradermal testing (which would usually mandate that the test is carried out in a specialist setting) the 40 minute total observation time is essential.

3.5 Skin prick test result reporting
3.5.1 Reporting forms
Following skin prick testing a report should be generated which is clear, legible and enables communication of results to other practitioners. Skin prick test result forms should contain the following information:

- Name, address and contact information of the supervising practitioner (letterhead)
- Name and date of birth of patient
- Date of test
- Region tested (eg. back, arm)
- Name of technician who carried out test
- Name of each allergen tested- the correct name on the extract bottle should be used, followed by any common or local name. In Australia it can be assumed that the majority of tests are carried out using Hollister-Stier extracts; if extracts are obtained from another company, or are prepared on site, or use fresh or frozen substances, this should be recorded.
- If the form contains a long list of allergens, some of which were not tested, these should be crossed out since leaving a blank space next to the allergen may lead to the assumption that they were tested but were negative.
- If the allergen solution is diluted from the standard concentration supplied, this should be recorded.
- Negative and positive controls should be listed; the positive control (and its concentration) should be identified.
- Size of the resultant wheal for each allergen.

3.5.2 Standardised quantitative reporting
The primary result of the test is the wheal mean diameter in millimetres, according to the measurement method and at the time suggested above, and this should always be clearly recorded. The mean diameter of the flare may also be recorded but this is optional. These parameters should be recorded for negative and positive controls as well.
3.5.3 Qualitative reporting
Reporting of skin prick testing by qualitative measures (ie, 0, +, ++ etc) alone is not satisfactory; such interpretive reporting has been shown to be highly variable. Practitioners may find a qualitative scale to be clinically useful for test interpretation (eg. in distinguishing borderline results, indicating clinical significance) but this should be a secondary part of the report. Such qualitative assessments should always be made by the medical practitioner inspecting the results after measurement. If a qualitative scale is used then the scale should be printed on the report form.

3.5.4 Qualitative scales
Qualitative scales quoted in the literature are highly variable and hence may confound communication and interpretation of results. Because a number of different scales are used, qualitative results may mean different things to different people. In addition, the attachment of a qualitative statement to a report may convey an unintended meaning. For example a “+” reading may be technically positive but not clinically significant (see next section); a “-” report does not exclude sensitivity to the substance tested by other, non-IgE mediated mechanisms (for example a negative skin prick test for wheat does not exclude celiac disease). Therefore qualitative reporting is subject to misinterpretation by those who are not experienced or trained in allergy. Finally, some scales are intended for intradermal testing and misapplication to skin prick testing has led to further confusion in their use.
4. **INTERPRETATION OF SKIN PRICK TEST RESULTS**

4.1 **Meaning of “positive” and “negative” tests**

The result of a skin prick test may have significant ramifications to the patient’s lifestyle, diet, or occupation, and may determine prolonged courses of treatment and/or expensive environmental modification measures. The decision of whether a patient is truly allergic to the substance in question depends on careful interpretation of the SPT result as well as consideration of other clinical factors. Skin prick test results need to be interpreted in the context of the patient’s history, clinical signs, and allergen exposures. In the presence of a history of an allergic condition (such as those listed in part 2.1.1) with a positive skin prick test and known exposure to the allergen, particularly when the pattern of symptom exacerbation relates to variations in allergen exposure, it is reasonable to conclude that the allergen is relevant to the symptoms, and the positive test is significant.

A wheal of 3mm or greater is taken to indicate the presence of specific IgE to the allergen tested. When properly conducted, the skin prick test is a highly sensitive and specific test for the presence of allergen-specific IgE antibody. However, the presence of IgE antibody (as defined by a positive skin prick test) does not prove that the patient is clinically reactive to the allergen. The 3mm lower cutoff was determined because of reproducibility of measurement rather than clinical relevance. Studies have compared skin prick test results to the “gold standard” of clinical reaction to controlled challenge testing with the allergen. It is evident that in general, larger skin test reactions predict a higher likelihood of a positive response to a challenge, but do not predict severity of symptoms. These studies have indicated that for many allergens, a wheal size (lower cutoff) set at a larger size than 3mm would correlate better with clinical allergen reactivity. For example, a wheal size of >6mm may provide more specificity for the diagnosis of clinical dust mite allergy than the 3mm wheal. However, this remains to be firmly established; it will vary with different allergens, extracts from different sources, and different populations. Therefore a wheal of 3mm or greater is considered a positive skin prick test, but this must then be subjected to clinical interpretation.

