Jean-Marie Lachapelle
Howard I. Maibach

Patch Testing and Prick Testing

A Practical Guide
Second Edition

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A Practical Guide
Official Publication of the ICDRG

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Preface to 2nd Edition

This second edition has been expanded taking into account the progress made in the field of contact dermatitis during the last 5 years.

The realm of dermato-allergology is constantly on the move; this implies a better knowledge of the mechanisms involved, improvements of patch and prick testing procedures, and adaptations of lists of allergens in relation with the ongoing changes in our environment.

The number of tables, flowcharts, and illustrations has been increased to offer more accurate guidelines for all practicing dermatologists.

J.-M. Lachapelle
H.I. Maibach

Preface to 1st Edition

This small book is a follow-up to the classic Manual of Contact Dermatitis by Siegfried Fregert, which was published on behalf of the International Contact Dermatitis Research Group and the North American Contact Dermatitis Group.

The format follows the succinct presentation of Professor Fregert. Every emphasis has been made on balancing brevity and clarity with sufficient details for the beginner in the field of diagnostic patch and prick testing.

Brevity is valued by the beginner. Fortunately, several major textbooks including those by Cronin, Kanerva, Rycroft, and Fisher are available and provide for the second level of detail.

The authors would greatly appreciate any corrections and suggestions – for future editions.

J.-M. Lachapelle
H.I. Maibach

Contents

List of Abbreviations

ACD	Allergic contact dermatitis
ACDS	Allergic contact dermatitis syndrome
AD	Atopic dermatitis
APT	Atopy patch test
CAD	Chronic actinic dermatitis
CADR	Cutaneous adverse drug reaction
CR	Current relevance
CUS	Contact urticaria syndrome
DMSO	Dimethylsulphoxide
EECDRG	European and Environmental Contact Dermatitis Research Group
EFTAD	European Task Force on Atopic Dermatitis
ESCD	European Society of Contact Dermatitis
ESS	Excited skin syndrome (angry back)
FDA	Food and Drug Administration
ICD	Irritant contact dermatitis
ICDRG	International Contact Dermatitis Research Group
ICU	Immunological contact urticaria
IDT	Intradermal test
IFRA	International Fragrance Association
IgE	Immunoglobulin E
IR	Index réactif
J	Joules
JCDS	Japanese Society for Contact Dermatitis
MED	Minimum erythema dose
NACDG	North American Contact Dermatitis Research Group
NICU	Non-immunological contact urticaria
NSAIDs	Non-steroidal anti-inflammatory drugs
PACD	Photoallergic contact dermatitis
PCD	Protein contact dermatitis

PLE	Polymorphic light eruption
PLR	Persistent light reactions (actinic dermatitis, actinic reticuloid)
PNU	Protein nitrogen units
PPT	Photopatch test
PR	Past relevance
PT	Patch test
PUT	Provocative use test
PVA	Polyvinyl alcohol
RAST	Radioallergosorbent test
ROAT	Repeated open application test
RCT's	Randomized controlled clinical trials
SAFT	Skin application food test
SDRIFE	Symmetrical drug-related intertriginous and flexural exanthema
SRCD	Systemic reactivation of contact dermatitis

The International Contact Dermatitis Research Group

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J.-M. Lachapelle and H.I. Maibach

1.1 Historical Background

The International Contact Dermatitis Research Group (ICDRG) was founded in 1967. It was (and still is) an informal association, without any statutes.

The founding members of the group were 11: C.D. Calnan, E. Cronin, D.S. Wilkinson (United Kingdom); N. Hjorth (Denmark); V. Pirilä (Finland), H.J. Bandmann (Germany); C.L. Meneghini (Italy); K.E. Malten (Holland); S. Fregert and B. Magnusson (Sweden). Niels Hjorth acted as Chairman of the Group.

The main aim of the group was to provide a standardization of Routine Patch Testing [1]. This standardization did not exist at the time "As long as clinics used different techniques, substances, concentrations and vehicles for testing, results obtained at various clinics in different countries could not be compared" [2]. The members of the ICDRG conducted extensive joint studies, and this resulted in the production of the so-called ICDRG standard series, known and used throughout the world.

The ICDRG promoted the foundation of several contact dermatitis national and international groups. This goal was reached in the 1980s [3].

Some groups, e.g., the European and Environmental Contact Dermatitis Research Group (EECDRG) and the North American Contact Dermatitis Group (NACDG) took over the task of standardization of series of allergens. In the meanwhile, Working Parties, created by the European Society of Contact Dermatitis (ESCD), conducted joint studies, leading to a continuous program of updated lists of additional series of patch tests. Furthermore, a similar task was achieved in different countries by national groups, which adapted series of tests to local needs, in relationship with the specific environment encountered in each country.

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1.2 Current Tasks of the ICDRG

The current tasks adopted by the present ICDRG committee are the following:

- To promote the dissemination of our knowledge in the field of environmental dermatology (with a special interest for contact dermatitis). This goal is reached by the organization of international symposia (on a 2-year schedule). The aim of the symposia is to allow dermatologists, occupational physicians, chemists, and pharmacists to be acquainted with updated information. The symposia are organized in different parts of the world.

  The strategy is focused on the following:

  a. Keynote lectures, pointing out the more recent advances in the field of contact dermatitis and other related problems
  b. Courses, mainly aimed to promote basic knowledge among participants, who are not acquainted with the "tricks" of the discipline

- To promote the publication of manuals, which are of practical use for practicing dermatologists and occupational physicians [4, 5].

1.3 ICDRG Members

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References

1. Lachapelle JM (2006) Historical Aspects. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis. 4th edn. Springer, Berlin, pp 1–7
2. Calnan CD, Fregert S, Magnusson B (1976) The International Contact Dermatitis Research Group. Cutis 18:708–710
3. Bruynzeel DP (2006) Contact Dermatitis Research Groups. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis. 4th edn. Springer, Berlin, pp 903–906
4. Lachapelle JM, Maibach HI (2009) Patch Testing/Prick Testing. A Practical Guide. Official Publication of ICDRG, 2nd edn. Springer, Berlin
5. Wahlberg JE et al. (eds) (2003) Management of Positive Patch Test Reactions, Springer, Berlin, 116 p

Part I

Patch Testing

The Spectrum of Diseases for Which Patch Testing is Recommended

2

Patients Who Should be Investigated

J.-M. Lachapelle

2.1 Allergic Contact Dermatitis

Allergic contact dermatitis (ACD) is observed in daily life by the practicing dermatologist. It is noteworthy that, in the vast majority of cases, its clinical presentation is an eczematous reaction. ACD is therefore synonymous with contact eczema.

2.1.1 Pathomechanisms in Allergic Contact Dermatitis

T. Rustemeyer

ACD is a T cell-mediated, delayed-type hypersensitive immune response induced by contact allergens. Although innate immunity plays a role in ACD, it is primarily mediated by an adaptive T cell-mediated immune response and, hence, can be divided into a sensitization phase and an elicitation phase.

2.1.1.1 Sensitization Phase

Contact allergens are small molecular weight chemicals, also termed haptens, which can easily penetrate the epidermal barrier. Penetration of the epidermal barrier can be facilitated by the irritant properties of the allergen itself or by the concomitantly present irritants. Penetrated allergens diffusely distribute into the skin due to their frequently lipophilic nature. The vast majority of contact allergens are too little to induce specific immune reactivity themselves. The chemically reactive haptens react first with various extracellular and cell-membrane-associated self-proteins (altered self), forming a neo-antigen ("hapten-carrier complex"), which can elicit a specific immune response. Hereto, hapten-carrier complexes have to stimulate professional antigen-presenting cells (dendritic cells) of the epidermis, called Langerhans cells, and/or the dermis, called dermal dendritic

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cells. Following encounter with an immunostimulatory allergen, dendritic cells become activated and undergo maturation.

These processes are stimulated by the release of pro-inflammatory mediators ("danger signals"), such as IL-1α, IL-6, and TNF- α, from residual cells (e.g., keratinocytes) and dendritic cells. Under the influence of IL-1β, TNF-α, and GM-CSF, the matured antigen-loaded dendritic cells emigrate from (epi)dermal tissues towards the draining lymph node. Hereto, they loose adherence to surrounding keratinocytes by downregulating the expression of E-cadherin, by upregulating the expression of basement membrane-dissolving enzymes, for example, MMP-9, and by the expression of chemokines receptors, in particular CXCR4 and CCR7. Following the chemotactic gradient of the CCR7 ligands CCL19 and CCL21 matured antigen-loaded dendritic cells reach the draining lymph node in less than 24 h. During their migration, matured dendritic cells upregulate the expression of antigen-presenting MHC molecules ("signal 1" of priming of antigen-specific T cells) and the so-called co-stimulatory molecules, such as CD54, CD80, CD83, and CD86 ("signal 2"). In the draining lymph node, they settle in the T-cell-rich paracortical areas and regain long dendrites, enabling the contact with randomly bypassing naive T cells. Next to the matured dendritic cells, naive T cells express the chemokine receptor CCR7 and, thus, both cell types are brought in contact attracted by the same chemokines. In the presence of the appropriate antigen and sufficient co-stimulatory signals, $CD45RA^+$ naive T cells can get activated, start secretion of IL-2 and proliferation. Thereby, they loose $CD45RA^+$ expression and acquire $CD45R0^+$ effector/memory phenotype. If the antigen is presented in the context of MHC class-I molecules, emerging allergen-specific T cells then belong to the $CD8^+$ population, whereas $CD4^+$ T cells can recognize antigen presented by MHC class-II molecules. Depending on further soluble and membrane-bound mediators (e.g., polarizing cytokines and stimulatory molecules), distinct subsets of primed T cells can be formed. In the presence of, for example, IL-12 and CD40-CD40 ligand interaction, T cells get polarized towards the Th1 cytokine-secreting profile characterized by the secretion of, for example, TNF-α and IFN-γ. In contrast, the presence of IL-4 in the lack of IL-12 leads to Th2 cytokine-secreting T cells characterized by the secretion of, for example, IL-4, Il-5, and IL-13. Both T cell types secrete inflammatory mediators, of which the former is associated with classical delayed-type hypersensitivity reactions and the latter among others with immediate-type allergy and atopic dermatitis. Only recently, Th2 cytokines were also identified to plat a role in ACD. Also, the newly described Th17 cells mainly secreting IL-17 and IL-22 can act as an effector T cell in ACD. Upon priming, certain T cells retain CCR7 expression. They belong to the central memory T cell pool and recirculate in the bloodstream and can migrate again to the primary lymphatic tissues. This T cell population represents the long-living immunological memory. Primed T cells that downregulate the chemokine receptor CCR7 belong to the pool of effector memory T cells. These T cells primed in skin draining lymph nodes start to express the skin homing molecule CLA (cutaneous lymphocyte-associated antigen), which enables effector memory cells to leave dermal blood vessels and to control skin tissues ("immunosurveillance"). The sensitization phase lasts for 10–15 days and is, except from an occasionally observed cutaneous lymphadenopathy, usually asymptomatic.

2.1.1.2 Elicitation Phase

Although sensitization can be clinically unapparent, repeated contacts with the specific allergen in the sensitized individuals can lead to ACD. For the initiation and amplification of the immune response, also participation of resident cells, in particular keratinocytes, mast cells, and endothelial cells, as well as mediators of the innate immunity are required. Allergen-exposed keratinocytes, fibroblasts, and other residual cells secrete pro-inflammatory cytokines (IL-1α, IL-6, TNF-α, and others). Along with the leakage of serum, these mediators stimulate the expression of adhesion molecules on dermal endothelial blood vessels. The increased expression of integrins, selectins, and chemokine receptors on endothelial cells facilitates unspecific extravasation of leukocytes from the blood flow and infiltration of the allergen-exposed skin sites. Among the cellular infiltrate, in particular CLA^+ T cells co-expressing $CXCR3^+$, $CCR4^+$, and $CCR10^+$ are attracted by the (epi)dermal secretion of their inflammatory chemokine ligands CXCL9-11, CCL17/22, and CCL27, respectively. If allergen-specific T cells recognize skin-penetrated allergen, presented in the context of MHC class-I and/or II molecules, they start to secrete large amounts of various inflammatory cytokines belonging to either Th1, Th2 of TH17 cytokines.

These mediators cause the inflammatory response of ACD reactions. In case of the involvement of cytotoxic $CD8^+$ T cells, keratinocytes are the main target cells of Fas-Fas ligand-driven apoptosis by the release of lytic enzymes (perforins and granzymes) from granules in cytotoxic $CD8^+$ T cells. Because of the necessary formation of an inflammatory infiltrate and the production of inflammatory mediators, the reaction shows a delayed-type reaction classically peaking at 48–72 h. Although, ACD is a highly allergen-specific process, it is important to note that only up to 10% of the infiltrated T cells are allergen specific. These relatively few inflammatory cells activate the vast majority of the cellular infiltrate to contribute to the clinical inflammation as seen in ACD.

For declining the inflammatory reaction, different types of regulatory mechanisms are involved. Secretion of regulatory/immunosuppressive mediators (e.g., TGF-β, PGE_2, and IL-10) from keratinocytes, fibroblasts, and macrophages suppresses the inflammatory reaction. Also metabolic degradation and transportation of allergen from skin sites can contribute to a declining immune reaction. Among others, regulatory T-cells of the Th3, Tr1, or Treg phenotype appear to be involved in suppressing the inflammatory processes.

2.1.2 Clinical Signs and Symptoms

The clinical picture of ACD is eczematous in almost all cases. It can vary depending on its location and duration. In most instances, acute eruptions (Fig. 2.1) are characterized by erythema and papules, vesicles (often coalescent), or bullae, depending on the intensity of the allergic response. In severe cases, this can lead to abundant oozing. In case of acute ACD occurring in certain areas of the body, such as the eyelids, penis, and scrotum, erythema and edema usually predominate rather than vesiculation.

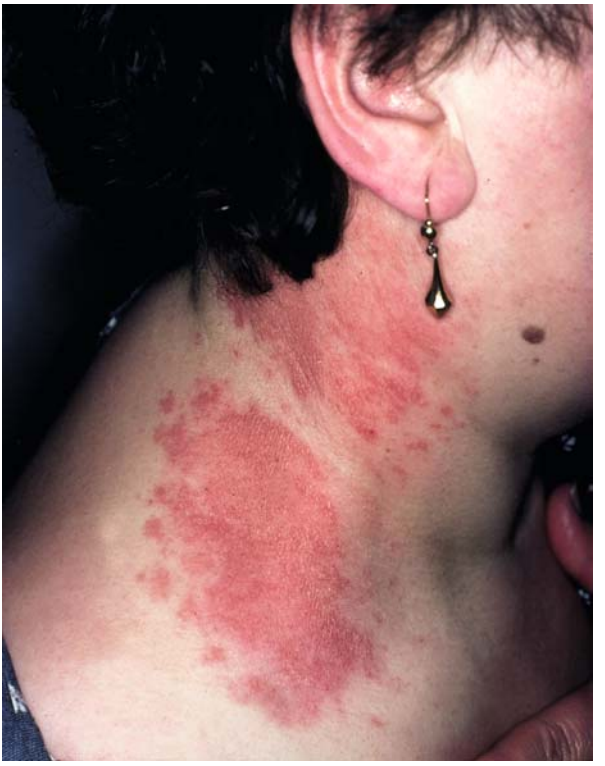

Fig. 2.1 Allergic contact dermatitis (ACD) to paraphenylenediamine from a permanent hair dye

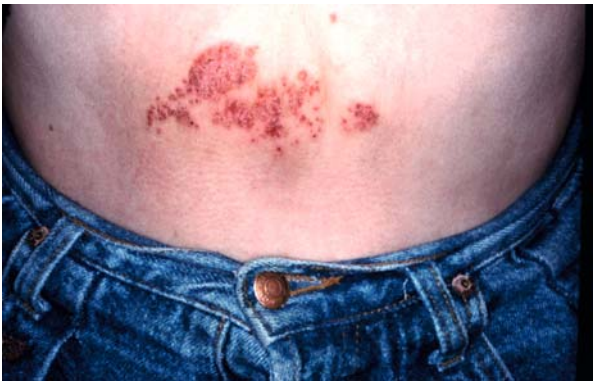

Fig. 2.2 Allergic contact dermatitis to a jean stud, extending far beyond the friction area. The nickel sulphate patch test was positive

In contrast, chronic ACD of nearly all cutaneous sites presents as a lichenified scaling, occasionally fissured dermatitis, with or without accompanying vesiculation [1]. The limits of the eczematous plaques, either vesicular (Fig. 2.2) or dry and scaly (Fig. 2.3), are usually ill-defined, extending beyond the site of application of the allergen(s) (Fig. 2.2). This is in contrast with the lesions of irritant dermatitis, which are usually sharply demarcated (see Sect. 2.3). Allergic contact stomatitis or vulvitis is diffusely erythematous, sometimes edematous, without vesiculation.

Itching is generally severe, but it can be mild in some cases.

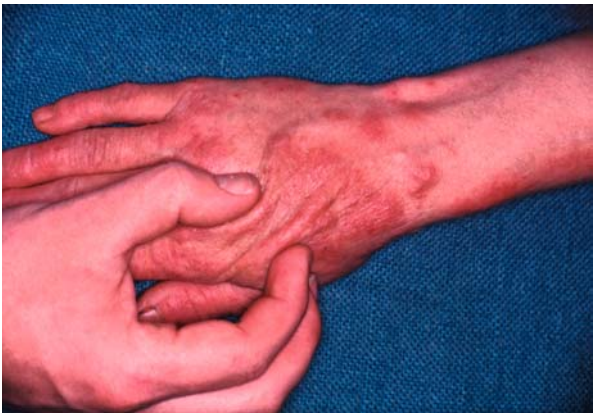

Fig. 2.3 Acute erythemato-vesicular and edematous allergic contact dermatitis to rubber gloves on the dorsa of the hands and fingers. The thiuram-mix patch test was strongly positive

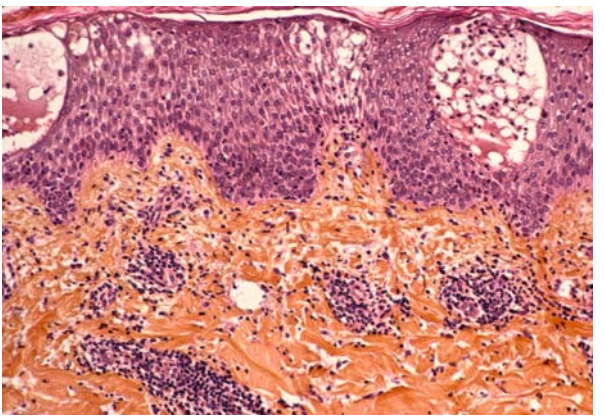

Fig. 2.4 Allergic positive patch test reaction to balsams of Peru (*Myroxylon pereirae*) at 48 h: spongiotic ve-siculation in epidermis with exocytosis of lymphocytes; in dermis, dense infiltrate of mononuclear cells around blood capillaries

2.1.3 Histopathological Features

The histopathological picture of ACD (Fig. 2.4) is a typical example of a spongiotic dermatitis. Features are very similar in all cases.

2.1.3.1 Epidermal Lesions

In the epidermis, spongiosis is an almost constant sign, resulting from the accumulation of fluid around the individual keratinocytes (exoserosis) and the consequent stretching of intercellular desmosome complexes (or "prickles").

Spongiosis is focally or evenly distributed along the length of the epidermis; it is either limited to the lower layers or extends from the basal to the granular layer. In some but not all cases, it spares the cells of the sweat duct unit. Hair follicles are usually involved in the spongiotic process.

A more plentiful accumulation of fluid results in the rupture of the intercellular prickles and in the formation of vesicles. Thus, in ACD, spongiotic vesiculation can be defined as an intra-epidermal cavity with ragged walls and surrounding spongiosis. There is migration of inflammatory cells into the epidermis (exocytosis). These cells, mainly lymphocytes and occasionally polymorphonuclear neutrophils and eosinophils, accumulate in the spongiotic vesicles.

Some vesicles are rounded and tense; they are located in the stratum spinosum, whereas others are flat and located in the stratum corneum. They finally rupture at the surface of the epidermis and vertical channels of fluid discharge are occasionally seen on the serial sections. These channels are sometimes colorfully described as "Devergie's eczematous wells" [2].

2.1.3.2 Dermal Changes

Papillary blood capillaries are often congested and dilated; dilatation of lymphatic vessels is very conspicuous in some but not all cases. Dermal edema is prominent. A dense mononuclear cell infiltrate is usually present around blood vessels of the lower dermis, and even in the subcutaneous tissue. The cells of the infiltrate migrate from the perivascular spaces to the epidermis and are found throughout the dermal tissue, either isolated or grouped in small clumps.

It is common to see a dermal infiltration of inflammatory cells around and within hair sheaths and sebaceous ducts, which show some degree of spongiosis and cellular degeneration. This picture could be partly due to direct penetration of the allergens through the pilosebaceous unit.

The infiltrate is of the lymphohistiocytic type, composed almost exclusively of mononuclear cells, varying in form and size. The occurrence of an intimate contact between the cell surfaces of lymphocytes and the cell processes of macrophages was demonstrated many years ago at the ultrastructural level. It was emphasized that, in delayed hypersensitivity, macrophages were thought to play an important role, together with lymphocytes. This view was later confirmed and broadened by the discovery of the role played by Langerhans cells.

Polymorphonuclear neutrophils are usually absent. Some eosinophils can be found in the edematous tissue of the upper dermis, migrating towards the epidermis [2].

2.2 The Allergic Contact Dermatitis Syndrome

We have developed the concept of the allergic contact dermatitis syndrome (ACDS) [3]. A syndrome can be defined as a group of signs and symptoms that actively indicate or characterize a disease [4].

A similar approach was made previously regarding irritation, i.e., the irritant contact dermatitis syndrome [5], and contact urticaria, i.e., the contact urticaria syndrome [6]. The concept of ACDS considers the various facets of contact allergy, including morphological aspects and staging by symptomatology.

The three stages of ACDS can be defined as follows:

1. *Stage 1*. The skin symptoms are limited to the site(s) of application of contact allergen(s);
2. *Stage 2*. There is a regional dissemination of symptoms (via lymphatic vessels), extending from the site of application of allergen(s);
3. *Stage 3*. Corresponds to the haematogenous dissemination of either ACD at a distance (stage 3A) or systemic reactivation of ACD (stage 3B).

Remember that patch testing is the mainstay of etiological diagnosis for all stages of ACDS.

The concept and stages of ACDS are summarized in Fig. 2.5.

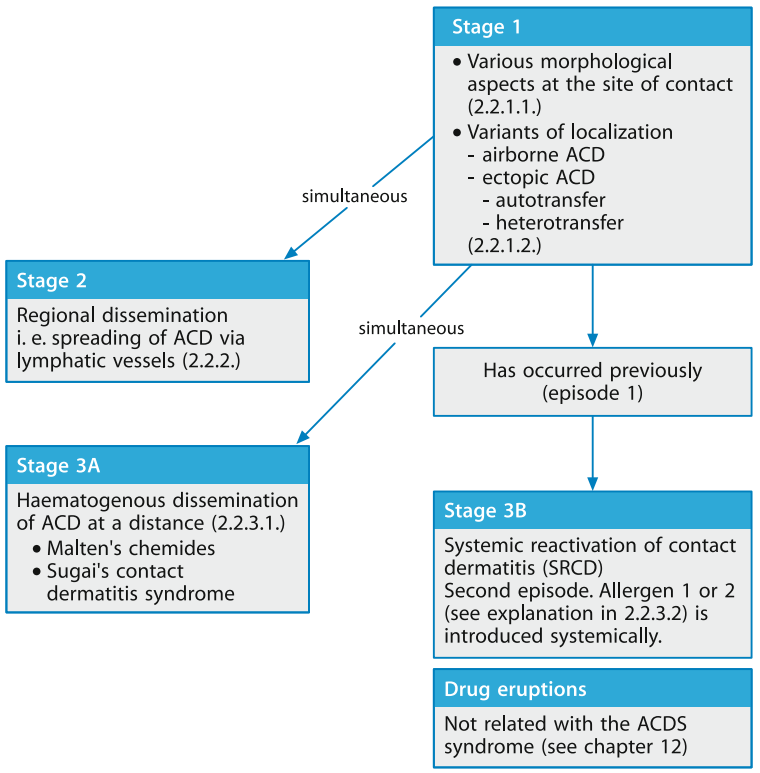

Fig. 2.5 The allergic contact dermatitis syndrome (ACDS): staging by symptomatology

2.2.1 Stage 1 of ACDS

By definition, stage 1 of ACDS includes all clinical aspects of ACD at the site (s) of application of contact allergen(s), in terms of morphological aspects and/or localizations.

2.2.1.1 Morphological Aspects

Morphological aspects of ACD are varied. The commonest are erythematous plaques (with or without edema) and/or erythemato-vesicular or erythemato-bullous eruptions, evolving sometimes to oozing dermatitis. In a chronic stage, clinical signs of ACD are those of an erythematous, dry and scaly dermatitis (see Sect. 2.12).

Clinical variants of ACD are infrequently observed. They are manifold, and can be described as follows:

1. *Purpuric ACD*. This variant is mainly observed on the lower legs (Fig. 2.6) and/or feet, and has been reported with a variety of allergens (i.e., anti-inflammatory non-steroidal topical drugs, textile dyes, etc.). Purpuric lesions are prominent or associated with eczematous symptoms (sometimes bullous on the lower part of legs and/or feet).

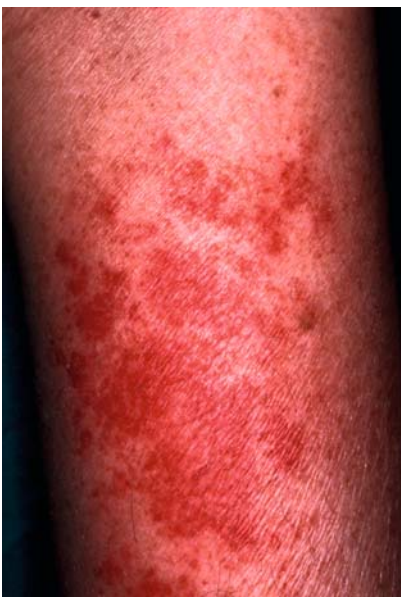

Fig. 2.6 Allergic contact dermatitis to a rubber boot. The lesions are distinctive in being not simply erythemato-vesicular but also markedly purpuric, as is frequent on the lower limbs. The mercapto-mix and mercaptobenzothiazole patch tests were positive

They may occur in other regions of the body. Purpura is the clinical manifestation of the extravasation of erythrocytes into dermal tissue and epidermis.

2. *Lichenoid ACD*. Lichenoid ACD is rare (Fig. 2.7a, b). Its clinical features mimic lichen planus (e.g., from metallic dyes in tattoos or from corals). Oral lichenoid ACD looks like oral lichen planus (e.g., from dental amalgams).
3. *Pigmented ACD*. It is mainly reported in Oriental populations; it is fully described in Sect. 3.15.
4. *Lymphomatoid ACD*. This variant cannot be defined as a clinical distinctive entity; it is based only on histopathological criteria. Clinical signs (non-diagnostic) are erythemato-edematous plaques, sometimes very infiltrated, at the site(s) of application of contact allergen(s). Histopathological examination reveals the presence of an important dermal (and sometimes subdermal) infiltrate, displaying features of pseudolymphoma, i.e., mainly lymphohistiocytic with a few neutrophils and/or eosinophils.

Immunopathological investigation permits the exclusion of malignant lymphocytic proliferation.

In all these variants of ACD, patch testing is equally useful; the clinical signs of positive patch test reactions are eczematous in nature, and therefore identical to those observed in "classic" ACD.

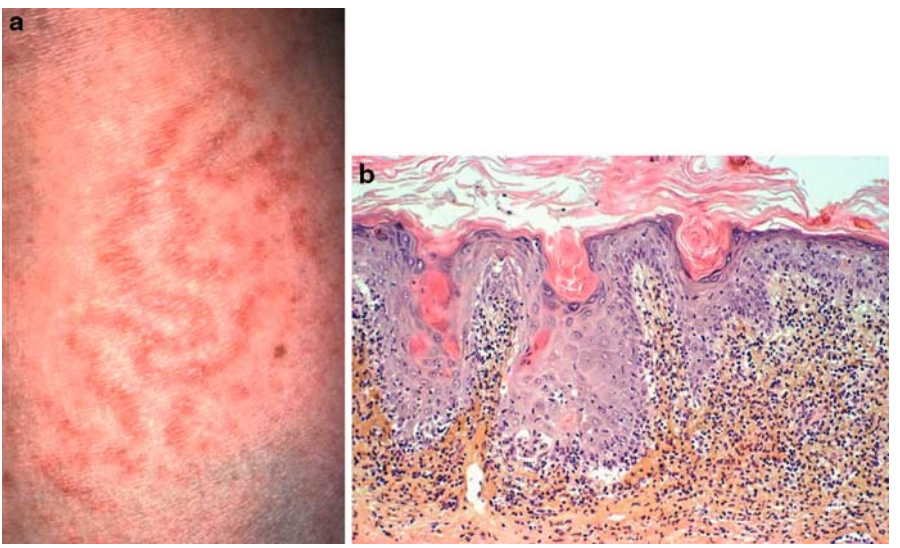

Fig. 2.7 Lichenoid allergic contact dermatitis to a red coral, 10 days after scaba diving (**a**). The histopathological picture is typical: vacuolar alteration of basal keratinocytes; cytoid bodies (apoptotic keratinocytes) in the stratum spinosum; lichenoid lymphocytic dermal infiltrate (**b**)

2.2.1.2 Topographical Variants

ACD can display some topographical peculiarities, that may be misleading for every trained dermatologist. This mainly refers to cases of "ectopic" ACD and airborne ACD.

Ectopic dermatitis can follow these:

1. *Autotransfer*. A typical example is nail lacquer ACD, located on the eyelids or lateral aspects of the neck (transfer of contact allergen by fingers).
2. *Heterotransfer*. The often-quoted example is transfer of the allergen(s) to the partner. Such events have been described as connubial ACD, or consort ACD, or ACD *per procurationem*; note that in these circumstances, the patient applying the allergen is usually free of any symptoms.

Another pitfall for clinicians is airborne ACD. Allergen(s) is (are) transported by air as dust particles, vapors or gasses. In most cases, ACD involves the face, neck and/or décolleté (Fig. 2.8a, b). There is usually no spared area, contrary to phototoxic and/or photoallergic contact dermatitis (see Sect. 5.3). Limits of eczematous lesions are ill-defined. There is no definite clue to make a clinical distinction between irritant and allergic airborne contact dermatitis. Patch testing is therefore of utmost diagnostic value. The occurrence of airborne ACD and airborne ICD is underestimated, because reports omit the term "airborne" in relation to dust or volatile irritants and/or allergens. An updated list of references is available [7].

2.2.2 Stage 2 of ACDS

Stage 2 of ACDS is linked with the regional dissemination via lymphatic vessels of ACD from the primary site of application of the allergen(s). In most cases, ACD lesions are more pronounced at the site(s) of application of the allergen(s), and disseminating lesions fade progressively from the primary site. They appear as erythematous or erythemato-vesicular plaques with poorly defined margins. In some other cases, extending lesions are more pronounced than those located at the primary site. This paradoxical observation is not fully understood. It sometimes occurs with, e.g., non-steroidal anti-inflammatory drugs or antibiotics.

Three clinical variants of regional dissemination involve more intricate immunological mechanisms. These include the following:

a. True *erythema multiforme lesions*, displaying both clinical and histopathological signs of erythema multiforme. Such reactions have been reported with several allergens [8]. The most frequently quoted are woods and plants (*Dalbergia nigra*, pao ferro, *primula obconica*, etc.); metals (nickel, cobalt); paraphenylenediamine, epoxy resin.
b. *Erythema multiforme-like lesions* presenting clinically as "targeted" lesions typical of erythema multiforme (Fig. 2.9), but histopathological signs of a spongiotic dermatitis, characteristic for eczematous dermatitis [8].

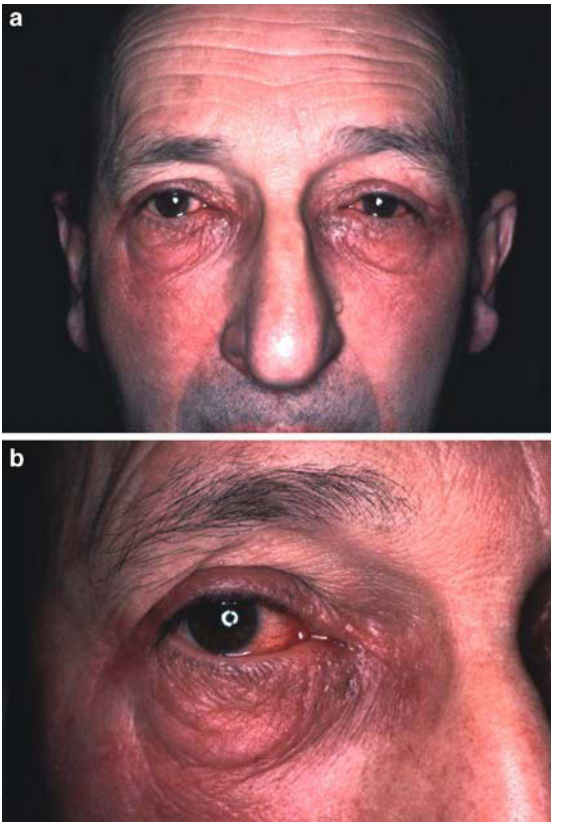

Fig. 2.8 **a**, **b** Allergic airborne contact dermatitis to *Frullania dilatata,* affecting mainly eyelids and cheeks. *Frullania* is a liverwort that grows on tree trunks (oak, beech, etc.) and rocks. The allergen is (+) frullanolide, a sesquiterpene lactone. The sesquiterpene lactone mix patch test was positive

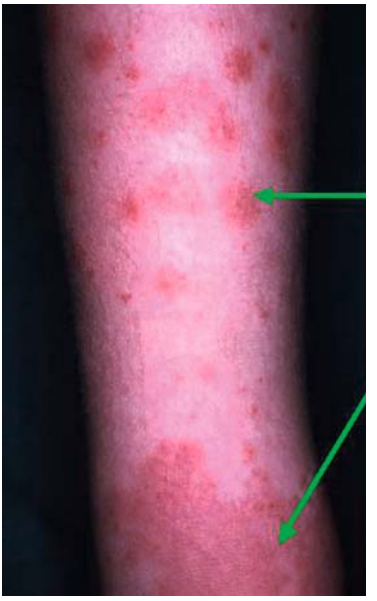

Fig. 2.9 Stage 2 of ACDS. ACD of the foot due to neomycin in a cream. Secondary *targeted* erythema multiforme-like lesions (ides) on the leg (see explanations in text)

c. The two syndromes (a) and (b) are well documented in some publications, whereas in some others there is no clear-cut distinction between both groups, due a lack of histopathological investigations.
d. An additional variant has been described by Goh [8] under the name of "*urticarial papular and plaque eruption,*" a term that is self-explanatory.

In the meantime widespread of secondary lesions can occur simultaneously at a distance of the primary site (stage 3A). In all these variants, patch testing is of diagnostic value; the clinical signs of positive patch test reactions are similar to those observed in "classic" ACD.

2.2.3 Stage 3 of ACDS

Stage 3 of ACDS includes two distinct entities, leading sometimes to unexpected confusion in the current literature. A clear-cut distinction between both entities is fully described below.

2.2.3.1 Stage 3A of ACDS

Stage 3A of ACDS can be defined as a generalized dissemination of skin lesions – via blood vessels – from the primary site of application of the allergen. It is considered that the allergen penetrates through normal and/or lesional skin and reaches distant skin sites (haematogenous dissemination) where it provokes secondary (or "ide") reactions. These reactions appear as symmetrical erythematous, sometimes slightly elevated plaques, more rarely vesicular or squamous (Fig. 2.3). They are of "pompholyx-type" on palmar and/or plantar skin.

Malten [9] coined the term "chemides" to describe the various skin manifestations at distant sites. Chemides are always concomitant with ACD lesions at the primary site(s) of application of the allergen.

Malten's historical description was rediscovered by Sugai, under the name of "contact dermatitis syndrome" [10]. Sugai makes a clear distinction between "systemic contact dermatitis syndrome" and "systemic contact-type dermatitis" (see Sect. 2.2.3, stage 3B of ACDS). The sensitization processes and pathways of these two conditions are different: contact dermatitis syndrome (syn: chemides) is provoked by percutaneous absorption of the causative allergen(s) from the primary site of application whereas, in systemic contact-type dermatitis, allergen(s) are introduced by systemic administration (ingestion, inhalation or injection). The consequence of the latter can be defined as a haematogenous contact-type dermatitis (see Sect. 2.2.3, stage 3B of ACDS).

Sugai added to Malten's initial description some clinical variants, such as true erythema multiforme lesions (Figs. 2.10 and 2.11), erythema multiforme-like lesions and/or Goh's "urticarial papular and plaque eruption", all types of lesions being similar to those reported in stage 2 of ACDS [11, 12].