Many precautions need to be taken in skin prick test interpretation:

- Positive tests (sometimes even with large wheal size) may occur without clinical symptoms. The test result indicates that IgE is present, therefore the test is technically positive, but symptoms may not occur on exposure to that allergen. This may be referred to as “clinically silent sensitisation”, or a “clinical false positive” test result (this individual may still be classified as atopic).
- The size of the skin prick test reaction may correlate with the likelihood that the patient is clinically reactive to that allergen. For example, in groups of patients, a subgroup with larger wheal size will contain a higher proportion of individuals who react to the allergen upon challenge than a subgroup with smaller wheal size.
- In general the size of the skin prick test reaction does not correlate with severity of the allergic manifestations.
- A positive skin prick test does not predict the nature of the allergic symptoms; different individuals with a positive test to the same substance may react in very different ways on exposure the allergen.
- Positive allergy tests may indicate a clinically true allergy but may be irrelevant (eg the patient is sensitized and clinically reactive but not exposed to that allergen, hence it is not the cause of their symptoms).
Skin prick tests may be positive when a patient has a previous history of allergy that has since resolved, for example hay fever may remit in adults but pollen skin tests often remain positive.

- Negative skin prick test results can occur even in the presence of true IgE-mediated allergy, due to inadequate representation of allergenic proteins in certain extracts.
- Negative skin prick tests in children do not rule out the possibility of the future development of allergy.
- Real false positive and false negative tests occur occasionally in clinical practice, for technical reasons or because of human error. Real false positive or false negative tests are defined by being non-reproducible in the same individual.
- Skin prick testing is not appropriate for the diagnosis of non-IgE mediated allergy or intolerance. In some cases it is clear from the history that the adverse reaction is not caused by type-1 (IgE-mediated) allergy. Negative skin tests in the presence of a good history of adverse reactions should prompt consideration of other mechanisms.
- When the skin prick test result is equivocal or does not correlate with the history, controlled challenge with the suspected allergen may be required (if clinically indicated and practical). Challenge testing is a specialized procedure.

### 4.2 Performance characteristics of skin prick testing

Theoretically skin prick testing is not a single test but a series of independent tests. Each test may have its own “performance characteristics” such as sensitivity, specificity, positive and negative predictive values etc. Ideally, the same rigor should be applied to technical aspects and interpretation of the results of skin prick tests as is applied to laboratory tests. Laboratory testing is subjected to strenuous quality control and ultimately, independent external assessment and accreditation; laboratory test results are evaluated with reference to populations of test subjects, and statistical analysis is used to determine the diagnostic significance of a test result at a particular level.

Studies evaluating the diagnostic utility of skin prick testing are of varying quality and frequently suffer from population selection bias, lack of appropriate gold standard, absence of blinding and absence of estimates of uncertainty. Published studies of skin prick test evaluation may be of great interest, but can be related only to the particular allergen and test method used. It is not advisable to directly translate wheal size in published studies to local practice unless the allergen extract is the same or is standardized, and the device, site of test and technique used is similar. Variability of skin prick test results using different devices and different brand extracts can be considerable and not only the size of the reaction but the result (eg positive/negative) can vary in the same individual²⁷,²⁸.

Evaluation of the performance of a test usually requires reference to a “gold standard”; for allergy tests this is usually the controlled challenge. There are a number of reasons why controlled challenges may not be entirely representative of natural exposure to the allergen. Nevertheless, challenge often allows figures such as positive and negative predictive value to be calculated. The positive predictive value is the probability that a positive test represents a true allergy. Many studies are emerging which attempt to determine the extent to which a particular wheal diameter can predict the risk of clinical reaction on challenge with a food. These studies have been used to suggest that challenge testing (in the case of suspected food allergy) may not be necessary to confirm the diagnosis when the wheal reaches a certain diameter²⁵. However it is crucial to recognize that the likelihood of true allergy for any given skin test size will depend on the pre-test probability that the study subject has the
allergy. For example the pre-test probability of peanut allergy is different in a child with a history of urticaria after eating nuts compared with a child who has eczema but no history of nut ingestion, in whom the test is performed for screening purposes. Therefore the predictive value varies in individuals with different histories, and may vary in hospital, specialist or general practice populations. A more useful figure is the likelihood ratio, which is a reflection of the degree to which the test result changes the probability that the patient has the allergy. These factors need to be taken into account not only in evaluating published studies but in applying the results of diagnostic testing to individual patients.

We should note that the importance of optimal interpretation of skin prick test results depends on the allergic condition in question and the allergen being tested. For example the erroneous interpretation of skin test results for aeroallergens in a patient with allergic rhinitis might result in inappropriate allergen avoidance strategies, which may be inconvenient, but erroneous interpretation of food allergy tests can have much more serious consequences such as inappropriate dietary restrictions which might be deleterious to health, or inappropriate exposure to foods which might be dangerous. Therefore, taken together with the fact that skin testing for food is inherently more difficult to interpret, we suggest that it should be restricted to specialist practitioners. When immunotherapy for inhalant allergens is being considered, the correct interpretation of skin prick test results becomes more critical since misdiagnosis may lead to inappropriate treatment, and again it should be carried out by specialists in these circumstances.

Therefore like any test used in clinical medicine the skin prick test is only one part of a comprehensive assessment of the patient and if the result is discordant with all of the other clinical indications, there may be grounds to repeat the test under different conditions or use another method (such as serum specific IgE [RAST] test, or diagnostic challenge). Interpretation of skin test results should be carried out by an experienced practitioner who is familiar with all of these factors.

4.3 Challenge tests
Challenge testing under controlled conditions can be used to confirm the presence of an allergy or rule it out, if the history and skin prick test results are not considered to be absolutely diagnostic. Challenge testing is also used in the research context with the specific purpose of validating the results of diagnostic tests. Detailed discussion of challenge tests are beyond the scope of this document. Challenge tests can be done by respiratory exposure (nasal or bronchial challenges) or using eyedrops of allergen solution (ocular challenge), generally with graded concentrations. Food challenges are done using graded amounts of food given orally. Challenge testing may also be done with drugs. The challenge is stopped once any objective reaction has occurred. Challenge testing is ideally done in a double-blinded fashion but open challenges are often used in clinical settings. Challenge testing, particularly for food and drugs, carries significant risk and must be done with full informed consent, under intense observation and monitoring, in a setting where all safety measures have been taken and equipment is readily available to treat any reactions including anaphylaxis.
5. PERSONNEL

Skin prick testing is routinely carried out (when indicated) by allergy specialists, where it is considered an extension of the physical examination. It is also carried out by general practitioners and other specialists (paediatricians, general physicians, thoracic physicians) who have an interest in allergy or where there are few allergy specialists available. In these circumstances it is therefore a POC (point of care) test, where the medical practitioner who is consulted by the patient provides the test and interprets the results. However there is currently no certification or accreditation for performance of this test. Skin testing carried out in a medical practitioner’s rooms should conform to the minimum and/or optimum standards for skin testing specified in this document (appendix 1) and endorsed by ASCIA. Skin prick testing is also carried out in some respiratory laboratories and pathology laboratories; the standards in appendix 1 should also apply in these settings.

5.1 Medical staff

Role of the medical practitioner in allergy skin prick testing:

- Ensure that an appropriate environment for skin prick testing is in place and that trained staff, equipment, reagents and facilities are available; according to standards set out in appendix 1.
- Assess the patient, history and examination, formulate a differential diagnosis, assess the likelihood of allergic disease, consider indications for skin prick testing, whether additional information is likely to be provided by skin prick testing and whether management will be altered by the results of skin prick testing.
- Carefully consider any contraindications or factors which might interfere with skin prick testing.
- Advise the patient of the procedure including risks and benefits.
- Decide on which allergens or panels of allergens should be tested, based on the symptom pattern, patient exposure, and using information about allergens in the local environment.
- Consider location to be tested, for example back, arms.
- In some cases the medical practitioner will personally carry out all steps of the skin prick test.
- If not carried out by the medical practitioner personally:
  - Advise paramedical staff of the test panel required and any patient characteristics that will need to be known to complete the test reliably and safely.
  - Be present and available in case of any adverse symptoms experienced by the patient.
  - Inspect the test site at the conclusion of the test to verify measurements taken by the person who carried out the test and determine whether there are any factors that might affect the interpretation of the results.
- Interpret the meaning of the measured results in the context of the clinical assessment.
- Consider whether technically positive skin test results are clinically important and whether negative test results are potentially false negative.
- Determine final diagnosis and management plan.
- Counsel the patient on the meaning of the results and their diagnosis and management.
Medical practitioners involved in allergy testing should maintain a good knowledge of allergic diseases, of allergens relevant in their area, and the significance of particular skin prick test reactions in relation to the condition in question. (An example might be the relative importance of allergy to dust mite, animals, pollens and foods in a case of atopic dermatitis). The evidence base for effectiveness or otherwise of allergen avoidance measures and immunotherapy must be taken into account when advising patients on management based on allergy test results.

5.2 Paramedical staff
Appropriately trained and experienced nursing staff and in some cases, technicians may play a role in certain aspects of the allergy skin test and resulting management.

Role of nurses or technicians in skin prick testing:
- Counsel the patient prior to the test on what to expect, put them at ease, position the patient appropriately and comfortably.
- Carry out the test according to the steps described above, ie apply numbers to skin, apply allergens, prick through, measure results.
- Management of the skin test record chart including patient details and recording results as described above.
- Monitoring patient for adverse reactions, reassurance regarding normal sensations.
- Aftercare of test site.
- Provision of patient education in allergen avoidance or Epipen use (on request by the medical practitioner, when indicated), if appropriately trained to do so.

5.3 Training
Allergy specialists (clinical immunologists and allergists) undergo extensive training at a postgraduate level under the College of Physicians and/or College of Pathologists, which includes proficiency and experience in all aspects of skin testing for allergy. There is no formal training for other specialists or general practitioners who conduct allergy testing. Allergy seminars and allergy testing workshops have been run from time to time by specialist units. A workshop on skin prick testing was held at the ASCIA Annual Scientific Meetings in 2005 and 2006.

Postgraduate training in allergy has been available for nurses from 2006 as part of the Allergy Nurses Course offered by UniSA (University of South Australia). This course covers a range of topics in allergy relevant to nurses, and include theoretical and practical training in skin testing, as well as hands-on training with a preceptor.