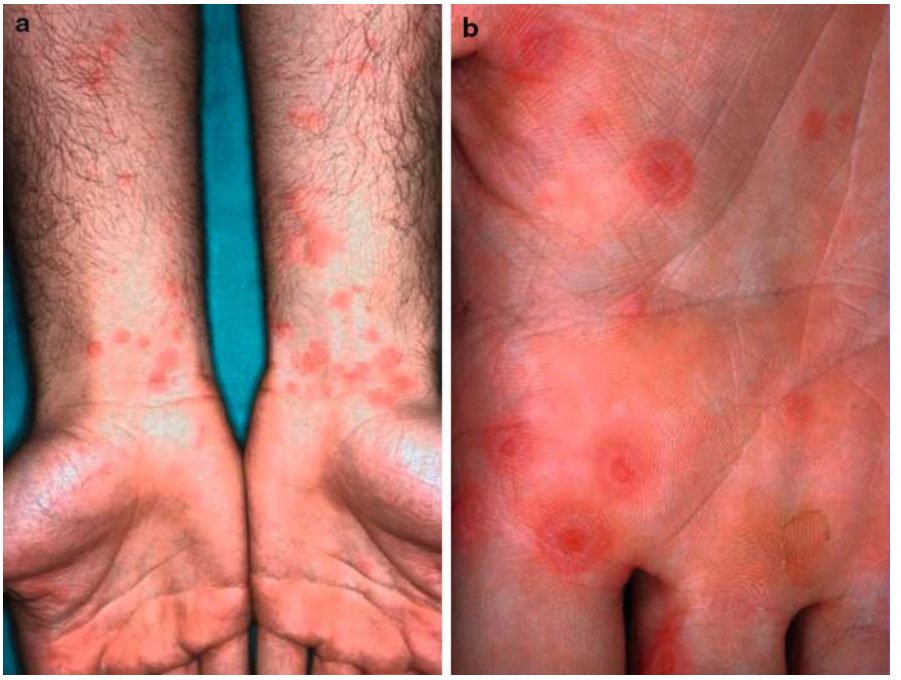

Fig. 2.10 Stage 3A of ACDS. True erythema multiforme symmetrical lesions at distant sites (hematogenous dissemination) from the primary site of sensitization (ides). (**a**) Case 1: contact allergy to dalbergiones. (**b**) Case 2: contact allergy to paraphenylenediamine

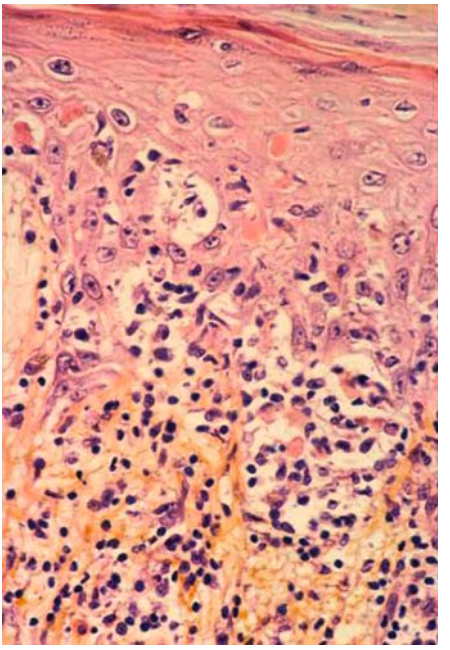

Fig. 2.11 Stage 3A of ACDS. Histopathology of a true erythema multiforme lesion (ide) displaying typical features. Apoptotic keratinocytes (cytoid or Civatte's bodies) at all epidermal levels; subepidermal initial bulla and dense lymphocytic infiltrate invading the epidermis

Stages 2 and 3A of ACDS can be present simultaneously in the same individual. The concomitant occurrence of both stages of lesions illustrates the clinical complexity of ACDS.

In stage 3A of ACDS, patch testing remains the milestone of investigation, providing accurate positive reactions, similar to those obtained in stage 2 of ACDS.

Among contact allergens involved in stage 3A of ACDS and reported in the literature, some deserve special interest: paraphenylenediamine, cobalt, nickel, mercury, mercuric chloride, corticosteroids and non-steroidal anti-inflammatory agents.

2.2.3.2 Stage 3B of ACDS

Stage 3B of ACDS has been described as follows:

1. *Baboon syndrome* [13]. This term is not satisfactory, since it tends erroneously to circumscribe symptoms to limited skin areas, i.e., buttocks, groin, perineal region; therefore it does not take into account other skin sites, which are involved as well.
2. *Fisher's systemic contact dermatitis*. The term is widely used in dermatology [14]. Nevertheless, it is a misnomer, due to the lack of precise meaning in the definition itself.

In essence, the most appropriate expression could be *systemic reactivation of allergic contact dermatitis* (SRCD) [3]. It considers the chain of events resulting in the occurrence of stage 3B of ACDS.

The successive steps are the following:

1. *First episode:* A first event of ACD to a well-defined contact allergen (allergen 1) has occurred in the past (weeks or even years before episode 2). All clinical symptoms have vanished completely, when contact with allergen 1 has ceased. Sometimes, patients have forgotten about it; this emphasizes the need for a complete clinical history (a general rule in the field of contact allergy).
2. *Second episode*: In some cases, the substance (molecule 1) is introduced systemically (ingestion, inhalation, injection) and its use is followed by a more or less generalized skin rash, usually in a symmetrical pattern (as in stage 3A of ACDS). The molecule is the true allergen (allergen 1). In other cases, another substance (molecule 2) is used systemically and provokes SRCD. This could be related with two different mechanisms:

    a. Molecule 2 is chemically closely related to molecule 1. Both are allergenic and there is cross-sensitization (see Sect. 3.13.1). Molecule 2 is therefore considered allergen 2.
    b. Another possibility is that molecules 1 and 2 are not allergenic as such, but are both transformed into another common molecule, which is the allergen (responsible for episodes 1 and 2).

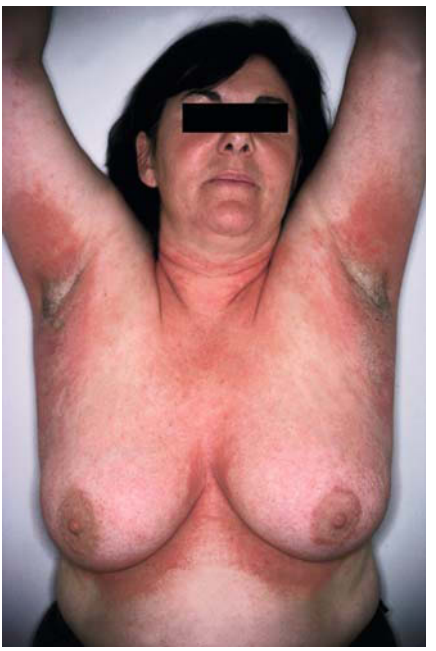

Fig. 2.12 Stage 3B of ACDS. Systemic reactivation of allergic contact dermatitis provoked by a drug containing aminophylline (theophylline + ethylenediamine) in a patient previously sensitized to ethylenediamine by skin contact

The clinical signs observed in stage 3B of ACDS share a similar pattern with skin lesions observed in stage 3A of ACDS (Fig. 2.12). The only difference is that in stage 3B, no current skin contact does occur (episode 2).

SRCD is a good indication for patch testing. Positive patch test reactions are diagnostic [15].

There is clear-cut frontier between stage 3B of ACDS (SRCD) and other immunologically related drug eruptions. In the latter, the allergens have never been applied previously onto the skin; no anterior process of skin sensitization has occurred (absence of episode 1). Among such drug eruptions, Hausermann et al. [16] and Arnold et al. [17] have coined the term SDRIFE (Symmetrical Drug Related Intertriginous and Flexural Exanthema), which differs from SRCD (or baboon syndrome).

SDRIFE specifically refers to a distinctive clinical pattern of drug eruption, and the following diagnostic criteria are proposed: (1) exposure to systemically administered drug either at the first or repeated dose (excluding contact allergens); (2) sharply demarcated erythema of the gluteal/perianal area and/or V-shaped erythema of the inguinal/perigenital area; (3) involvement of at least one other intertriginous/flexural localization; (4) symmetry of affected areas, and (5) absence of systemic symptoms and signs. Patch testing in drug eruptions is discussed at length in Chap. 12.

2

2.3 Allergic Contact Dermatitis vs. Irritant Contact Dermatitis: Criteria for Differential Diagnosis

Differential diagnosis between ACD and irritant contact dermatitis (ICD) is a major clinical problem. There are some trails to guide the dermatologist, but there is no definite "clue," as both conditions partly share similar signs and symptoms. Table 2.1 summarizes some clinical differences between ACD and ICD [18]. Histopathological examination has no real interest. Therefore, patch testing and other tests (see later) are of prime importance. When patch tests are positive, it is still possible that the clinical condition is mixed, i.e., associating symptoms of ACD and ICD.

Table 2.1 Clinical differences between ICD and ACD

	ICD	ACD
Clinical course	Acute ICD may appear after first exposure (at least with strong irritants).	Sensitizing exposure(s) is required. Clinical lesions appear after subsequent challenges with re-presentation of the antigen to already primed (memory) T-cells.
	In acute ICD lesions appear rapidly, usually minutes to few hours after exposure, but delayed reactions can be seen.	Lesions usually appear 24–72 h after the last exposure to the causative agent, but they may develop as early as 5 h or as late as 7 days after exposure.
	Irritant reactions are characterized by the "decrescendo phenomenon." The reaction reaches its peak quickly, and then starts to heal.	Allergic reactions are characterized by the "crescendo phenomenon" and the kinetics of resolution may be slower.
Morphology	Acute ICD includes erythema and edema and sometimes vesicles or bullae, oozing and pustules. Necrosis and ulceration may also be seen with corrosive materials.	Pustules, necrosis, or ulceration are rarely seen.
	Subacute or chronic ICD is characterized by hyperkeratosis, fissuring, glazed, or scalded appearance of the skin.	Intense vesiculation increases the suspicion of ACD, but it may not be present in chronic ACD.
	Lesions are characteristically sharply circumscribed to the contact area (Fig. 2.13). Usually there is absence of distant lesions, but sometimes dermatitis may be generalized depending on the nature of the exposure.	Clinical lesions are stronger in the contact area but their limits are usually ill-defined. Dissemination of the dermatitis with distant lesions may occur.
Symptoms	Symptoms of acute ICD are burning, stinging, pain, and soreness of the skin (pruritus may be present).	Pruritus is the main symptom of ACD.

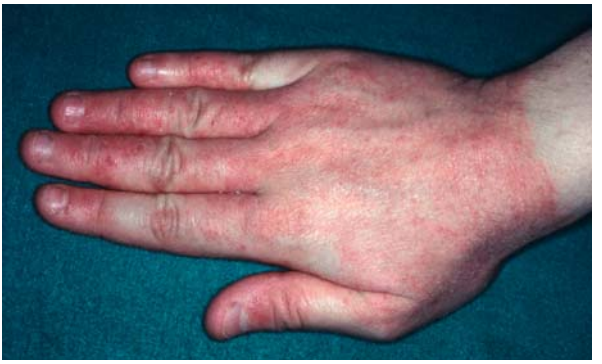

Fig. 2.13 Irritant contact dermatitis. Pruritic, discretely painful, sharply demarcated plaque of the dorsum of the hand due to repeated contact with household detergents

2.4 Other Skin Diseases in Which Patch Testing is of Major Interest

Patch testing is also highly recommended in patients suffering from various eczematous conditions, considered (partly or entirely) endogenous. The philosophy behind this strategy is related to the fact that in many cases ACD may worsen underlying dermatitis.

Thus, the purpose of patch testing is clearly defined: its results permit further avoidance of contact allergens in the management of eczematous conditions. A list of eczematous (endogenous) diseases is presented in Table 2.2.

In other words, the practising dermatologist is confronted with the problem of various types of eczematous eruptions, which are attenuated by the use of topical corticosteroids, but are relapsing when tapering is recommended.

Histopathological investigation is not contributory in those cases: there are almost no epidermal changes and dermal lesions are limited to a perivascular nonspecific lymphocytic infiltrate.

Hence, superimproved ACD to topical corticosteroids has to be kept in mind. This approach concerns also the use of other topical drugs, such as tacrolimus, pimecrolimus, vitamin D_3 analogues, antibiotics, etc. The allergens may be the active molecule itself or one of the components of the vehicle.

Table 2.2 Eczematous (endogenous) diseases in which patch testing is recommended

Atopic dermatitis
Nummular dermatitis (nummular eczema)
Seborrhoeic dermatitis (when presenting episodes of acute inflammation)
Asteatotic eczema
Stasis dermatitis
Eczematous lesions around leg ulcers
Pompholyx and/or dyshidrotic eczema (see Sect. 2.5)
Lichenification
“Eczematous psoriasis” (palms and soles)

Accurate patch testing needs to be performed not only with standard allergens, but also with topical corticosteroids and preservatives, and of course more precisely concerned allergens in each individual case.

2.5 An Algorithmic Approach: The Key Role of Patch Testing

Each patient, presenting (or having) presented clinical signs suggestive of ACD, requires a complete investigation, built on grounds of evidence-based dermatology. An algorithmic approach of problems is an efficient way to reach a good evaluation in terms of diagnosis and management ("holistic approach"). The procedure is extremely useful, in particular when dealing with hand dermatitis, a daily challenge for dermatologists. In this perspective, patch testing is one of the pieces of the jigsaw puzzle (see Fig. 2.5). A similar approach can be applied to other situations.

2.6 Hand Dermatitis: Procedures Applied in Differential Diagnosis

Hand dermatitis is a difficult problem, the management of which requires skill and expertise [19]. Positive and differential diagnosis is crucial. Hand dermatitis may be multifactorial, so that more than one diagnosis has to be kept in mind. The systematic use of an algorithmic approach, including targeted patch testing, is very informative.

2.6.1 Hand Dermatitis: Exogenous and Endogenous Factors

The occurrence of hand dermatitis in a patient may imply exogenous and/or endogenous factors. In each case the balance between these two factors needs precise evaluation (Fig. 2.14) as stressed many years ago by Fregert [20].

2.6.2 A Classification of Hand Dermatitis

The following classification of hand dermatitis is proposed, taking into account the occurrence of exogenous and/or endogenous factors (Table 2.3) [21]. It is obvious that several other dermatoses can affect hands. This classification is willingly limited to the most common situations, being either eczematous, or involving differential diagnosis with eczema. Some skin diseases deserve a precise definition.

Tinea manuum. Tinea manuum is synonymous with fungal infection of the hands by dermatophytes. The clinical picture on the back of the hands is similar to that observed on other

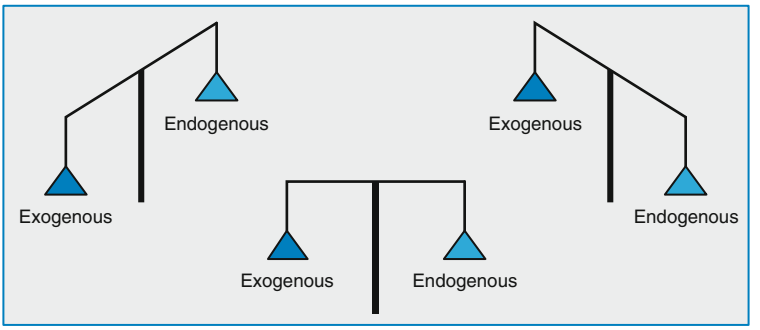

Fig. 2.14 The evaluation of exogenous and endogenous factors in hand dermatitis [21]

Table 2.3 Proposal for a classification of hand dermatitis [from [21])[a]

A. Exogenous
Irritant contact dermatitis (ICD): frictional, chemical (Fig. 2.13)[a]
[a]Allergic contact dermatitis[a]
Protein contact dermatitis (see Sect. 10.2) and contact urticaria (see Sect. 10.1)[a]
(*Tinea manuum*) (Figs. 2.15 and 2.16)[a]
B. Endogenous
• 3 Atopic dermatitis (see Chap. 9)
• 4 Nummular dermatitis (nummular eczema)
• 5 Pompholyx and/or dyshidrotic eczema (Figs. 2.17 and 2.18)
• 6 Hyperkeratotic palmar dermatitis (Figs. 2.19 and 2.20)
• 7 Psoriasis and "eczematous psoriasis"
• 8 Fingertip dermatitis (Fig. 2.21)

[a]In some cases, hand dermatitis is the result of the occurrence of two (or more) combined conditions, e.g., irritant and allergic contact dermatitis, nummular dermatitis, ICD, etc. Atopic dermatitis can involve both exogenous and/or endogenous factors. Some authors prefer the term "ICD with an atopic background"; this is misleading, since not only irritants but also contact allergens and proteins can penetrate into the skin and be responsible for clinical manifestations

parts of the body, i.e., round-shaped erythematosquamous lesions, with an elevated margin, either scaly or vesicular. In contrast, *tinea manuum* of the palms is a whitish often unilateral (Fig. 2.15) scaly dermatosis without any inflammatory component. Skin creases appear as white prominent crossing lines (Fig. 2.16). Erythema is generally absent. Abundant floury material is peeled off easily by curettage. Microscopic investigation is diagnostic.

Nummular dermatitis. Nummular dermatitis (nummular eczema) is a variety of eczema of unknown origin. It is claimed that an atopic background does exist in certain cases. Eczematous lesions are round or oval-shaped, either vesicular and oozing, or dry and scaly. The localization on the palms is sometimes described as "apron dermatitis".

Pompholyx. Pompholyx is defined as a clinical variant of eczematous lesions, involving exclusively palmar skin and/or lateral aspects of the fingers (Fig. 2.17). Pompholyx is

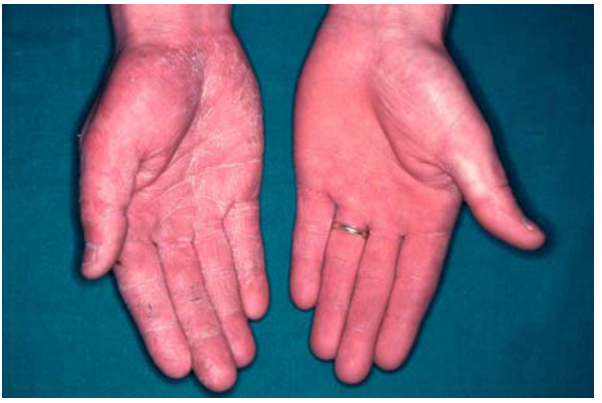

Fig. 2.15 *Tinea manuum*. It is a diagnostic trap with chronic palmar eczema. In most cases, it is strictly unilateral, which provides a first clue to the diagnosis

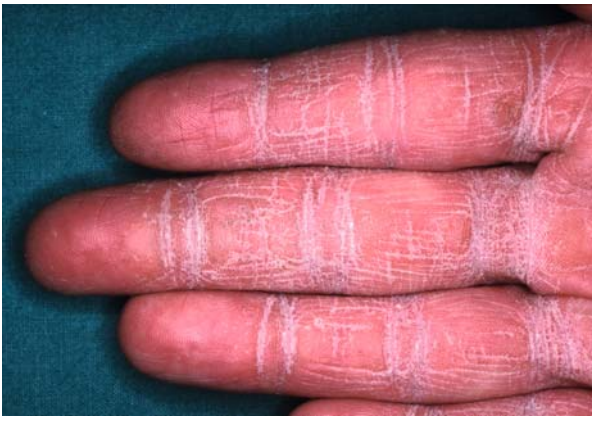

Fig. 2.16 *Tinea manuum* of the palmar aspect of the fingers. Dusty desquamation on an erythematous background with pearl white accentuation of the palmar flexor folds. The appearance is very similar to that of some cases of hyperkeratotic palmar dermatitis, but, in *tinea manuum*, scraping yields a flurry of disistegrating scales

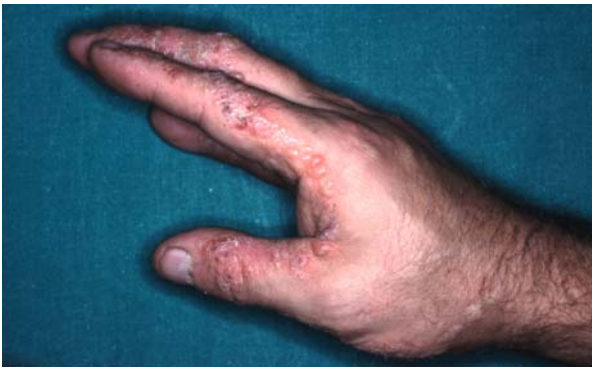

Fig. 2.17 Pompholyx. The typical vesicles are bunched on the lateral aspects of the fingers. They are hard to touch, embedded in epidermis, and translucent. They are associated with intense pruritus