It is suggested that at least 10 skin tests over several days on a variety of patients should be carried out under supervision of an experienced nurse and allergy specialist to ensure basic competency. Evaluation of proficiency has been suggested by a test in which (for example) 10 histamine pricks are carried out on each of 5 different individuals, and the CV (SDx100/mean) should be less than 20%\(^1\). ASCIA plans to attempt to develop methods for proficiency testing and maintenance which will be presented in future versions of this manual.
6. SAFETY AND RISKS

6.1 Safety/risks of skin prick testing

Skin prick testing is an extremely safe procedure, with minimal discomfort. Rarely, adverse events can occur; these can be classified into allergic, test-related non allergic, and nonspecific. Examples of test-related non-allergic might include transmission of infection (theoretical but never documented); examples of nonspecific are syncope, headache etc. Vasovagal syncope is relatively common and if the test is done on the patient in the sitting position, facilities should be available for the patient to lie down if feeling faint.

The expected reaction to a skin prick test is a localised wheal and flare. Delayed local skin swelling (the late phase response) which is often tender or painful may occur uncommonly as a result of an IgE-mediated late-phase reaction (seen more commonly with intradermal testing). Rarely this can cause quite marked swelling and discomfort, however it does not usually last more than 36 hours.

Systemic introduction of allergen may occur as an unintended consequence of the skin prick. Systemic reactions from skin prick testing have been recorded, including the typical manifestations of anaphylaxis such as generalised urticaria, angioedema including airway angioedema, bronchospasm, and hypotension. These reactions are generally mild and respond to treatment with standard measures. There are many case reports of systemic allergic reactions from prick testing\(^{30}\) (Liccardi 2006) although in large case series this is exceedingly rare. In a survey of 16,000 individuals tested with eight routine allergens, the rate of adverse reactions was 0.04%\(^{31}\) but most of these were syncope, near-syncope or malaise. In another large survey, the rate of systemic allergic reactions was 0.033%, all occurring in asthmatics\(^{32}\). A small number of fatalities are recorded as a result of intradermal skin tests; there is only one reported fatality from skin prick testing (however this was an atypical case and many of the risk factors mentioned below were present)\(^{14}\). Rarely, delayed systemic reactions in association with large late-phase responses have been reported; these usually consist of wheezing in asthmatic patients who had strongly positive skin prick tests (unpublished personal communications) commencing several hours after the test. All asthmatics should have an appropriate action plan in place, particularly where there are multiple strong positive skin prick test reactions.

Case reports or small series describing anaphylaxis from skin testing have suggested certain risk factors. Amongst a paediatric population, systemic reactions occurred exclusively in infants <6 months of age with atopic dermatitis, when tested with fresh food allergens\(^{33}\). Further case reports suggest that a history of anaphylaxis to food, particularly when testing with fresh food allergens and multiple allergens, is a risk factor\(^{34}\). Systemic reactions from skin testing with latex extracts have been well described.
Putative risk factors for anaphylaxis in skin prick testing:

<table>
<thead>
<tr>
<th>Risk Factor</th>
</tr>
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<tbody>
<tr>
<td>Less than 6 months of age (though possible at any age)</td>
</tr>
<tr>
<td>Previous history of food anaphylaxis, testing with foods</td>
</tr>
<tr>
<td>Testing with fresh foods, non-commercial extracts</td>
</tr>
<tr>
<td>Testing with latex allergens</td>
</tr>
<tr>
<td>Asthma, particularly if active or unstable</td>
</tr>
<tr>
<td>Widespread atopic dermatitis in children</td>
</tr>
</tbody>
</table>

It should be noted that since atopic dermatitis and asthma are very common and systemic reactions are extremely rare, the presence of atopic dermatitis and/or asthma should not preclude skin testing in the appropriate setting.

### 6.2 Safety measures and safety equipment required

Skin prick testing must always be performed in a medical setting with the ready availability of medical practitioners competent to treat systemic allergic reactions, and appropriate equipment. It is recommended that patients who have undergone skin prick testing and have positive results, who have asthma or a history of anaphylaxis, should remain in the centre for at least 20 minutes following completion of the skin prick test (total of 40 minutes after skin pricking).

**Suggested minimum standards for available emergency equipment and medications:**

- Availability of oxygen, 6l/min via mask
- Facility for intravenous cannulation and intravenous fluids for rapid infusion in case of hypotension.
- Ready availability of adrenaline for intramuscular injection.
- Salbutamol via nebuliser or spacer.

*Detailed information on the treatment of systemic allergic reactions and anaphylaxis are beyond the scope of this document.*
7. REGULATORY ISSUES

The following points apply to regulations in Australia and the situation in New Zealand is significantly different.

7.1 Positive Control Solutions

Products that are sold for use in human skin prick allergy testing are regulated by the Therapeutic Goods Administration (TGA), which is responsible for all therapeutic goods (drugs or devices) used in Australia.