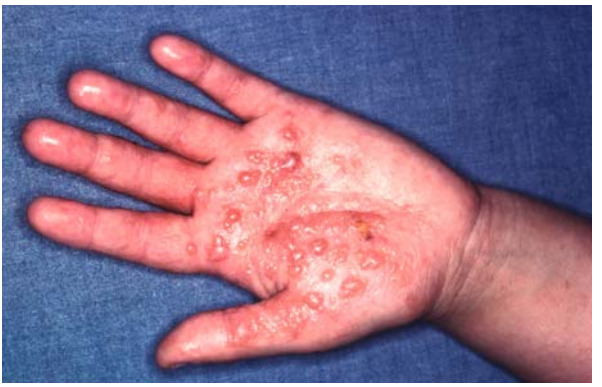

Fig. 2.18 Palmar pompholyx. Isolated and confluent vesicles with bullae are scattered over the palms

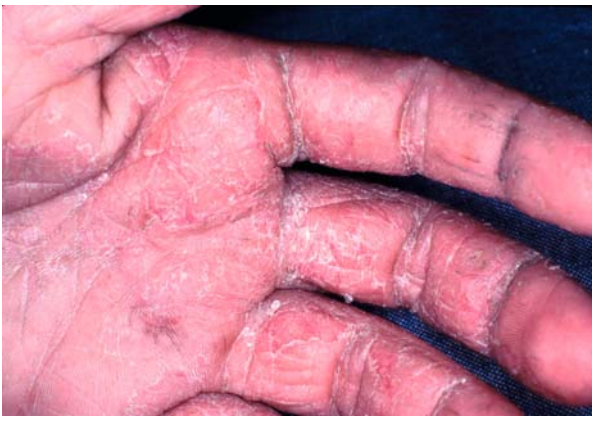

Fig. 2.19 Hyperkeratotic palmar dermatitis. Clinical presentations vary and probably encompass different entities produced by a combination of endogenous and mechanically repetitive exogenous factors. In some cases, the differential diagnosis with palmar psoriasis can be difficult

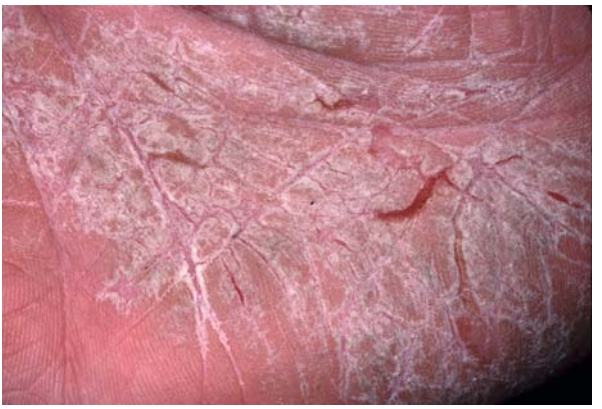

Fig. 2.20 Hyperkeratotic palmar dermatitis. Well-demarcated erythematosquamous plaques are traversed by deep fissures due to the absence of cutaneous elasticity on skin traction

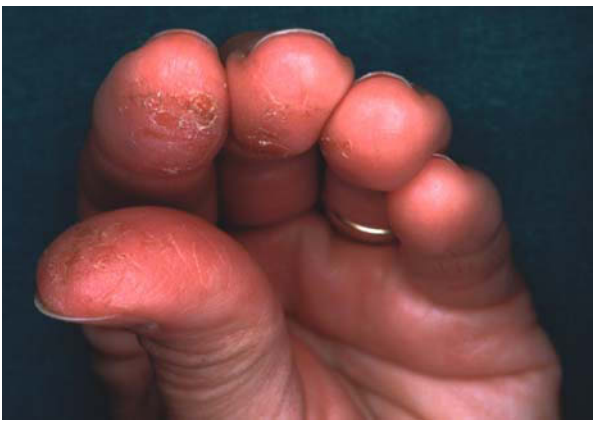

Fig. 2.21 Fingertip dermatitis. ACD to garlic in a female cook handling cloves of garlic. Positive patch test to diallylsulphide, one of the garlic allergens

synonymous with dyshidrotic eczema [19]. Clinical symptoms of dyshidrotic eczema are characterized by the occurrence of numerous vesicles or bullae, either isolated or grouped in crops that appear on normal skin of the palms or underlying erythema (Fig. 2.18). Itching is often severe. Considered in many cases endogenous (an atopic background has been advocated mainly in children), it can be triggered by environmental factors, such as tobacco smoking, wet/and or hot work conditions, and hot climate.

Research for etiological factors may be useful; indeed it has been argued that, in some cases, pompholyx reflects an "ide" reaction to ACD or mycotic infections; in some others, it could be a clinical manifestation of SRCD, in particular to drugs or food ingredients, like spices. A particular relationship between pompholyx and nickel ingestion in nickel-sensitive patients has been advocated [22], but it remains controversial. Oral challenge with nickel is sometimes positive [22].

When pompholyx evolves to a chronic stage, lesions are dry and scaly. At this erythematosquamous stage, differential diagnosis may be difficult with other eczematous conditions or psoriasis.

Hyperkeratotic palmar dermatitis. This condition is characterized by the outcome on the palms of hyperkeratotic sharply demarcated plaques (Fig. 2.19). Deep, painful, sometimes bleeding crevices are common (Fig. 2.20). Erythema is usually very pronounced with well-defined margins, extending around hyperkeratotic plaques, but, in some cases, it is totally absent. Itching, if any, is usually moderate. Mechanical factors can sometimes be implied (hyperkeratotic variant of frictional dermatitis), but, in most cases, environmental factors cannot be traced; therefore hyperkeratotic palmar dermatitis is considered endogenous. This optional view reflects our incomplete understanding of the mechanisms involved in the impaired keratinization of the stratum corneum, in relation or not with an inflammatory process.

Psoriasis. Psoriasis of the hands is common. Lesions are typical on the dorsal hands. Palmar psoriasis is often difficult to diagnose, when not associated with lesions on other

skin sites. In some cases, it cannot be differentiated from hyperkeratotic palmar dermatitis, with which it shares common features. Biopsy is of no help. Nail examination is important, since psoriatic nail lesions are diagnostic.

Fingertip dermatitis. Chapping of the fingertips is a common event. Painful crevices and bleeding do occur in severe cases. We have stressed [21] that fingertip dermatitis limited to the thumb, index (and eventually medius) of one or both hands frequently implies irritant (frictional and/or chemical) or allergenic factors. In those cases, fingertip dermatitis may be typical of (a) ICD; (b) ACD (Fig. 2.21) or (c) protein contact dermatitis. We have coined the term "gripping form" of fingertip dermatitis [21]. Such considerations are far too simple; in many of these cases the skin condition remains unclear and it is therefore considered endogenous, environmental factors playing only an adverse role. When some fingers are randomly involved, whereas others are spared, or in case of complete involvement of all fingers of both hands, etiology is even more obscure.

2.6.3 Tools of Investigation

Several procedures are available in the diagnostic approach of hand dermatitis. They are listed in Table 2.4.

2.6.4 Hand Dermatitis: Some Examples of an Algorithmic Approach

Two examples of an algorithmic approach applied to the diagnosis of hand dermatitis are presented in Figs. 2.22 and 2.23.

2.6.5 Management of Chronic Hand Dermatitis

In cases of ICD and/or ACD, the eviction of irritants and/or contact allergens clears the skin condition, provided that no endogenous factors are involved.

Table 2.4 Hand dermatitis: tools of investigation[a]

Accurate clinical history, obtained by questionnaire
Careful clinical examination
Patch testing
Prick testing
Microscopic examination of scales collected by curettage (in search of dermatophytes)
IgE blood level (of minor interest, to precise an atopic background)

[a]Skin biopsy provides questionable results in most cases

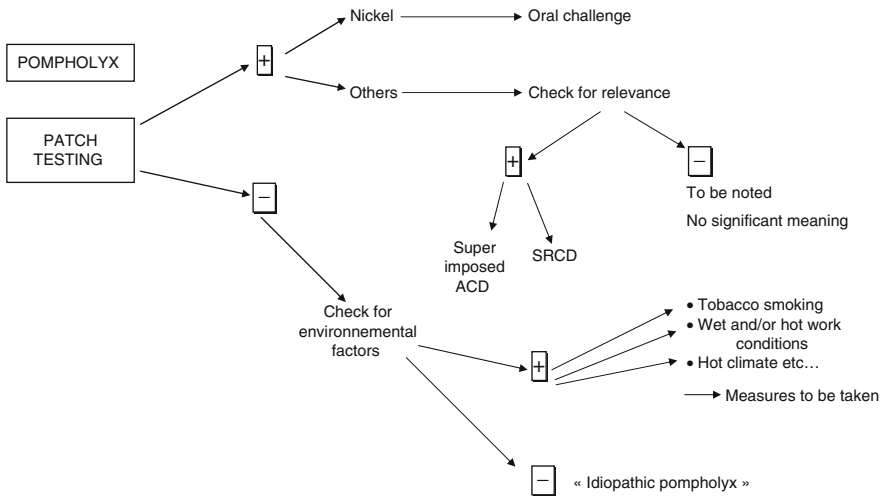

Fig. 2.22 An algorithmic approach to pompholyx

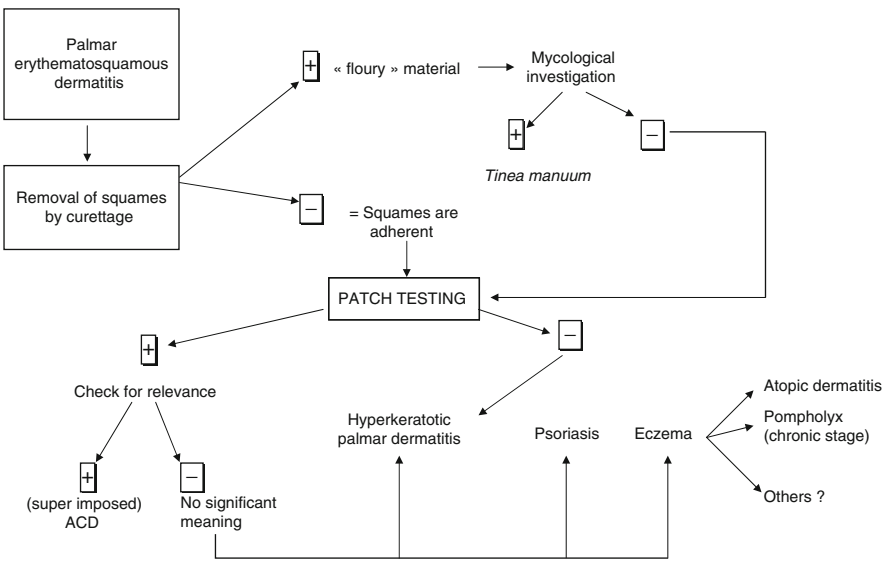

Fig. 2.23 An algorithmic approach to palmar erythematosquamous dermatitis

It is obvious that chronic hand dermatitis represents a difficult problem in terms of clinical diagnosis, even when all procedures of investigation have been carefully conducted. After exhausting all potential "clues" to encircle etiopathogenic factors involved, mystery still remains in some cases. It is sometimes claimed that each individual case is "unique." Therefore, it is not surprising that treatment options according to an evidence-base approach

(randomized controlled clinical trials, RCTs) are still lacking. It can be concluded that despite the abundance of topical and systemic treatment options, disease management in patients with severe hand dermatitis is partly unsatisfactory.

There is a strong need for RCTs of existing and new treatment options based on clearly diagnosed subtypes of hand dermatitis and its severity [23].

Despite of their limitations, topical treatments include emollients, corticosteroids, immunomodulators (tacrolimus and pimecrolimus), and UV light (many variants) adapted to each individual cases. Many systemic treatments have been proposed: ciclosporine, azathioprine, methotrexate, and retinoids [23]. A new systemic retinoid, alitretinoin, seems to offer promising results in refractory cases and is under clinical evaluation [24, 25].

References

1. Belsito DV (2003) Allergic contact dermatitis. In: Freedberg IM et al. (eds) Fitzpatrick's Dermatology in General Medicine, 6th edn. Mc Graw-Hill, New York, pp 1164–1177
2. Lachapelle JM, Marot L (2006) Histopathological and immunohistopathological features of irritant and allergic contact dermatitis. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 107–116
3. Lachapelle JM (2001) Dermato-allergologie de contact. Nouv Dermatol (Strasbourg) 20:450–452
4. Grosshans E, Lachapelle JM (2008) Signs, symptoms or syndromes? Ann Dermatol Venereol 135:257–258
5. van der Valk PGM, Maibach HI (1995) The irritant contact dermatitis syndrome, CRC Press, Boca Raton, 393 p
6. Maibach HI, Johnson HL (1975) Contact urticaria syndrome. Contact urticaria to diethyltoluamide (immediate type hypersensitivity. Arch Dermatol 111:726–730
7. Santos R, Goossens A (2007) An update on airborne contact dermatitis: 2001–2006. Contact Dermatitis 57:353–360
8. Goon A, Goh CL (2006) Noneczematous contact reactions. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 349–362
9. Malten KE, Nater JP, Van Ketel WG (1976) Patch testing guidelines. Dekker and van de Vegt, Nijmegen, 135 p
10. SugaiT (1988) Contact dermatitis syndrome and unusual skin manifestations. Skin Research 30:8–17
11. Sugai T (2000) Contact dermatitis syndrome (CDS) Environ Dermatol (Nagoya) 7:543–544
12. Kato Y, Sugiura M, Hashimoto R, Ogawa H, Hayakawa R (2001) Eight cases of contact dermatitis syndrome. Environ Dermatol 8:41–47
13. Andersen KE, Hjorth N, Menné T (1984) The baboon syndrome: systemically induced allergic contact dermatitis. Contact Dermatitis 10:97–101
14. Fisher AA (1986) Systemic contact-type dermatitis. In: Contact Dermatitis, Lea and Febiger, Philadelphia, pp 119–131
15. Veien NK, Menné T (2006) Systemic contact dermatitis. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 295–307
16. Hausermann P, Harr Th, Bircher AJ (2004) Baboon syndrome resulting from systemic drugs: is there strife between SDRIFE and allergic contact dermatitis syndrome? Contact Dermatitis 51:297–310

17. Arnold AW, Hausermann P, Bach S, Bircher AJ (2007) Recurrent flexural exanthema (SDRIFE or baboon syndrome) after administration of two iodinated radio contrast media. Dermatology 214:89–93
18. Ale SI, Maibach HI (2006) Irritant contact dermatitis versus allergic contact dermatitis. In: Chew AL, Maibach HI (eds) Irritant Dermatitis. Springer, Berlin, pp 11–18
19. Menné T, Maibach HI (2000) Hand Eczema, 2nd edn. CRC Press, Boca Raton, 431 p
20. Fregert S (1981) Manual of Contact Dermatitis, 2nd edn. Munksgaard, Copenhagen, 139 p
21. Lachapelle JM (2001) Les dermatites des mains: approche algorithmique des onze diagnostics différentiels de base. In: Progrès en Dermato-Allergologie, Bruxelles, 2001, Lachapelle JM, Tennstedt D (eds) John Libbey Eurotext, Montrouge (France) pp 1–10
22. Veien NK, Menné T (1990) Nickel contact allergy and a nickel-restricted diet. Semin Dermatol 9:197
23. Diepgen TL, Agner T, Aberer W et al (2007) Management of chronic hand eczema. Contact Dermatitis 57:203–210
24. Elsner P, Diepgen TL, Coenraads PJ et al (2008) Alitretinoin relieves signs and symptoms of severe hand eczema. Contact Dermatitis 58(Suppl 1):33
25. Diepgen TL, Ruzicka T, Gemec G et al (2008) Alitretinoin induces complete responses in steroid-refractory CHE. Contact Dermatitis 58(Suppl 1):33

Patch Testing Methodology

3

J.-M. Lachapelle and H.I. Maibach

3.1 Historical Background

Jozef Jadassohn is the father of patch testing [1]. At the time of his discovery in 1895, he was Professor of Dermatology at Breslau University (now Wroclaw in Poland). He initially reported a patient who had developed an eczematous reaction to mercury plasters. He recognized the potential for eczematous reactions to occur in some (sensitized) patients when chemicals were applied to their skin; he thereby introduced the world to the contact test, then referred to as "Funktionelle Hautprüfung" [2].

Bruno Bloch (Professor at Basel and Zurich Universities) is considered by the international community as one of the more outstanding pioneers in the field of patch testing, continuing, and expanding Jadassohn's clinical and experimental work. In some textbooks and papers, patch testing is sometimes quoted as the Jadassohn-Bloch technique.

In retrospect, it is difficult to assess the real place of the patch test procedure for the diagnosis of contact dermatitis between 1895 and the 1960s. Some points seem obvious:

- The technique was used extensively in some European clinics, and ignored in others.
- No consensus was reached concerning material, concentrations of allergens, time of reading, reading scores, etc.
- Differential diagnosis between irritant vs. allergic contact dermatitis was often unclear.

It is no exaggeration to say that patch testers were acting like skilled craftsmen. Nevertheless, they provided, step by step, new information on contact dermatitis.

During that long period, clinicians often equated a positive patch test with the fulfilment of Koch's postulate [3]. They inferred that because a patient with dermatitis was shown to develop a positive reaction to compound X, the same compound must therefore be the cause of the dermatitis. In other words, there was little attempt to interpret correctly patch test results. Relevance was a neglected concept.

Credit must be paid to the former members of the International Contact Dermatitis Research Group (ICDRG) for their invaluable contribution to the standardization and interpretation of patch test procedures. Their efforts have encouraged many dermatologists, immunologists, chemists, and pharmacists.

Patch testing is now a well-recognized diagnostic tool, constantly being refined.

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3.2 Definition and Aims

General considerations need to be pointed out about patch testing methodology.

First of all, patch testing aims to reproduce "in miniature" an eczematous reaction, by applying allergens under occlusion on intact skin of patients, suspected to be allergic. It is the in vivo visualization of the elicitation phase of a delayed-type hypersensitivity (type IV) reaction. Therefore, it is not intended to reflect an irritant reaction, considering its occurrence an untoward event, to be avoided by any means.

It is primarily aimed to detect "culprit" allergens in ACD, but its field of interest has been extended to some cutaneous systemic drug eruptions (see Chap. 12). It is submitted to general rules of evidence-based medicine applied to investigative procedures [4].