7.1.1 Histamine

There is no histamine solution registered or marketed in Australia. Since positive controls are essential, this creates a difficult situation for some practitioners. Currently there are two ways in which histamine solutions can be legally obtained:

- **Histamine can be supplied by a hospital pharmacy as an extemporaneous preparation which can be used by doctors, on specific patients for use within the hospital.** In theory good clinical practice would dictate that a prescription for each individual tested would be required by the hospital pharmacy, but this is not a legislative requirement.

  \[ \text{Whether histamine can be legally dispensed from a hospital pharmacy for use in private practice within the same state or territory will depend on state health regulations. It cannot be sent to another state or territory.} \]

- **Histamine can be obtained from an overseas supplier via the TGA Special Access Scheme (SAS), e.g. Positive Skin Test Control – Histamine® manufactured by Hollister-Stier, USA, or Stallergenes Histamine Hydrochloride 10mg/ml; i.e.**

  Doctors can apply for a Category B SAS approval from the TGA [Therapeutic goods act (1989) section 19(1)]. Approval is given for a single patient, on a case-by-case basis. Applications should be made in writing, preferably on a Category B form, available from Therapeutic Goods Administration (TGA) website [www.tga.gov.au/hp/sas.htm](http://www.tga.gov.au/hp/sas.htm)

or

Doctors can apply for an authority to use a specific drug or class of drugs without the need for prior approval in respect of each patient, i.e. Authorised Prescriber [Therapeutic goods act (1989) section 19(5)]. Endorsement from an Ethics Committee (or if this is not available, a specialist college) must be obtained prior to completing an “Agreement to Treatment Directions Authorisation of Prescribers Under Section 19(5) of the Therapeutics Goods Act 1989” form. Records must be kept of the number of patients who receive the product. Information about this is available on the Therapeutic Goods Administration (TGA) website [www.tga.gov.au/hp/ap.htm](http://www.tga.gov.au/hp/ap.htm)

\[ \text{In both cases written consent is required and patients should be monitored appropriately to determine both efficacy and the occurrence and severity of any adverse drug reactions.} \]

7.1.2 Codeine phosphate

Codeine phosphate is an alternative positive control, in a solution of 9%. Currently, there is no marketed codeine phosphate solution for skin prick allergy testing available in Australia. However, it can be obtained on prescription for an individual patient from a pharmacy (i.e. extemporaneous preparation by a registered pharmacist). Each patient tested should have a prescription submitted. Skin prick testing is a non-approved use of a marketed product and
therefore the manufacturer assumes no liability for its use; the liability is assumed entirely by the prescriber.

7.2 **Allergen solutions**

Allergen extracts manufactured for the purpose of skin prick testing by Hollister-Stier are currently registered for use in Australia, except for food allergens, as Hollister-Stier ceased producing these in 2008.

Skin testing extracts from Stallergenes (Alyostal) and ALK-Abello are unregistered skin prick testing allergens which are available under Section 19(1) and/or Section 19(5) of the Therapeutic Goods Act 1989. The options for accessing these allergens are as follows:

1. **Section 19(5): Obtain “Authorised Prescriber status” by endorsement from a medical College.** ASCIA has obtained endorsement for its Full (Ordinary) members from the Royal Australasian College of Physicians (RACP) and the Royal College of Pathologists of Australasia (RCPA), on behalf of its Full (Ordinary) members who are Fellows of the RACP or RCPA for the full ranges of Stallergenes (Alyostal) and ALK-Abello skin prick testing reagents.

2. **Section 19(5): Obtain “Authorised Prescriber status” by endorsement by an ethics committee.** This is possible for some but not all doctors who work outside hospitals/universities.

3. **Additional provisions (Section 19A) that allow another manufacturer to supply reagents to a market where a currently registered reagent is no longer available.** This option is available for ALK-Abello food allergen extracts, which have been discontinued by Hollister Stier. As it does not require Authorised Prescriber status it is open to all medical practitioners, without endorsement.

Therefore if a doctor is ineligible for Authorised Prescriber endorsement they can still access skin prick testing reagents by using Option 3 to obtain food allergens from ALK-Abello and order other allergens from **Hollister Stier, which are TGA registered.**


7.3 **Skin prick test devices**

This section is yet to be developed.

7.4 **Personnel carrying out the test**

Certain restrictions apply if the patient is intending to claim a rebate from Medicare for the performance of an allergy test. The following items from the Medicare Benefits Schedule apply:

12000 SKIN SENSITIVITY TESTING for allergens, USING 1 TO 20 ALLERGENS
12003 SKIN SENSITIVITY TESTING for allergens, USING MORE THAN 20 ALLERGENS

With regard to who actually carries out the test, the following is stated in the MBS schedule “general explanatory notes” section 12:

12.1 Professional Services
12.1.1 Professional services which attract Medicare benefits include medical services rendered by or on behalf of a medical practitioner. Medical services which may be rendered "on behalf of" a medical practitioner include services where a portion of the service is performed by a technician employed by or, in accordance with accepted medical practice, acting under the supervision of the medical practitioner.

12.1.2 The health insurance regulations specify that the following medical services will attract benefits only if they have been personally performed by a medical practitioner on not more than one patient on the one occasion (i.e. two or more patients cannot be attended simultaneously although patients may be seen consecutively), other than an attendance on a person in the course of a group session (i.e. Items 170-172). The requirement of "personal performance" is met whether or not assistance is provided in the performance of the service according to accepted medical standards:

(a) All Category 1 (Professional Attendances) items (except 170-172, 342-346);

(b) Each of the following items in Group D1 (Miscellaneous Diagnostic):- 11012, 11015, 11018, 11021, 11212, 11304, 11500, 11600, 11601, 11627, 11701, 11712, 11724, 11921, 12000, 12003;

However a ruling has previously been sought which clarifies this issue: “The requirement that skin sensitivity tests be performed by the doctor is fulfilled when the medical practitioner attends the patient to take, or to review, the history and to decide if, and which, allergies are to be tested, and afterwards again attends the patient for interpretation of the results. The scratch or patch tests may be applied or the intradermal allergen may be injected by a technician or nurse.” (Director-General of Health, Department of Health, 22 Feb 1984).

Therefore, as long as a medical practitioner reviews the history and orders the panel, and attends the patient to interpret the results, then the patient is entitled to a rebate from Medicare for the service provided by that practitioner.

7.5 Consent
In most practices, patient informed consent has not routinely been sought for skin prick testing. If unregistered commercial histamine or unregistered allergen extracts [under section 19(1) or 19(5)] are used, informed patient consent is mandatory according to TGA regulations. The basis for this consent is not any actual increase in risk from the use of these products, but rather a notification to the patient that “the product is not approved in Australia by the TGA and therefore that the Commonwealth can give no guarantee as to its safety, quality or efficacy and accordingly the Commonwealth can accept no liability for its use”.
REFERENCES
2. Portnoy J. Diagnostic testing for allergies. Ann Allergy Asthma Immunol 2006; 96:3-4
5. McCann WA, Owby DR. The reproducibility of the allergy skin test scoring and interpretation by board-certified/board eligible allergists. Annals of Allergy, Asthma and Immunology 89:368-371 2002
18. Dreborg S (editor), Subcommittee on skin tests of the European Academy of Allergology and Clinical Immunology. Skin tests used in type 1 allergy testing- Position paper. Allergy 1989; Supplement 10, 44:1-59
20. Piette V, Bourret E, Bousquet J, Demoly P. Prick tests to aeroallergens: Is it possible simply to wipe the device between tests? Allergy 2002 57:940-942
23. Bernstein IL, Storms W, and the Joint Task Force on Practice Parameters, American Academy of Allergy, Asthma and Immunology (AAAAI) and the American College of Allergy, Asthma and Immunology (ACAAI): Practice parameters for allergy diagnostic testing. Annals of Allergy, Asthma and Immunology, 1995 75:543-624
26. Clark AT, Ewan PM. Interpretation of tests for nut allergy in one thousand patients, in relation to allergy or tolerance. Clin Exp Allergy 2003; 33:1041-1045
REVIEWS ACCESSED IN FULL FOR THE PREPARATION OF THIS MANUAL:

Dreborg S (editor), Subcommittee on skin tests of the European Academy of Allergology and Clinical Immunology. Skin tests used in type 1 allergy testing- Position paper. Allergy 1989; Supplement 10, 44:1-59

Bernstein IL, Storms W, and the Joint Task Force on Practice Parameters, American Academy of Allergy, Asthma and Immunology (AAAAI) and the American College of Allergy, Asthma and Immunology (ACAAI): Practice parameters for allergy diagnostic testing. Annals of Allergy, Asthma and Immunology, 1995 75:543-624


Turkeltaub PC. Percutaneous and intracutaneous diagnostic tests of IgE-mediated diseases (immediate hypersensitivity). Clin Allergy Immunol 2000; 15:53-87


O’Brien, RM. Skin prick testing and in-vitro assays for allergic sensitivity. Australian Prescriber 2002 25; 91-93

APPENDIX 1

STANDARDS FOR SKIN PRICK TESTING

This set of standards is based on ASCIA expert consensus as well as published evidence where available. It should be taken in conjunction with the ASCIA skin prick testing manual which provides full explanation, references and justification.