3.2.1 Requirements for an Ideal Patch Testing Procedure

Several requirements are advocated to reach an ideal patch testing procedure [5]:

- A perfect patch test should give neither false-positive nor false-negative reactions.
- It should cause as few adverse reactions as possible, particularly no patch test sensitization. False-positive, false-negative, and adverse reactions are all dose-dependent.
- Simplicity, safety, and low cost of patch testing methodology is highly recommended.
- Patch testing must have a very good positive predictive value, defined as the percentage of true cases in those with a positive test, when this test is used in a given population.
- Patch testing must also have a very good negative predictive value, defined as the percentage of disease-free individuals in those with a negative test, when this test is used in a given population.
- Positive and negative predictive values depend on several parameters, which cannot be dissociated:
    - Sensitivity defined as the probability of a positive test in an individual with the disease
    - Specificity defined as the probability of a negative test in an individual without the disease
    - The prevalence of the disease in the given population
- A good screening test has also to be reliable, which means that it has to be precise and to have good intraobserver and interobserver reproducibility.

3.2.2 Is Patch Testing the "Gold Standard" to Investigate Patients with Allergic Contact Dermatitis?

"Tests reactions properly performed and interpreted are acceptable as scientific proof of a state of allergic sensitization."

The question is: can Rietschel's statement [6] be fulfilled by patch testing? At present the answer is as follows: patch testing even with optimum concentration and

vehicle for a given allergen is, like most diagnostic tests, neither 100% sensitive nor 100% specific [5].

Despite its limitations, patch testing is by no means the cornerstone of the diagnostic procedure.

Its reliability is increased if it is sustained by additional tools of investigation, such as the following:

- Use of complementary testing approaches, that is, semi-open tests, ROATs, etc. (see Chap. 7).
- Other methods for assessment of clinical relevance of patch test reactions (see Chap. 8).

Conventional patch testing, as described in this chapter, is used worldwide. Allergens are produced and purchased separately from patch test units plus tapes.

TRUE Test is an alternative way of patch testing described in Chap. 6.

3.3 Patch Test Units

Earlier (nonchamber) patch tests, such as Leukotest, Porotest, Neo-Dermotest, Curatest, and others have been withdrawn from the market.

Retrospectively, their design was unsatisfactory, since the amount of allergens applied was not standardized and varied considerably when comparing methodologies in patch test clinics.

3.3.1 Finn Chamber

Finn Chamber is a round aluminum patch test device which provides good occlusion because of the chamber design [7]. The 8 mm inner diameter provides a 50 mm^2 area and about 20 µL volume. The outer diameter is 11 mm and the distance between the chambers is 20 mm. Finn Chambers are available mounted on an acrylate-based adhesive tape, Scanpor Alpharma AS, Norgesplaster Facility, Kristiansand, Norway.

Finn Chambers on Scanpor (Fig. 3.1) are available in strips of 10 (2 × 5) and one chamber(s). The strips of 10 chambers are practical when testing with a large number of substances, for example, with routine tests. Smaller strips are suitable for small test series and individual tests.

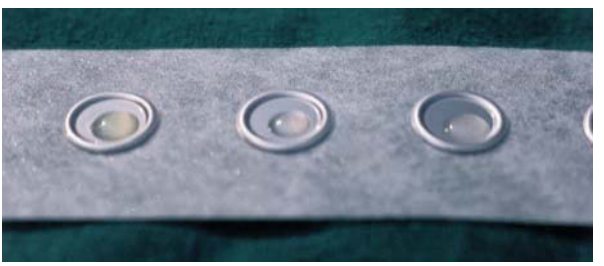

Fig. 3.1 Finn chambers filled with allergens dispersed in petrolatum

Most commercial test substances are suitable for Finn Chambers. The substances incorporated in petrolatum are applied directly into the chamber. For liquids (e.g., formaldehyde), a filter paper disc is placed in the chamber and saturated with the liquid.

Finn Chambers may be safely used for patch testing mercurials if these are dispersed in petrolatum [8], but are unsuitable for aqueous solutions of some mercurials, due to a complete corrosion of the aluminum chamber [8]. Polypropylene-coated chambers (thus avoiding corrosion) are available on request.

The Finn Chamber Tray keeps the test strips in good order when applying the test substances. The trays are stackable, which saves space on the work surface. When removing the tests, the occlusion is verified by a ring-shaped depression around each test.

For locating the test sites, a special device, Reading Plate, is recommended. Reading Plate can also be used when removing the tests (Fig. 3.2).

Apart from standard 8 mm (inner diameter) Finn Chambers®, large 12 mm (inner diameter) Finn Chambers can be purchased (200 strips of one chamber). These are of special interest when using the Atopy Patch Test (see Sect. 9.2.). Extra-large 18 mm (inner diameter) Finn Chambers are intended to be used only for special experimental purposes.

The methodology of use of Finn Chambers is as follows:

a. Lay out, with backing removed, all of the chambers to be used.
b. Start with no. 1 of the standard tray, and apply a small amount of allergen to each disk. A 5 mm ribbon of petrolatum-based allergen is sufficient. Proceed in sequence through the trays to be tested.
c. For liquid allergens, place a filter paper disk in the chamber, and apply one drop of liquid, just sufficient to soak the disk. Petrolatum patches can be made up a few hours in advance; liquid patches should be made up at the last minute.

When all patches in a Finn Chamber patch test show red infiltrated papular rings, contact sensitivity to aluminum should be suspected [9], but it can be considered exceptional.

Finn Chambers® are manufactured and distributed by SmartPractice®Finland Oy, Rannankoukku 22, 04300, Tuusula, Finland, Tel.: +358-9-2755366 Fax: +358-9-2754335, E-mail: epitest@epitest.fi

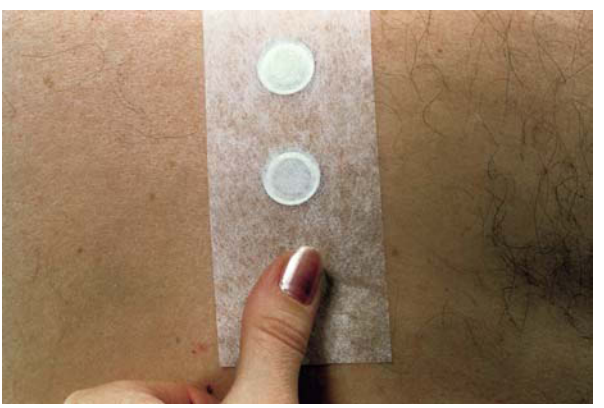

Fig. 3.2 Finn chambers: mode of application. After removal, skin sites of application can be checked by the Finn chamber reading plate

3.3.2 Plastic Square Chambers

Several companies have different models of square plastic chambers as an alternative. The square shape of the chambers is intended theoretically to differentiate allergic and irritant reactions.

3.3.2.1 IQ Square Chamber Chemotechnique

The IQ chamber Chemotechnique is made of additive-free polyethylene plastic. Undesired side effects in the form of allergic reactions to the test unit itself are avoided due to the chemical stability of the polyethylene plastic.

The IQ chambers are supplied in units of 10 square chambers (in two rows of 5 chambers per row) on a hypoallergenic acrylic-based nonwoven adhesive tape, providing good occlusion and fixation of the test unit to the skin (Fig. 3.3). The tape with the chambers is protected by a stiff plastic cover, with ten compartments that correspond to the chambers on the tape.

The volume of the chamber is 65 µL and the inside area of the chamber is 9 × 9 mm^2 (81 mm^2). The bottom of the chamber is filled with filter paper. The distance between the chambers is 12 mm in the row and 20 mm between the rows. The width of the tape is 68 mm and the length is 142 mm.

IQ Chambers are delivered in two sizes of cardboard boxes containing either 100 units or 50 units per box. A variant of the IQ chamber has been introduced more recently, named IQ Ultra patch test unit. The IQ Ultra patch test unit has important advantages.

Each chamber has a filter paper incorporated, which eliminates adding loose filter papers.

The rim of each chamber has an adhesive layer to optimize adhesion to the skin and to eliminate leakage. This makes IQ Ultra a closed-cell system enhancing occlusion and confining the test reaction within the chamber parameter.

The size of the IQ Ultra is small to allow the application of multiple test units to patients' backs.

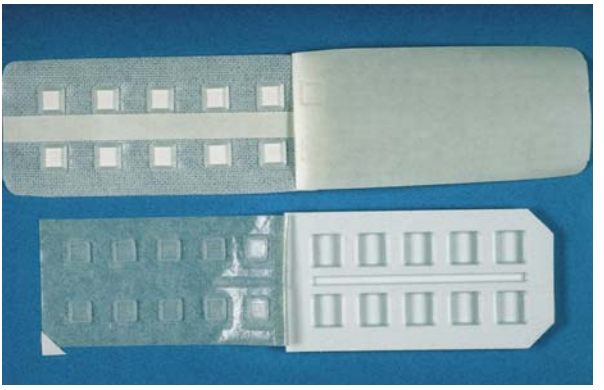

Fig. 3.3 Plastic chambers: van der Bend Square Chamber (*up*); IQ Square Chamber Chemotechnique (*down*)

The chambers are made of thin and soft polyethylene foam material, thus making them even more comfortable for the patients.

The highest quality hypoallergenic surgical tape is used for the IQ Ultra. Each strip of 10 chambers of IQ Ultra is attached to a protective plastic cover with corresponding compartments, which makes it possible to reattach the tape after advance filling of the chambers with the allergens.

The IQ Ultra Application device makes advance filling of test substances even easier. The device is specifically designed for the IQ Ultra. It is cost effective and saves nurses/technicians time, as they can prepare test series up to two weeks prior to use.

The IQ Chamber and the IQ Ultra Chamber are marketed by Chemotechnique Diagnostics, Modemgatan 9, 23539 Vellinge, Sweden (Tel.: +46-40-466077, Fax: +46-40-466700, e-mail: info@chemotechnique.se).

3.3.2.2
van der Bend Square Chamber

The van der Bend Square Chamber is made of an additive-free polymer. Undesired side effects in the form of allergic reactions to the test unit itself are avoided due to the chemical stability of the polymer.

van der Bend Chambers can be delivered already fixed on tape and also joined in a row without tape, which makes it easy to apply the test on a porous adhesive (e.g., Fixomull Beiersdorf) that can be chosen by the dermatologist carrying out the test.

The volume of the chamber is 100 µL and the inside area of the chamber is $10 \times 10\,mm^2$ ($100\,mm^2$). The distance between the chambers is 15 mm.

There is a standard Whatman filter paper $1 \times 1\,cm^2$, mechanically fixed without glue in each chamber (Fig. 3.3).

The van der Bend Square Chamber is marketed by van der Bend B.V., Postbus 73, 3230 AB Brielle, The Netherlands (Tel.: +31-18-1418055, Fax: +31-18-1417450, e-mail: info@vanderbend.nl).

3.3.2.3
Haye's Test Square Chamber

The Haye's Test Square Chamber is made of a white speenlaced hydrophilic unbleached nonwoven polyester, devoid of any allergenic properties. The chambers are supplied in units of 10 square chambers (in two rows of 5 chambers per row) on a hypoallergenic solventless acrylic adhesive (MED5761U). The tape with the chambers is protected by a transparent protection cover.

The volume of the chamber is 40 µL and the inside area of the chamber is $8 \times 8\,mm^2$ ($64\,mm^2$). The bottom of the chamber is filled with Whatman filter paper, $0.6\,cm^2$, fixed without adhesive. The distance between the chambers is 9 mm in the row and 23 mm between the rows. The width of the tape is 70 mm and the length is 120 mm. Chambers are delivered in a box containing 100 units.

For the use of Haye's Test Chambers (when kept in the refrigerator), Haye's Test Chambers Sealings have been developed (designed covers). They are made of environmentally

responsible synthetic material and cover all 10 test chambers of the plaster without having to remove the Kraft release liner beforehand. Haye's Test Chambers Sealings are delivered in boxes containing 60 pieces.

The Haye's Test Square Chamber is marketed by HAL Allergenen Lab.B.V. Parklaan 125, 2011 KT Haarlem, The Netherlands (Tel.: +31-23-5319512, Fax: +31-23-5322418, e-mail: sales@hal-allergic.nl).

3.3.2.4
allergEAZE Patch Test Chamber

Each patch test panel consists of two rows of 5 square ($8 \times 8\,mm^2$) inert acetal copolymer (Kepital) chambers. The chambers are mounted on a rectangular patch ($125 \times 70\,mm^2$) made of nonwoven polyester.

The nonwoven panel material flexes to allow improved freedom of movement. The chamber volume is 40 µL. The spacing between chambers is 10 mm. The spacing between rows is 24 mm.

The panel adhesive is an acrylic copolymer emulsion, consistent with state-of-the-art hypoallergenic surgical tapes.

allergEAZE(tm) Patch Test Allergens* and Chambers are marketed by: SmartPractice® Canada, 2175 29th Street NE Unit 90, Calgary, AB T1Y 7H8, Canada, Tel: +1 866-903-2671 Fax: +1 866-903-2672, E-mail: info@allergeaze.com, manufactured by brial allergen GmbH, Germany.

3.3.3
Reinforcement of Patch Test Units

The patch test units may be reinforced by extra tape stuck at the margins or covering the total surface of the original tape and extending over its margins. The procedure is particularly recommended in hot climate to avoid detachment of the strips. Its use is also advisable but facultative in temperate climate.

Various tapes are convenient for this purpose: Fixomull Beiersdorf, Scanpor Alpharma, Micropore 3 M.

3.4
A General Overview of Allergens

3.4.1
Allergens

The first standardized allergens (in the 1970s) were manufactured and marketed by Trolab in Denmark. At that time, the company has worked in close cooperation with former ICDRG members.

Nowadays, the standard and/or additional series of patch test allergens are sold by three companies, working in close connection with the ICDRG or other international and/or national groups.

- Trolab Patch Test Allergens, Allmiral Hermal GmbH, 21462 Reinbek, Germany (Tel: + 49-40-727040, Fax: + 49-40-7229296, e-mail: info@hermal.de).
- Chemotechnique Diagnostics, Modemgatan 9, 23539 Vellinge, Sweden (Tel: + 46-40-466077, Fax: + 46-40-466700, e-mail: info@chemotechnique).
- allergEAZE/SmartPractice Canada, Inc., 2175 29th Street NE Unit 90 Calgary, AB TIY 7H8, Canada, (Tel: +1 602225-0595, e-mail: info@smartpractice.com) (manufactured by Brial allergen GmbH, Germany).

According to those supplier's product catalogues, the allergens can be considered chemically defined and pure. The vast majority of allergens of the standard and/or additional series are dispersed in white petrolatum (Fig. 3.4). The petrolatum used as a vehicle is considered to be the purest on the market [10].

White petrolatum can be considered inert when applied onto the skin, but may be responsible in exceptional cases for an irritant reaction.

A few allergens cannot be dispersed in petrolatum due to their chemical instability. This is the reason why they are supplied in aqueous solutions. Some examples include formaldehyde, Cl + Me-isothiazolinone, phenylmercuric acetate, coco- amidopropyl-betaine, ammonium thioglycolate, chlorhexidine digluconate, benzalkonium chloride, etc. Hydrocortisone-17-butyrate is dissolved in ethanol 70%. An extensive list of chemicals not available in marketed lists of allergens has been gathered in de Groot's textbook [11]. This provides useful and accurate information about test concentrations and vehicles.

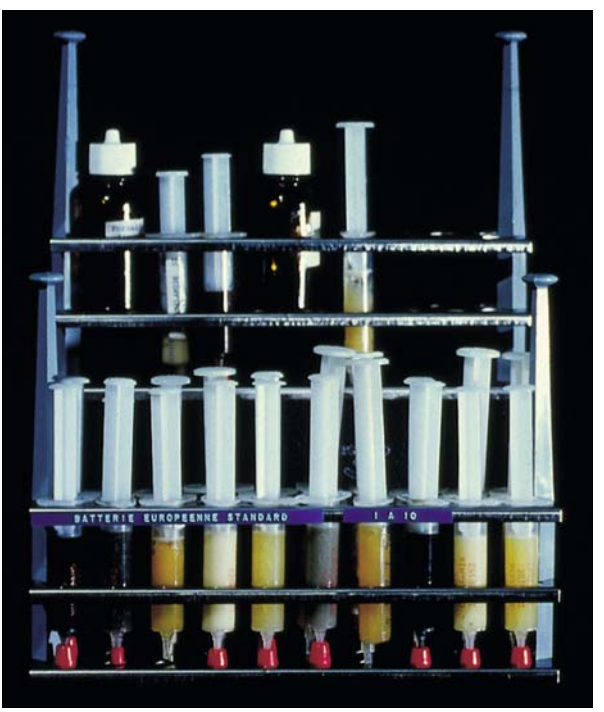

Fig. 3.4 Tray with contact allergens (standard series)

The vehicles that are referred are water, acetone, ethanol 70%, methylethylketone, olive oil, and petrolatum. Liquid vehicles are recommended for some allergens, since they facilitate penetration into the skin, but they have also some drawbacks. Solvents may evaporate, which does not favor exact dosing, and most test solutions must be freshly prepared. Liquid vehicles are used mainly when testing chemicals and products brought by patients and in research projects.

In textbooks on contact dermatitis and patch testing, and in suppliers' catalogues, the concentration of an allergen is given as a percentage. In one catalogue [12], molality (*m*) is given together with percentage (weight/weight). The traditional method of presenting concentrations as a percentage is simple and probably practical, but has been questioned [13], as we do not know if this means weight/weight, volume/volume, volume/weight, or weight/volume. Especially when comparing substances and in research projects, it is the number of moles applied that is of interest.

Finding the ideal test concentration is complicated; the currently recommended concentrations have been determined taking many important factors into account.

The general principle has always been to use the highest concentration that does not provoke any irritation when testing in groups of patients enrolled in prospective joint studies. Doing so, false-positive (irritant) and false-negative (due to a too-low concentration) reactions are avoided. Therefore, the choice of the concentration tends to reach an ideal (but sometimes unattainable) compromise.

In Trolab and Chemotechnique series, the substances with petrolatum vehicle are supplied in 5 mL polypropylene syringes, while those in a liquid solution are supplied in 10 mL polypropylene dropper bottles. The allergEAZE allergens, in either petrolatum or liquid base, are supplied in 5 mL color-coded polypropylene tubes.

The allergens should be kept in a cool dark place (refrigerator) to minimize degradation. In accordance with their stability, it is recommended that all substances should be renewed according to the expiry stated on the labels of the allergens. Nonmarketed allergens are prepared freshly; allergens diluted in liquids should be kept in dark bottles.

3.4.2 Bioavailability of Allergens

To obtain optimal bioavailability of an allergen, one can influence the following five parameters:

- Intrisic penetration capacity
- Concentration
- Vehicle
- Occlusivity of patch test system and tape
- Exposure time

Since it is desirable to remove all test strips at the same time, usually at day 2 (48 h), four factors remain and can be varied and optimized by the manufacturers of patch test materials and allergen preparations and by the dermatologist responsible for the testing.

3.4.3 Quality Control of Allergens

The dermatologist is recommended to obtain protocols of chemical analyses and data on purity from suppliers of test preparations.

3.4.4 Appropriate Amounts of Petrolatum to be Applied at Patch Testing

The prerequisite for a patch test is the requirement that the whole test area is covered with the allergen.

The ideal test situation is (a) the test area completely covered with the test preparations and (b) without any spreading outside the test area, to avoid overlapping at reading.

So far, there were no recommendations related to the amounts of petrolatum to be applied to reach this goal. Bruze et al. [14] have recently conducted a study on behalf of the ESCD to answer this important question. After several trials, they concluded that, when using the Finn Chambers, the optimal dose for pet. preparation was 20 mg. Similar studies were conducted with the van der Bend Chamber. The authors could not draw a definite conclusion, but a minimal dose of 35 mg seems advisable. No similar studies do exist for the other plastic square chambers.