MINIMUM STANDARDS FOR SKIN PRICK TESTING

1. Patients must be screened for suitability for skin prick testing by a medical practitioner taking into account indications and contraindications.
2. Asthma, pregnancy, beta-blocker use are relative contraindications to skin prick testing.
3. Allergens to be tested (or panels of allergens) must be ordered individually based on patient history and exposure.
4. Patients should not be tested if they have recently taken antihistamines or other medications which interfere with the test response, or on skin with active dermatitis or open lesions.
5. A medical practitioner must be present on the premises during the conduct of the procedure.
6. Appropriate medications and equipment to treat anaphylaxis must be readily available.
7. The test must be conducted by a practitioner (nurse or doctor) who has training and experience.
8. The test must incorporate a positive control (histamine or codeine) and an appropriate negative control.
9. Test sites should be at least 2cm apart.
10. The test result should be inspected and measured at 15 minutes.
11. The diameter or mean diameter of the wheal must be recorded as the primary result of the test.
12. A wheal of 3mm is the minimum size to be considered a positive result.
13. An experienced medical practitioner should inspect the results of the test.
14. Allergen extracts should be obtained from reliable commercial sources, stored at 2-8°C when not in use, and discarded after the use-by date.
15. Appropriate devices should be used for skin pricking (hypodermic needles are not suitable).
16. Sharps must be disposed of appropriately, with universal precautions for infection control observed.
17. A report should be provided stating the supervising medical practitioner, the patient’s name, the date, the site used, the allergens tested, and results as wheal diameter (in mm).
18. Results must be interpreted in the context of the patient’s history.
19. Post-test counseling must be provided based on the results.
20. Patients with a history of asthma who have positive skin prick test results should be observed for at least 20 minutes after completion of the test. This holding time is also applied in any other higher risk patients (see below, "skin test procedures that should only be conducted by allergy specialists"). If the test is negative, or positive
for aeroallergens where there is no history of asthma, a holding time is not mandatory.

Minimum requirement to claim a Medicare rebate for allergy skin prick testing:
In order to claim the item, the medical practitioner must review the patient history and order the panel, and attend the patient to interpret the results. (HIC advice).

OPTIMUM STANDARDS FOR SKIN PRICK TESTING

1. The same medical practitioner who orders the test should inspect the results and provide post-test counselling (or the medical practitioner may conduct the entire test).
2. A new pricking device should be used for each allergen and control.
3. Standardised extracts should be used where possible.
4. The flare diameter as well as the wheal should be recorded.
5. The histamine result should be read at 10 minutes, the allergens at 15-20 minutes.
6. Following the test, comprehensive patient education on allergen avoidance should be provided if indicated.

SKIN TEST PROCEDURES THAT SHOULD ONLY BE CONDUCTED BY ALLERGY SPECIALISTS OR EQUIVALENTLY TRAINED MEDICAL PRACTITIONERS

1. Skin prick testing for foods, particularly fresh foods.
2. Skin prick testing for latex allergy and drug allergy.
3. Intradermal skin tests (drugs, venoms).
4. Skin prick testing on infants <2 years.
5. Skin testing in the presence of relative contraindications such as pregnancy, use of beta-blockers, severe or unstable asthma.

SKIN TEST PROCEDURES THAT ARE USUALLY INAPPROPRIATE/CONTRAINDICATED

1. Intradermal skin testing for foods (very high risk), aeroallergens (lack specificity).
2. Skin tests are not indicated for the diagnosis of food intolerance, adverse reactions to food additives, and allergy to most drugs.
## APPENDIX 2