3.4.5 Appropriate Amounts of Liquids to be Applied at Patch Testing

The prerequisities are similar to those described in Sect. 3.4.4. The Malmö team conducted a study [15], the aims of which were similar to those of the previous study (see Sect. 3.4.4) [14].

The conclusions are clear-cut:

- For water solutions, the Finn Chamber is highly recommended. The amount of liquid, delivered by a calibrated pipette, is 15 or 20 µL.
- For ethanol and acetone solutions, the van der Bend Chamber represents the unequivocal choice. The amount of liquid that fulfils requirements is 20 µL.

The chambers are immediately applied onto the skin to avoid evaporation of liquids. No irritation from ethanol or acetone is noted.

No similar studies do exist for the other plastic square chambers.

3.5 Specific Recommendations When Considering Patch Testing Patients

Some general rules as well as recommendations have to be taken into consideration when patch testing patients. This seems useful in practice.

3.5.1 Patch Testing on Intact Skin is Critical

The general rule is to avoid by any means patch testing at skin sites presenting currently or recently any type of dermatitis, to avoid false positive reactions and/or the angry back syndrome (see Sect. 3.14.2). This includes not only contact dermatitis (either primary or "id" reaction) but also atopic dermatitis, nummular eczema, and seborrhoeic dermatitis. Similar considerations are applied to various skin diseases, such as pityriasis versicolor, psoriasis, lichen rubber planus, pityriasis rubra pilaris, pityriasis lichenoides, pityriasis rosea, Darier's disease, and others. Complete healing or remission is needed before patch testing.

Atopic dermatitis is of special concern: it is up to the clinician to decide when patch testing can be performed. A good criterion is perhaps to consider that the patient is free of any inflammatory phase of the disease, does not require any "active" topical drugs (tacrolimus, pimecrolimus, corticosteroids) and is exclusively treated by emollients, useful for treating xerosis.

3.5.2 Medicaments and Patch Testing

3.5.2.1 Corticosteroids

Treatment of test sites with topical corticosteroids [16] can give rise to false-negative reactions.

Testing a patient on oral corticosteroids creates uncertainty. The problem was studied 25–30 years ago [17] by comparing the intensity of test reactions before and during treatment with corticosteroids (20–40 mg prednisone). Diminution and disappearance of test reactions were noted in several cases, but not regularly. These findings have been interpreted as allowing us to test patients on oral doses equivalent to 20 mg of prednisone without missing any important allergies. However, the test reactions studied were strong (+++), and fairly questionable reactions were not evaluated. A recent study called this dogma in question again [18]. When patch testing with serial dilution tests with nickel, it was found that the total number of nickel patch tests decreased significantly when the patients were on 20 mg of prednisone compared to those on placebo. The threshold concentration to elicit a patch test reaction increased and the overall degree of reactivity to nickel shifted toward weaker reactions. We conclude that interpretation of patch test results in patients treated with corticosteroids needs great caution; repeating patch testing after treatment discontinuation can be useful when in doubt.

3.5.2.2 Antihistamines

The interference of antihistamines on patch test results is a subject of controversy.

There are very few studies referring to this specific question. In one study, oral loratadine was found to reduce patch test reactions, evaluated clinically and echographically

[19]. These results also give the dermatologist a feeling of uncertainty. Therefore, in most clinics, antihistamine treatment is discontinued during testing, which is deferred. However, this option is not universally accepted [20].

3.5.2.3 Immunomodulators

There was so far no comparison of patch test reactions in allergic patients before, during, and after treatment with oral ciclosporine.

We treated 12 patients suffering from current allergic contact dermatitis of the hands (from cement) with oral ciclosporine (3 mg kg^{-1} day^{-1}). After 3 weeks of treatment, the symptoms were notably reduced; on the other hand, patch tests with potassium dichromate, applied before and during treatment, did not show any differences in terms of positivity and intensity of the reaction [21].

No information exists regarding the influence of azathioprine and cytostatic agents on patch test results.

At present, caution is needed as regards the current use of the new topical immunomodulators tacrolimus and pimecrolimus, since it has been demonstrated that they are efficient in treating atopic dermatitis.

3.5.2.4 Irradiation

Irradiation with UVB [22] and Grenz rays [23] reduced the number of Langerhans' cells and the intensity of patch test reactions in humans. Repeated suberythema doses of UVB depressed reactivity even at sites shielded during the exposures. This indicates a systemic effect of UVB [22].

From a practical point of view, avoid patch testing on markedly tanned persons, and a minimum of 4 weeks after heavy sun exposure should be allowed before testing.

3.5.3 Pregnancy and Patch Testing

There are no indications that the minute amounts of allergens absorbed in patch testing could influence the fetus, but in cases of miscarriage or deformity it is natural to blame several things, including medical investigations. Therefore, the general rule adopted by the members of the ICDRG is: do not test pregnant women, taking into account medico-legal considerations, not scientific ones. In some clinics, this view is also adopted for lactating women.

3.5.4 Patch Testing in Children

In children, patch testing has the same indications as in adults. Most authors agree that patch testing in children is safe, and the only problem being mainly technical because of

the small patch test surface [24]. It is usually advised to use the Finn Chamber. Reinforcement of patch test units is suitable due to hypermobility of children, which may result in loss of patch test materials.

Instructions should be given to parents about the test procedure and the measures that may be taken to optimize the patch test conditions [24].

There has been much debate about the concentrations of allergens to be used in children. Some authors have recommended lower concentrations, but nowadays, there is a general consensus of using the same concentrations as in adults. Nevertheless, it is well-known that irritant reactions from patch testing are more frequent in children. When in doubt, the clinician is advised to retest with a lower test concentration. The problem is raised mainly in children under the age of 5. Similarly, most authors agree upon the fact of applying in children the classical standard series, as well as additional series, if needed [24]. Some authors have advocated the use of a limited series of patch tests [25] adapted for the usually more restricted environment of children, but there is no general agreement about this opinion.

3.6 Application of Patch Tests on the Skin: Some Practical Suggestions

The accurate application of patch test units onto the skin is a prerequisite to ensure optimal reading and interpretation of patch test results.

Some suggestions to optimize the technique of application are listed below:

3.6.1 Test Sites

The preferred site is the upper back (Fig. 3.5). For a small number of allergens, for example, at retesting, the outer aspect of the upper arm is also acceptable. False-negative results can be obtained when testing on the lower back or on the volar forearms.

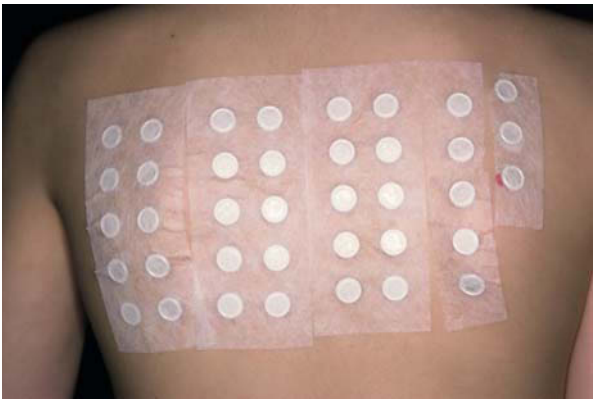

Fig. 3.5 Application of patch tests (Finn Chambers) on the upper back

The avoidance of applying patch tests on naevi or seborrhoeic keratoses is self-evident, but not always respected. When lesions are numerous, and do not allow proper application of tests, the choice of another patch testing site is mandatory.

3.6.2 Removal of Hair

On hairy areas of the back, it is difficult to get acceptable skin contact, and for this reason clipping is recommended. However, a combination of clipping, petrolatum, and tapes sometimes contributes to the irritation seen, which makes reading somewhat difficult. It is advisable to clip hair one or two days before patch testing, whenever possible. This procedure does not offer absolute guarantee in terms of skin irritation.

3.6.3 Degreasing of Test Site

In cases of oily skin, gentle treatment with ethanol or other mild solvents could be recommended. The solvent must evaporate before the test strips are applied. Practically, no degreasing is performed in European clinics.

3.6.4 Application of Test Strips

Test strips should be applied from below with mild pressure to remove air pouches, followed by some moderate strokes with the back of the hand to improve adhesion [26].

3.6.5 Instructions to Patients

Patients should be informed as to the aim of the test: about avoidance of showers, wetting the test site, irradiation and excessive exercise, and about symptoms such as itch and discomfort. Occasional loosening of patches can occur; frequent check by the patient is advisable during the application period. Reinforcement of test strips is recommended (material delivered to the patient when patch tests are applied). Such written instructions and guidelines for patients are highly recommendable.

3.7 Reading Time

Reading is the most important step in the patch test procedure. It should be done by the clinician him or herself and interpreted carefully. There is a need for constructive dialogue between clinician and patient. This requires time, skill, and perseverance to

achieve the specific aim of tracing the source of allergy. The reading allows the clinician to complete past and current history in each individual patient. It cannot be dissociated from the search for relevance or nonrelevance (see Chap. 8). A decision must be made about whether to continue the investigations by additional patch tests and/or other tests such as repeated open application test (ROAT) for instance (see Sect. 7.4). Therefore, it may be considered that in many cases the reading is only an intermediate step in the investigatory process.

There are controversies in the literature regarding the optimal reading time, as discussed in the following sections. Therefore, the "best" reading time is always a matter of compromise.

3.7.1 Standard Patch Test Occlusion and Reading Time

The standard patch test technique involves application of the test allergen strips onto the skin under occlusion for 2 days (48 h). Conventionally, patch test reading is performed 15–30 min after the removal of the occlusive strips to allow the transient erythema caused by the occlusive effects of allergens and plasters to subside [27]. This will eliminate false-positive reactions. The 2-day occlusion ensures that adequate allergen penetration has occurred to provoke an allergic contact reaction on the test site.

Reading is further performed at day 3, 4, and 7 after occlusion (i.e., 1, 2, and 5 days after the removal of the patch test strips) thereafter.

3.7.2 Conventional Patch Test Reading Time

Conventionally, patch test reading is performed in most patch test clinics at day 2 when the patch test strips are removed, and again at day 4. Allergic reactions are then identified and checked for relevance. Patients are then instructed to report back to the dermatologist if any additional positive reaction appears at day 5 or beyond to detect any late reactors or sensitization that may have occurred.

3.7.3 Reading at Day 2, Day 3, Day 4

Positive reactions at day 2 after the removal of the test strips should not be considered positive unless the reactions persist into day 3 and beyond [28]. True allergic reactions should persist or may appear at days 3 and 4.

3.7.4 Reading at Day 7

Reactions occurring at day 7 or later are regarded as late reactions. Some allergens are "late reactors," and delayed positive reactions may appear at day 5 or later. Examples of such late reactors include neomycin, corticosteroids [29], and many others. This is particularly

true for corticosteroids: in many instances, when readings are made only on day 2 and day 4, some positive reactions are missed, since they appear later on [29]. In some cases, late reactions reflect active sensitization (see Sect. 3.14.1), but this latter interpretation requires cautious appreciation. To corroborate this point, a late reaction to paraphenylenediamine is often considered an active sensitization. It is certainly not always the case, as demonstrated in a recent observation [30].

3.7.5 Single Reading vs. Multiple Reading

Single reading carried out at day 2 may result in false-negative reactions. Reading of diagnostic patch test should not cease at day 2, as numerous allergic reactions need more time to evolve to become positive. Further recommended reading times include day 3, day 4, and day 7. In most patch test clinics around the world, patch test reading is carried out at day 4.

3.7.6 Day 3 vs. Day 4 Reading

Recent studies have indicated that day 4 reading yields better results (fewer false-negative results) than day 3 reading alone, because some positive results appear only after day 3 [31].

At this stage, it must be recalled that several exogenous factors, e.g., surface concentration of the allergen, total amount applied, penetration properties of the allergens and the vehicle, patch test technique, and allergen exposure time are major determinants in the elicitation of positive patch test reactions [32].

3.7.7 One-Day Occlusion vs. Two-Day Occlusion

Most authors advocate an exposure time of 48 h. A few comparisons of 1-day (24 h) and 2-day (48 h) allergen exposure show some reactions positive only at day 1 (24 h) and some positive only at day 2 (48 h). A 1-day exposure would reduce the number of questionable reactions. No definite conclusion can be drawn from the studies published to date [33].

In tropical climates where the environmental temperature and humidity are high, 1-day occlusion may be adequate to elicit positive patch test reactions. The shorter occlusion will be more tolerable to the patients and is more likely to improve compliance and cooperation from patients to accept the patch test procedure [34].

3.7.8 Marking the Skin

When several readings are carried out, it is extremely useful to “mark” the patch test sites.

The Chemotechnique Skin Marker is a suitable marking pen designed for marking efficiently the patch test sites. Its content is methylrosanilin (gentian violet), 1%; silver

nitrate, 10%; denaturated ethanol/aqua in equal parts ad 100%. The duration of the marking is approximately 5–7 days. Marking may be repeated to ensure durable staining.

For dark skin types or when a nonstaining ink is required, the Chemotechnique UV Skin Marker (yellow fluorescent ink) provides a good alternative. Its content is disulphonic acid derivate of stilbene, 2%; dimethylsulphoxide (DMSO)/denaturated ethanol in equal parts ad 100%. DMSO increases fixation of the ink to the outer layer of the skin. The tip has tapered edges, which facilitates precise markings. The duration of the marking is approximately 5–7 days. The UV Skin Marker requires the use of a Wood's light at each reading session (Fig. 3.6).

Some authors do not use skin markers, but a reading plate (i.e., Reading Plate for Finn Chambers on Scanpor Epitest), which is a real template for the patch skin sites (Fig. 3.2).

A practical, clean, durable, and inexpensive alternative method of marking was reported recently [35]. It requires A4 (21 × 29.7 cm^2) transparencies used for transparent photocopies, and two or three colors dry erasable pens. Contours of patch test areas are carefully marked with a pen. The transparency is used for further readings.

3.7.9 Immediate Urticarial Reactions to Some Allergens

Seldom, some allergens (e.g., balsams of Peru, cinnamic aldehyde) are responsible for an immediate urticarial reaction about 20–30 min after applying patch tests. It is the reason why some authors remove the tests for a short while at 30 min and reapply them immediately at the same site. This practice, that is in essence wise, is not usually carried

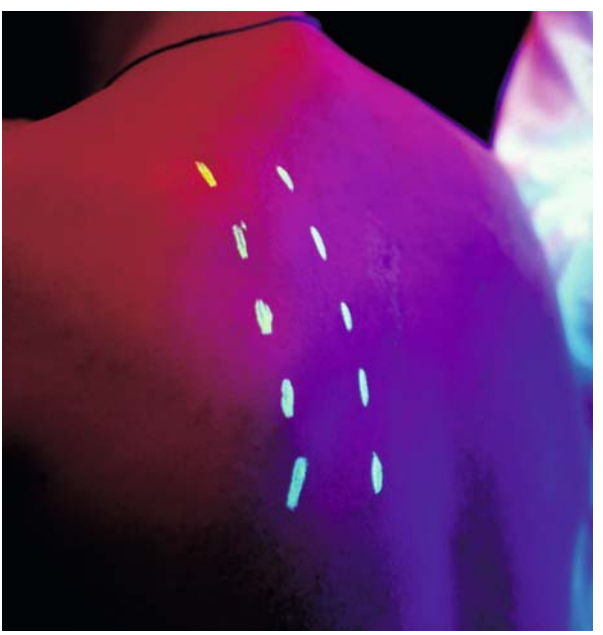

Fig. 3.6 Marking the skin with the Chemotechnique UV Skin Marker: examination under Woods's light

out by dermatologists. The reaction can be reproduced when applying the allergen in an open test. Meticulous investigators apply systematically in each patient balsams of Peru on the volar aspect of the left forearm, and cinnamic aldehyde on the volar aspect of the right forearm, as a open test (see Sect. 7.2). Readings occur at 20 and 30 min. In some cases, this observation has no clinical meaning, but in some others, it reflects the existence of a contact urticaria syndrome (see Sect. 10.1), coexisting eventually with ACD.

3.8 Reading and Scoring Patch Test Results

3.8.1 Nomenclature: Scoring Codes

It is important for patch tests to be scored according to the reaction seen and not only according to the interpretation placed on the reaction by the reader (Fig. 3.7). Irritant reactions should be recorded as positive irritant and not as negative. In our view, the best scoring system remains that recommended by Wilkinson et al. [36] and reproduced in Table 3.1. Some variants of scoring do exist in textbooks of contact dermatitis; they include the occasional occurrence of papules, as an additional clinical sign of + and ++ reactions. Papules are purposely omitted in our scoring system for two reasons: they do not provide any complementary useful information, and histopathological examination of papules observed in some positive patch test reactions reveals that they are in fact tiny vesicles.

3.8.2 Rating Patch Test Reactions Based on Digital Images

A recent study has been conducted in Germany [37] to assess the diagnostic validity of readings of 20 digital images of various patch test reaction grades by congress attendants. One hundred twenty-two volunteers took a patch test quiz offered during the 8th ESCD meeting, September 2006, Berlin. The "gold standard" grading determined by an EECDRG expert panel was disclosed while the quiz was open. The distinction between ?+ and + reactions proved rather difficult, but most images prompted a fair proportion of correct classifications.

Results were largely valid. Thus, the method could be used for continuing medical education and standardization in multicentre networks.

3.8.3 Some Remarks About Reading and Scoring

3.8.3.1 Size of the Reaction

The size of the reaction is different from case to case. The use of current patch test units (i.e., chambers) has limited the size of the reaction to the patch area in most cases;

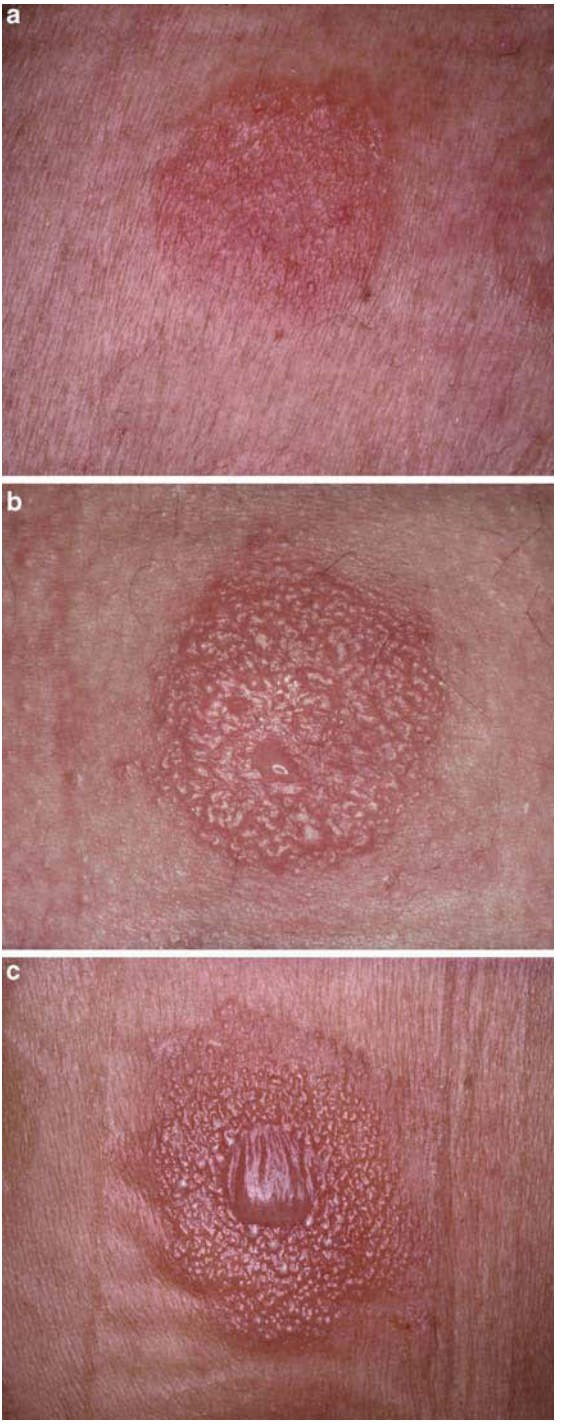

Fig. 3.7 Scoring positive allergic patch test reactions. (**a**) + reaction; (**b**) ++ reaction; (**c**) +++ reaction (see explanations in text)

Table 3.1 Scoring of patch test reactions according to Wilkinson et al. [36]

Score	Interpretation
–	Negative reaction
?+	Doubtful reaction[a]; faint erythema only
+	Weak (nonvesicular) reaction[b]; erythema, slight infiltration
++	Strong (edematous or vesicular) reaction; erythema, infiltration, vesicles
+++	Extreme (bullous or ulcerative)[c]
IR	Irritant reactions of different types
NT	Not tested

Note that photopatch tests (see Chap. 5, Sect. 5.5) are graded similarly with a prefix Ph: Ph–, Ph ?+, Ph+, Ph++, Ph+++, Ph IR, Ph NT
Reading and scoring have to be repeated at each individual visit to check the progression or regression of the reaction (day 2, day 4, day 6, or day 7)
[a]?+ is a questionable faint or macular (non-palpable) erythema and is not interpreted as a proven allergic reaction
[b]+ is a palpable erythema, suggestive of a slight edematous reaction
[c]From coalescing vesicles

nevertheless, the reaction may sometimes spread all around the patch area, outside the chamber's margins (see Sects. 3.4.4 and 3.4.5). It can be concluded that the reactions are more limited nowadays (thus more comfortable for the patient) than previously, when older patch tests (i.e., nonchamber) units were used. Readings are therefore easier because of the absence of overlap between neighboring positive reactions.

3.8.3.2 Edge Effect

The occurrence of "ring-shaped" allergic positive patch test reactions to allergens dissolved in a liquid vehicle (i.e., formaldehyde) is not uncommon [38]. Such reactions can be explained by the accumulation of the chemicals at the periphery of the patch test site. We previously coined the term "edge effect," because some patch test units are square in shape. When using such units, the liquids accumulate at the "edges" of the squares. The occurrence of the "edge" or "ring" effect could be due to pressure [39]. Besides this pressure mechanism, capillary migration could be responsible for an enhanced edge effect. Exceptionally, "ring-shaped" reactions can occur with allergens dispersed in petrolatum, the explanation of which could also be the effect of pressure (Fig. 3.8).

A particular type of edge effect (Fig. 3.8) can be seen when patch testing with corticosteroids [40]. The margins of the positive test are red, while the central area is whitish. This could be related to the vasoconstrictive effect of the corticosteroid, due to an enhanced penetration of the chemicals in the central area. Vasoconstriction and reduction of the inflammatory process most probably counteract the expression of the allergic response.

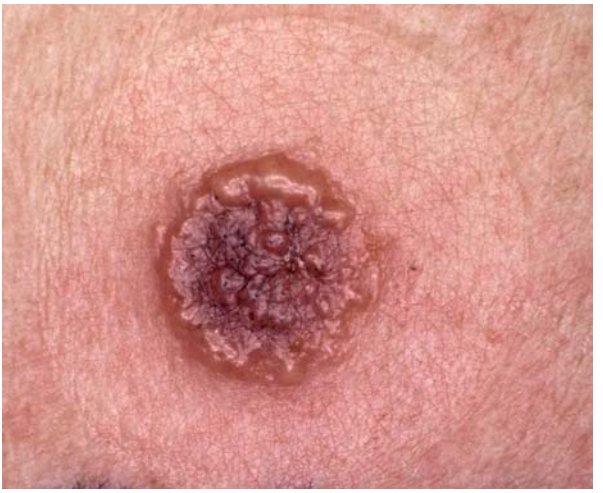

Fig. 3.8 Edge effect. Allergic positive ++ patch test reaction to paraphenylenediamine. Such a reaction can be explained by the accumulation of the chemicals at the periphery of the patch test chamber

3.8.3.3 What Must be Done in Case of "?+" (Doubtful/Questionable) Reactions?

"?+" reactions are labeled "doubtful" in the files. There is no real problem when allergens of the standard and/or additional series are concerned, since that type of reaction reflects in a few cases the true allergic nature of the reaction.

More attention must be paid if the reading occurs in a hot climate, due to the potentially increased irritancy of some allergens, such as the fragrance mix.

A caveat does exist: "?+" reactions cannot be easily interpreted as irritant or allergic when patch testing with less common allergens, and even more so with products of unknown content, the irritancy of which is to a large extent unknown.

To circumvent these difficulties, the following strategy can be adopted by the clinician:

a. Repeat the patch test in the patient to check its reproducibility. This may include serial dilutions of the suspected allergen (dose/concentration relationship).
b. Apply the same test in control subjects.
c. Conduct additional investigations in the patient, such as open tests, semi-open tests and ROATs, and eventually use tests.

To strengthen the validity of such investigations, note that, when applying patch tests in the same patients (left vs. right sides of the back), most discrepancies in patch test readings do occur with "?+" and/or "+" reactions [41].

3.8.3.4 What Must be Done in Cases of Pustular Reactions?

The occurrence of pustules in positive allergic patch test reactions is common. This is particularly true with metallic salts (chromate, nickel, cobalt, etc.) mainly, but not exclusively, in atopics. If some doubt does exist in relation with its allergic meaning, repeating the

tests would be wise, including a serial dilution test. This step-by-step procedure can avoid false-positive reactions and permits an unequivocal positive or negative reassessment of the allergic nature of the test.

3.9 Irritant Patch Test Reactions

In older days, when patch testing did not respond to definite rules (due to the lack of international standardization), irritant reactions were not uncommon in practice. This was due to (a) the nature of substances and/or mixtures applied to the skin, (b) a too-high concentration of some allergens, above the threshold of irritation.

Such irritant reactions may still occur nowadays when inappropriate methodology is used (Fig. 3.10).

The clinical signs of irritant patch test reactions are varied in relation with the nature and/or concentration of irritants [42].

They are classically described as follows:

a. Erythematous reactions
   Erythema is strictly limited to the site of application of substances, with sharp well-delineated margins. This means that when a square patch test unit is used, erythema has a square shape. The reaction is sometimes discretely scaly, but usually not edematous.
   Allergens from the standard and/or additional series may provoke in some patients mild erythematous irritant reactions; they occur "at random" and are probably related to skin hypersensitivity in these patients.
   Among allergens of the standard series, fragrance mix and thiuram mix are usually quoted as candidates for such marginal irritant reactions. In those cases, strategies to be applied for further patch testing are explained elsewhere (see Chap. 7).
b. Purpuric reactions
   Purpuric patch test reactions are common with some allergens, in particular, cobalt chloride. About 5% of patients tested with 1% cobalt chloride in petrolatum show this petechial hemorrhage (Fig. 3.9). Histopathologic examination reveals slight perivascular

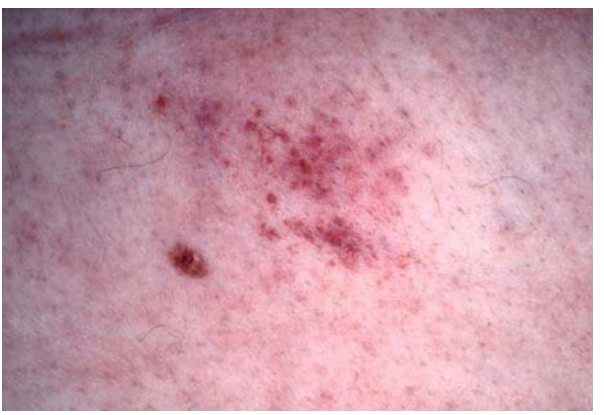

Fig. 3.9 Purpuric patch test reaction. Purpuric macules are scattered at random on the patch test application site. Mainly observed with cobalt chloride (see explanations in text)

lymphocytic infiltration, swollen endothelium, and extravasation of erythrocytes, mainly localized to the epidermis and acrosyringium. Purpuric reactions can also be observed when patch testing with paraphenylenediamine, IPPD, and some drugs.

c. "Soap or shampoo effect" reactions
   These are so named because they are typically produced by patch tests with soaps and detergents. The skin is red or slightly shiny and wrinkled; there are usually no vesicles; pruritus is uncommon. It is therefore not recommended to test with soaps or detergents.

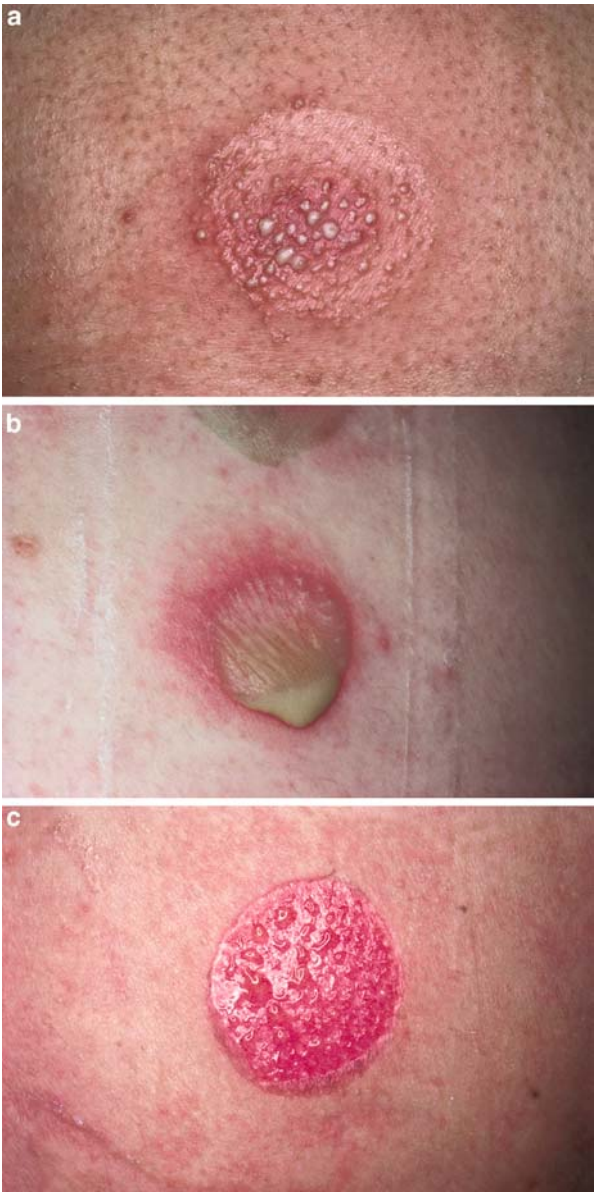

Fig. 3.10 Examples of irritant reactions. (**a**) pustular follicular; (**b**) pustular diffuse; (**c**) necrotic

Such reactions may still occur with soluble oils (which do contain detergents), when the test concentration is inappropriate.

d. Blistering (or bullous) reactions
   Blistering occurs after testing with nondiluted or overly concentrated caustic products, such as gasoline, kerosene, and turpentine. Patch tests with quaternary ammonium salts may blister even when low concentrations are used.
e. Pustular reactions
   These are sometimes consecutive to bullous reactions. Pustules are the result of an influx of polymorphonuclear neutrophils (sterile pustules) or are less often due to superinfection (by staphylococci). In those circumstances, a unique large pustule is observed at the site of application (Fig. 3.10b).

   Another type of pustular reaction may occur. The application area, uniformly erythematous, is dotted with small follicular pustules (Fig. 3.10a). This type of reaction mainly occurs with metallic salts (such as chromate, cobalt, nickel, copper) in atopic patients. The reaction can be exclusively irritant in nature or be superimposed onto a true allergic reaction. Formerly, a similar pattern of reaction (purely irritant, nonallergic) was observed when croton oil was applied to the skin ("croton oil effect").
f. Necrotic or escharotic reactions
   These are the most violent irritant reactions. For example, caustic soda or kerosene provokes such reactions (Fig. 3.10c).

3.10 False-Positive Patch Test Reactions

False-positive reactions can be defined as positive patch test reactions occurring in the absence of contact allergy [4]. These are manifold; nevertheless, the following list (Table 3.2) is mainly related to technical errors (which can be avoided) or to a misinterpretation of the test results, in particular, when using inadequate concentrations of allergens.

Table 3.2 False-positive patch test reactions

1. Too high a test concentration for a defined allergen
2. Impure or contaminated test substance
3. The vehicle is irritant (especially solvents and very rarely petrolatum)
4. Excess of test preparation applied
5. The test substance, usually as crystals, is unevenly dispersed in the vehicle. This can occur when prepared at the hospital (i.e., not by manufacturers)
6. Current or recent dermatitis at test site (Excited Skin Syndrome) [43]
7. Current dermatitis at distant skin sites (Excited Skin Syndrome) [43]
8. Pressure effects of tapes, mechanical irritation of solid test materials, furniture, and garments (see Sect. 3.14)
9. Adhesive tape reactions
10. The patch itself has caused reactions

Some of them are self-evident and can be predicted and monitored by the dermatologist carrying out patch testing, while others cannot.

3.11 False-Negative Patch Test Reactions

False-negative reactions can be defined as negative patch test reactions occurring in the presence of contact allergy [4]. The most common causes have been summarized in Table 3.3.

Some of them are self-evident and can be predicted and monitored by the dermatologist, while others cannot. Examples of the latter category may arise when (a) testing has been carried out in a refractory or "anergic" phase [43]; (b) the test does not reproduce the clinical exposure (multiple applications), where some adjuvant factors are present (sweating, friction, pressure, damaged skin), or penetration at the site is lower than that of clinical exposure (eyelids, axillae). A stripping skin technique is recommended in the last case, where the test sites are stripped with tape before application of test preparations (see Sect. 7.1).

The differential diagnoses photoallergy (see Chap. 5) and contact urticaria (see Sect. 10.1) should also be considered.

3.12 Compound Allergy

The concept of "compound allergy," popular among dermatologists, cannot stricto sensu be considered a false-positive or negative patch test reaction. It is the reason why it is described in a separate section.

Table 3.3 False-negative patch test reactions

1. Insufficient penetration of the allergen
   (a) Too low a test concentration for a defined allergen
   (b) The test substance is not released from the vehicle or retained by the filter paper
   (c) Insufficient amount of test preparation applied
   (d) Insufficient occlusion
   (e) Duration of contact too brief; the test strip has fallen off or slipped
   (f) The test was not applied to the recommended site: the upper back.
2. The reading is made too early, e.g., neomycin and corticosteroids are known to give delayed reactions
3. The test site has been treated with corticosteroids or irradiated with UV (see Sect. 3.5.2)
4. Systemic treatment with corticosteroids or immunomodulators (see Sect. 3.5.2)
5. Allergen is not in active form, insufficiently oxidized (oil of turpentine, rosin compounds, d-limonene) or degraded

The term "compound allergy" is used to describe the condition in patients who are patch test-positive to formulated products, usually cosmetic creams or topical medicaments, but test negative to all the ingredients tested individually [44]. This phenomenon can sometimes be explained by irritancy of the original formulation, but in some cases it has been demonstrated that the reactivity was due to the combination of the ingredients to form reaction products. Another reason might be that the ingredients were patch tested at the usage concentrations, which are too low for many allergens (e.g., MCI/MI, neomycin). Pseudo-compound allergy, due to faulty patch testing technique, is likely to be more common than true compound allergy. In a recent review [45, 46], several proven or possible compound allergens were listed. The formation of allergenic reaction products can take place within the product ("chemical allergenic reactions") but also metabolically in the skin ("biological allergeneic reactions") [45].

The "quenching phenomenon" is a consistent finding whereby cinnamic aldehyde alone induces sensitization, but when mixed with other fragrance compounds such as eugenol or d-limonene induces no sensitization. Patients who are sensitive to cinnamic aldehyde can sometimes tolerate perfumes containing this allergen because of presumed chemical changes (quenching) that occur during the usual aging process of a "mature" perfume [47].

3.13 Cross-Sensitization, Concomitant Sensitization, Polysensitization

This section deals with situations wherein patients present several (two or more) contact allergies.

3.13.1 Cross-Sensitization

Cross-sensitization (syn.: cross-sensitivity, cross-allergy) means that contact allergy caused by a primary allergen is combined with allergy to other chemically closely related substances. In other words, in those patients who have become sensitized to one chemical (primary allergen), an allergic contact dermatitis can be provoked or worsened by several other related chemicals (secondary allergens).

A few examples are the following:

- A patient positive to *p*-phenylenediamine not only reacts to the dye itself, but also to immunochemically related chemicals that have an amino group in the *para* position, for example, azo compounds, some local anesthetics, and sulfonamides [48]
- Cross-sensitization occurs with some antibiotics: neomycin, framycetin, kanamycin, gentamycin
- Cross-sensitization is often mentioned with nonsteroidal anti-inflammatory drugs. This issue is controversial in the literature: in some cases, true cross-sensitization seems to occur (ketoprofen and tiaprofenic acid [49, 50], whereas in some others, reactions are interpreted as examples of concomitant sensitization (see Sect. 3.13.2)

- In the realm of plant dermatitis, true examples of cross-sensitization do occur (e.g., catechols from different species of Rhus), but some are misinterpreted [51], since they are representative of a concomitant sensitization (see Sect. 3.13.2)

When investigating cross-sensitization, it is essential to use pure test compounds.

3.13.2 Concomitant Sensitization

Concomitant sensitization (syn.: cosensitization, cosensitivity, simultaneous sensitization) should not be confused with cross-sensitization.

It refers to the circumstance that certain substances often occur together in some products and that sensitization to the different substances often takes place on the same occasion. Thus, often cosensitization occurs to nickel and cobalt on contact with nickel items where cobalt is present as an impurity, and towards chromates and cobalt on contact with cement. Lisi et al. [52] have conducted an extensive study on concomitant sensitization between different metals. The same applies to sensitization to various rubber chemicals (e.g., thiurams and thioureas) [53].

The synonym "simultaneous sensitization," preferentially used in some papers, only means that at reading positive patch test reactions to some noncross-reacting substances do occur at the same time, that is, during the same test session. This does not imply that the patient has been sensitized "simultaneously" (or not) to those substances; this cannot be assessed retrospectively.

3.13.3 Polysensitization

Polysensitisation (syn: multiple sensitization) refers to a specific population of patients who are "polysensitized," that is, sensitized to different categories of chemically nonrelated allergens. It has been arbitrarily stated that this concerns patients who are allergic to three or more categories of allergens [54]. A lack of knowledge still persists, as regards the respective role played by environmental and genetic factors [54, 55].