**DRUGS WHICH ARE ANTIHISTAMINES OR HAVE ANTIHISTAMINE ACTIVITY AND WHICH MAY INTERFERE WITH SKIN TESTING**

<table>
<thead>
<tr>
<th>Antihistamines</th>
<th>Withholding period</th>
<th>Comment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Generic</td>
<td>Commercial*</td>
<td></td>
</tr>
<tr>
<td>Azatidine</td>
<td>Zadine</td>
<td>2</td>
</tr>
<tr>
<td>Brompheniramine</td>
<td>Dimetapp (some)</td>
<td>5</td>
</tr>
<tr>
<td>Cetirizine</td>
<td>Zyrtec, Zilarex, Xyzal, other OTC</td>
<td>4</td>
</tr>
<tr>
<td>Chlorpheniramine</td>
<td>numerous OTC</td>
<td>4</td>
</tr>
<tr>
<td>Cyproheptadine</td>
<td>Periactin</td>
<td>4</td>
</tr>
<tr>
<td>Desloratadine</td>
<td>Aerius</td>
<td>4</td>
</tr>
<tr>
<td>Dexchlorpheniramine</td>
<td>Polaramine</td>
<td>4</td>
</tr>
<tr>
<td>Diphenhydramine</td>
<td>Unisom Sleepgels, other OTC</td>
<td>2</td>
</tr>
<tr>
<td>Dimenhydrinate</td>
<td>Dramamine, Travacalm</td>
<td></td>
</tr>
<tr>
<td>Doxylamine</td>
<td>numerous OTC</td>
<td>2</td>
</tr>
<tr>
<td>Fexofenadine</td>
<td>Telfast, Fexal, other OTC</td>
<td>4</td>
</tr>
<tr>
<td>Loratadine</td>
<td>Claratyne, Lorano, other OTC</td>
<td>10</td>
</tr>
<tr>
<td>Mepyramine</td>
<td>Relaxa-tabs</td>
<td></td>
</tr>
<tr>
<td>Methdilazine</td>
<td>Dilosyn</td>
<td>3</td>
</tr>
<tr>
<td>Pheniramene</td>
<td>Avil, Fenamine</td>
<td>4</td>
</tr>
<tr>
<td>promethazine HCl</td>
<td>Phenergan, other generic</td>
<td>4</td>
</tr>
<tr>
<td>trimeprazine</td>
<td>Vallergan, other generic</td>
<td>2</td>
</tr>
<tr>
<td>triprolidine</td>
<td>numerous OTC</td>
<td>1</td>
</tr>
<tr>
<td><strong>H-2 antagonists</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>cimetidine</td>
<td></td>
<td>1</td>
</tr>
<tr>
<td>ranitidine</td>
<td></td>
<td>1</td>
</tr>
<tr>
<td>famotidine</td>
<td></td>
<td>1</td>
</tr>
<tr>
<td><strong>Antidepressants</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>amitriptyline</td>
<td>Endep, Tryptanol</td>
<td></td>
</tr>
<tr>
<td>clomipramine</td>
<td>Anafranil, Clopram, Placil, generic</td>
<td></td>
</tr>
<tr>
<td>dothiepin</td>
<td>Dothep, Prothiaden</td>
<td></td>
</tr>
<tr>
<td>doxepin</td>
<td>Depran, Sinequan</td>
<td>7</td>
</tr>
<tr>
<td>imipramine</td>
<td>Melipramine, Tofranil</td>
<td></td>
</tr>
<tr>
<td>mianserin</td>
<td>Lumin, Tolvon</td>
<td></td>
</tr>
<tr>
<td>mirtazapine</td>
<td>Avanza, Mirtazon, Remeron</td>
<td></td>
</tr>
<tr>
<td>nefazodone</td>
<td>Serzone</td>
<td></td>
</tr>
<tr>
<td>nortriptyline</td>
<td>Allegron</td>
<td></td>
</tr>
<tr>
<td>trimipramine</td>
<td>Surmontil</td>
<td></td>
</tr>
<tr>
<td><strong>Anti-migraine</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>diphenhydramine</td>
<td>Ergodryl</td>
<td></td>
</tr>
<tr>
<td>pizotifen</td>
<td>Sandomigran</td>
<td></td>
</tr>
<tr>
<td><strong>Anti-emetics</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>prochlorperazine</td>
<td>Stemetil, Stemzine</td>
<td>weak antihistamine</td>
</tr>
</tbody>
</table>
### Neuroleptics

<table>
<thead>
<tr>
<th>Neuroleptics</th>
<th>Brand Names</th>
<th>Withholding period</th>
</tr>
</thead>
<tbody>
<tr>
<td>chlorpromazine</td>
<td>Largactil</td>
<td>not established, may be up to 2 weeks or more.</td>
</tr>
<tr>
<td>clozapine</td>
<td>Clopine, Clozaril, generic</td>
<td></td>
</tr>
<tr>
<td>flupenthixol</td>
<td>Fluanxol**</td>
<td></td>
</tr>
<tr>
<td>fluphenazine</td>
<td>Anatensol, Modecate, generic</td>
<td></td>
</tr>
<tr>
<td>olanzapine</td>
<td>Zyprexa</td>
<td></td>
</tr>
<tr>
<td>pericyazine</td>
<td>Neulactil**</td>
<td></td>
</tr>
<tr>
<td>quetiapine</td>
<td>Seroquel</td>
<td></td>
</tr>
<tr>
<td>risperidone</td>
<td>Risperdal</td>
<td></td>
</tr>
<tr>
<td>thioridazine</td>
<td>Aldazine, Melleril</td>
<td></td>
</tr>
<tr>
<td>trifluoperazine</td>
<td>Stelazine</td>
<td></td>
</tr>
<tr>
<td>zuclopenthixol</td>
<td>Clopixol**</td>
<td></td>
</tr>
</tbody>
</table>

* OTC- various brand names available over the counter (S2, S3) in pharmacies; check labels.

** Antihistamine effect not formally demonstrated but thought likely due to structural and functional similarities with other drugs.
APPENDIX 3

IMPORTERS/DISTRIBUTORS OF SKIN PRICK TESTING REAGENTS IN AUSTRALIA (as of March 2009)

Link Medical Products Pty Ltd

Website: www.linkpharma.com.au
Email: allergy@linkpharma.com.au or jhillas1@bigpond.com
Address: Unit 18, 6A Prosperity Parade Warriewood NSW 2102 Australia
Postal address: PO Box 135 Avalon NSW 2107 Australia
Telephone: 1800 824 166 (toll-free)
Facsimile: 1800 824 199 (toll-free)

Supplier of:
Hollister-Stier skin prick testing extracts (registered)
Stallergenes Alyostal skin prick testing reagents (currently unregistered) *

Australasian Medical & Scientific Limited (AMSL)

Website: www.amsl.com.au
Email: amsl@amsl.com.au or orders@amsl.com.au
Address: Unit 2, 19-21 Gibbes Street Chatswood NSW 2067 Australia
Postal address: PO Box 5197 Chatswood West NSW 1515 Australia
Telephone: 02 9882 3666
Facsimile: 02 9882 3999

Supplier of:
ALK-Abello skin prick testing extracts (currently unregistered) *

* NOTE - See section 7.2 for information about accessing unauthorized products.