3.14 Unwanted Adverse Reactions of Patch Testing

The greatest hazard is omission of patch testing procedures in the management of patients who have certain dermatoses. Such omission dooms these patients to repeated attacks of avoidable contact dermatitis [56].

Side effects of patch testing patients are listed in Table 3.4. Some are described in detail. Such unwanted effects are seldom encountered in daily practice. In this respect, it must be emphasized that the risk-benefit equation of patch testing is much in favor of the benefit.

Table 3.4 Unwanted adverse reactions of patch testing

Patch test sensitization	("Active sensitization") see Sect. 3.14.1
The excited skin syndrome	("Angry back") see Sect. 3.14.2
«Ectopic» flare of dermatitis	On rare occasions, a positive patch test reaction may be accompanied by a specific flare of an existing or pre-existing dermatitis that was caused by the test allergen. This side effect can be minimized by testing patients free of any current active dermatitis
Persistence of a positive patch test reaction	A notorious patch test reaction for persisting for more than 1 month is that due, for example, to a 0.5% aqueous solution of gold chloride in a gold-sensitive patient. Its meaning is partly understood (see Sect. 2.1.3).
Pressure effect	This consists of a red, usually depressed mark "imprinted" into the skin. It is a transient effect due to the application of solid materials. In practice, it can be due to (a) the pressure of chamber's rings or squares. This is a physically induced edge effect, distinct from the chemically induced edge effect (see Sect. 3.8.2); (b) the use of allergens in a solid form
The Koebner phenomenon	A positive patch test reaction in a patient who has active psoriasis or lichen planus may reproduce these dermatoses at the patch test site during the weeks following patch test application [57]. The use of a topical corticosteroid usually quickly clears the lesion. Rarely, a similar Koebner phenomenon is observed in patients with lupus erythematosus [58] and lymphocytic infiltration of the skin (Jessner-Kanof) [59]
Hyperpigmentation	Hyperpigmentation from patch tests occurs infrequently and is most likely in darkly pigmented persons. If fades progressively after applying repeatedly topical corticosteroids. Sunlight or artificial UV exposure, immediately following removal of patch tests especially to fragrance materials, leads to hyperpigmentation of patch test sites in relation with photosensitivity, as in berloque dermatitis. This side effect is more common in Oriental populations (see Sect. 3.15.2).
Hypopigmentation	Post-inflammatory hypopigmentation may occur at the sites of positive patch test reactions. It is usually a transient event (e.g., phenol)
Bacterial and viral infections	These adverse reactions have been occasionally described, but are exceedingly rare
Necrosis, scarring and keloids	Foolhardy testing, with strong irritants (acids, alkalis, or chemicals of unknown composition) may produce such adverse reactions. Good practice of patch testing has entirely suppressed the occurrence of these complications, which are only of historical interest
Anaphylactoid reactions	Anaphylactoid reactions or shock from, e.g., neomycin, bacitracin have been reported. These can be considered exceptional

3.14.1 Patch Test Sensitization ("Active Sensitization")

By definition, a negative patch test reaction followed by a flare-up after 10–20 days, and then a positive reaction after 3 days at retesting, means that sensitization was induced by the patch test procedure. There is a risk of active sensitization from the standard and/or additional series. Common examples are *p*-phenylenediamine, thiuram mix, epoxy resin, sesquiterpene lactone mix, primula extracts, and in recent years, isothiazolinones [60] or acrylates [61]. The risk, however, is uncommon when the testing is performed according to internationally accepted guidelines. Sensitization by a patch test very rarely causes the patient any subsequent dermatitis or affects the course of a previous dermatitis.

In recent years, there has been a lot of concern about active sensitization from *p*-phenylenediamine. Gawkrodger and English [62] have made an extensive review of the literature and, when analyzing the different studies, they conclude the following:

- The overall percentage of active sensitization is very low (1–1.5%)
- Even in case of active sensitization, the risk of developing allergic contact dermatitis from hair dying is very small.

Moreover, late reactions to *p*-phenylenediamine are not always an indication of active sensitization [63].

In conclusion, it must be emphasized that the overall risk-benefit equation of patch testing patients is much in favor of the benefit. On the other hand, we advise against "prophetic" patch testing of non-dermatitic potential employees, because in that case the risk-benefit equation is very much in favor of the risk of active sensitization.

3.14.2 Excited Skin Syndrome ("Angry Back")

This represents an important issue. Mitchell [64] used the term "angry back" to describe a regional phenomenon caused by the presence of a strongly positive reaction, a state of skin hyperreactivity in which other patch test sites become reactive, especially to marginal irritants, such as formaldehyde or potassium dichromate. He believed that these concomitant "positive" reactions cannot be relied on. Indeed, when retesting, these reactions were negative. He suggested that the true index of sensitivity was falsely exaggerated by concomitant patch testing. Nickel sulphate and potassium dichromate were considered best examples of such false-positive reactions. To confirm or deny the significance of individual reactions found on the "angry back," he recommended sequential testing later with each substance alone.

Because patch test may be performed elsewhere besides the back, Maibach [65] and Mitchell [66] broadened the term "angry back" to the "excited skin syndrome" (ESS), which was extensively reviewed later on [67]. The pathogenesis of ESS has not yet been clearly elucidated.

When in doubt about the occurrence of ESS in a patient, the strategy to be conducted is individual *sequential retesting*, with each incriminated allergen, preferably on a different

skin site. This procedure can be completed by additional tests, such as ROAT tests (see Sect. 7.4). It is a matter of the utmost importance in medico-legal situations.

It is our experience that the ESS is less frequent nowadays, possibly for two main reasons: (a) patch testing only on intact skin in patients free of any current dermatitis; (b) using smaller amounts of allergens, in relation with new patch test units (chambers).

The ESS is distinct from the "status eczematicus," contrary to what is written in most textbooks on contact dermatitis. Status eczematicus means that, at many patch test sites, there are positive nonspecific reactions, due to a state of skin hypersensitivity. This does occur when general rules of patch testing are not respected, such as patch testing patients with active atopic dermatitis or other types of dermatitis. Status eczematicus makes reading impossible; it can be avoided by using correct procedures.

3.15 Patch Test Readings in Different Ethnic Populations

Most publications dealing with patch test readings refer to Caucasian populations. It seems important to know whether differences may occur when reading patch test results in different ethnic populations.

3.15.1 Patch Test Reading in Oriental Populations

3.15.1.1 Particular Aspects of Reading

The skin color in Oriental races (Japanese, Chinese, Korean, etc.) varies from white fair skin (equivalent to Fitzpatrick classification types II to IV) to dark complexion (equivalent to Fitzpatrick classification skin types V and VI).

For dark-skinned individual's (skin types V and VI) skin marking of patch test sites is important because by the second and fourth day, it is often difficult to identify the location of the patch test sites. Special markers incorporating silver nitrate (though it may cause irritant reactions) may be more effective than marking the skin test sites in a conventional way.

Goh in Singapore uses the following marker solution:

Gentian violet 1%
Meth Spirit (95%) 50%
Silver nitrate 20%
Distilled water to 100%

The fluorescence skin marker is an alternative.

For fair skin (type II to type IV), a patch test reaction is not difficult to interpret. Allergic patch test reactions are usually easily discernible. The erythema, papules, and mild

edema of allergic patch test reactions are usually very obvious in skin types II and III. In darker skin types (types V and VI), a mild positive allergic patch test reaction may be overlooked as the erythema may not be obvious. However, the edema and papules/vesicles are usually obvious and palpable.

In darker skin of Malays and Indians, allergic patch test reactions may be difficult to discern. Erythema is barely visible. Much will have to depend on the appearance of papules/vesicles and edema. Palpation of the patch test site may help to detect allergic reactions. Associated pruritus on papular eruptions on the patch test site helps to affirm the possibility of the presence of a positive allergic patch test reaction.

Finally, there is little evidence of statistically significant differences in the irritant response between Oriental and Caucasian groups [68]. Therefore, it can be anticipated that patch test irritant reactions are not more frequent in Asian than in Caucasian populations.

3.15.1.2 Pigmented Contact Dermatitis

Pigmented contact dermatitis is a particular entity characterized by a diffuse brown, slate-colored, greyish-brown, reddish-brown, or bluish-brown pigmentation. It occurs in the weeks following an acute episode of irritant or allergic contact dermatitis [69]. Pigmented contact dermatitis is rare in Caucasians but common in Mongoloids. Most recent cases have been reported from Japan. Various allergens have been incriminated, namely Naphthol AS, 1 phenyl-azo-2 naphthol, parabens, trichlorocarban, jasmine oil, rose oil, benzylsalicylate, musk ambrette, and some others. Positive patch tests to these allergens become hyperpigmented in the days or weeks following patch test application and remain so for long periods of time.

3.15.2 Patch Test Reading in Black Populations

It is surprising that in most textbooks on contact dermatitis, no mention is made about particular aspects of patch test reading in black populations. In practice, reading does not cause insurmountable difficulties.

Two specific points deserve special attention:

- Erythema is distinctly visible in some cases, or may present itself as a darker black hue in some others.
- In black skin, vesicles of eczematous reactions (including positive patch test reactions) do not tend to burst readily (Fig. 3.11); since they exhibit a yellowish hue (Fig. 3.12), they can be confounded with tiny pustules (Fig. 3.10). This particular aspect is certainly related to the fact that, in black skin, stratum corneum has more cell layers and requires more tape strips to remove it than that of Caucasoid stratum corneum [70].

The darker the skin, the more difficult it is to mark. For very dark skin, a fluorescent marking ink is probably best, the dots being located by a Wood's light in a dark room.

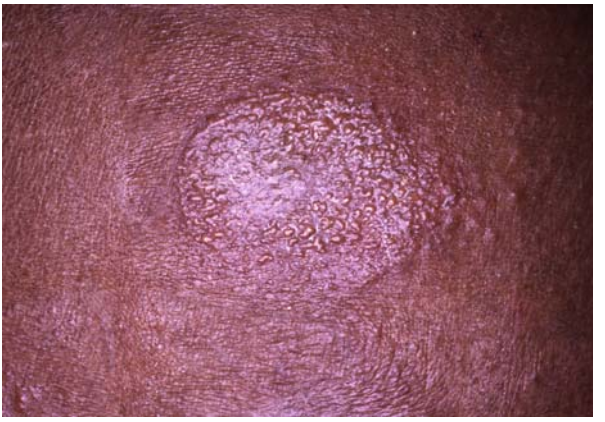

Fig. 3.11 Patch test scored ++ on a black skin. Darkening of the skin color replaces erythema. Infiltration and vesicles. Read at 72 h

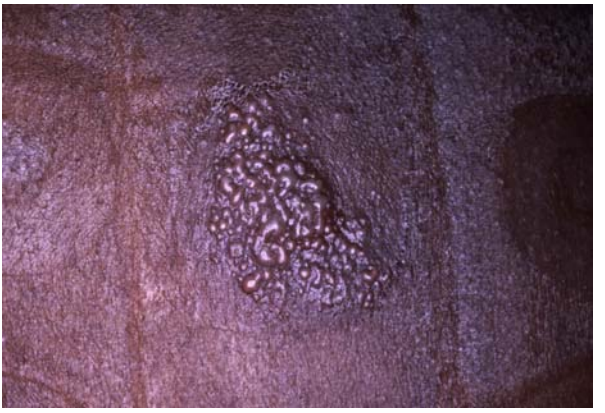

Fig. 3.12 Patch test scored ++; major infiltration of the central part, whitish tense vesicles mimicking minipustules. This particular image (*yellowish hue*) is due to the greater thickness of the stratum corneum in Blacks. The vesicles only burst as tension increases. Read at 48 h

Once again, there is little evidence of statistically significant differences in the irritant response between Black and Caucasian groups [68]. Therefore, it can be anticipated that patch test irritant reactions are not more frequent in Blacks than in Caucasoids.

Nevertheless, it is possible that intraindividual variations do exist, but further studies need to be conducted before a definitive statement can be made. Therefore, vigilance is requested at patch test reading to evaluate correctly potential irritant reactions.

3.16 Patch Testing Techniques in Different Climatic Environments

The patch testing procedures should be modified in different climatic conditions. This is because of the adherence of the tape and moisture of the skin surface under different climatic environments.

3.16.1 Temperate Climates

In some temperate countries, patch testing is performed only during the cooler seasons and discontinued during summer time, because the hot humid climate in summer may cause the tape to be dislodged more readily and patients generally find it uncomfortable to have strips of tape left on their skin for 48 h.

In many places, there is no real need to interrupt patch testing activities during summer time. The only reason why this habit does occur is for practical convenience, in relation with personnel holidays.

Useful information is related to seasonal variations in patch test reading in temperate countries.

- Chapping of the skin during winter predisposes to irritant contact dermatitis and also increases the incidence of false-positive reactions to substances such as formaldehyde, mercurials, and propyleneglycol.
- Some authors found many positive reactions in summer, but far fewer during cooler weather. Thus, occlusion and sweating may increase the number of positive reactions to some substances, whereas propyleneglycol, which is hygroscopic, and some other marginal irritants may often appear to be more of an irritant in winter.

3.16.2 Tropical Climates

Allergic contact dermatitis from whatever cause can be aggravated by environmental factors such as heat, high humidity, and dust [71].

In the tropics where there is little seasonable variation, there is no "ideal" season when patch testing can be done most comfortably. Patch testing is usually performed throughout the year. Because of the high ambient temperature and high humidity, the patch testing procedure may need some modification to ensure that the occlusive effects of the patch test chamber are maintained and that patients comply with the instructions carefully [72].

In addition, because of the higher ambient temperature, it is recommended that the patch test allergens be stored in a cool place when not in use. The test allergens should be kept in a refrigerator.

3.16.3 Patch Testing Procedures in the Tropics

The warm humid environment in the tropics makes patch testing an uncomfortable experience for the patients. Miliaria can occur at the sites of patch testing due to occlusion. Patients should be given clear instructions on the patch testing procedures.

3.16.3.1 Instructions for Patient

To ensure compliance, the following instructions may be given to the patients:

- Patients will be allowed to continue to take light showers or bathes to clean their face, chest, limbs, and lower torso. They should avoid washing the back (patch test sites) with water.
- The back where the patch test tapes are placed will be allowed to be cleaned daily with light moist towels, avoiding the test strips area.
- Patients should avoid outdoor activities and remain in a cool air-conditioned environment whenever possible.

3.16.3.2 Technical Adaptations

Patch testing can be performed with the various patch test chambers available commercially. The Finn chambers are widely used for patch testing in the tropics. However, the hot, humid environment causes sweating and makes plaster adhesion to the skin poor. Patch test plasters tend to come off easily. Reinforcement of the patch test plaster is useful to ensure proper occlusion. An effective way is to reinforce strips of plasters on the edges of the patch test tapes.

The conventional skin marker does not remain on the skin due to perspiration. The silver nitrate skin marker is a useful marker for identifying patch test sites.

3.17 Additional Note: Proposal for Modified Scoring Codes of Positive Patch Test Reactions

T. Menné, Editor-in-Chief, and I. White, Co-Editor of the journal "Contact Dermatitis," suggested a modification of the scoring codes to be submitted to the ESCD [73]. Their concern was based upon discrepancies in the reading of the + reaction encountered in the current literature.

Two schools have developed: one which defines the "+" reaction as homogeneous redness in the test area with scattered papules; the other requires homogeneous redness and homogeneous infiltration in the whole test area. The conflict is well-known. The stronger the patch test reaction, the higher the degree of relevance and reproducibility. Yet a weak positive reaction may be relevant and reproducible as well. The classification of patch test reactions depends exclusively on descriptive morphology. A pragmatic way, which will allow comparison between the different databases, is to introduce an extra grade of patch test reaction. To encompass the two main schools of current practice, the following scale is therefore suggested for debate:

+ homogeneous redness in the test area with scattered papules
++ homogeneous redness and homogeneous infiltration in the test area
+++ homogeneous redness and infiltration with vesicles
++++ homogeneous redness and infiltration with coalescing vesicles

To date, no real consensus has been reached in the matter.

References

1. Adams RM (1993) Profiles of greats in contact dermatitis. I Jozef Jadassohn (1863–1936). Am J Contact Dermatitis 4:58–59
2. Jadassohn J (1896) Zur Kenntnis der medikamentösen Dermatosen. Verhandlungen der Deutschen Dermatologischen Gesellschaft, Fünfter Congress, Graz, 1895. Wilhelm Braunmüller, Wien und Leipzig, pp 103–129
3. Gallant CJ (1994) Patch testing a century later. Chapter 4. In: Hogan DJ (ed) Occupational Skin Disorders, Ikagu-Shoin, New-York and Tokyo, pp 41–53
4. Wahlberg J, Lindberg M (2006) Diagnostic tests. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 365–390
5. Uter W, Schnuch A, Gefeller O (2006) Statistical methods in clinical epidemiology. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 150–163
6. Rietschel RL, Fowler JF Jr. (2008) Fisher's Contact Dermatitis, 6th edn. BC Decker, Hamilton, Ontario
7. Grob JJ, Sambuc R, Gouvernet J (1997) Introduction to epidemiology and prevention in dermatology. Chapter 1. In: Grob JJ, Stern RS, MacKie RM, Weinstock WA (eds) Epidemiology, Causes and Prevention of Skin Diseases, Blackwell Science, Oxford, p 6
8. Lachapelle JM, Douka MA (1985) An evaluation of the compatibility between aluminium Finn Chambers and various mercurials dissolved in water or dispersed in petrolatum. Dermatosen 33:12–14
9. Böhler-Sommeregger K, Lindemayr H (1986) Contact sensitivity to aluminium. Contact Dermatitis 15:278–281
10. Dooms-Goossens A (1982) Allergic contact dermatitis to ingredients used in topical applied pharmaceutical products and cosmetics. Katholieke Universiteit, Leuven, Belgium, Thesis
11. de Groot AC (2008) Patch testing. Test Concentrations and Vehicles for 4350 Chemicals, 3rd edn. acdegroot publishing, Schipsbootweg 5, NL 8351 HV Wapserveen
12. Patch Test Products Catalogue (2008) Chemotechnique Diagnostics
13. Benezra C, Andanson J, Chabeau G, Ducombs G, Foussereau J, Lachapelle JM, Lacroix M, Martin P (1978) Concentrations of patch test allergens: are we comparing the same things? Contact Dermatitis 4:103–105
14. Bruze M, Isaksson M, Gruvberger B, Frick-Engfeldt M (2007) Recommendation of appropriate amounts of petrolatum preparation to be applied at patch testing. Contact Dermatitis 56:281–285
15. Isaksson M, Gruvberger B, Frick- Engfeldt M, Bruze M (2007) Which test chambers should be used for acetone, ethanol and water solutions when patch testing? Contact Dermatitis 57:134–136
16. Sukanto H, Nater JP, Bleumink E (1981) Influence of topically applied corticosteroids on patch test reactions. Contact Dermatitis 7:180–185

17. O'Quinn SE, Isbell RH (1969) Effect of oral prednisone on eczema patch test reactions, Arch Dermatol 99:380–389
18. Anveden I, Lindberg M, Andersen KE et al. (2004) Oral prednisone suppresses allergic but not irritant patch test reactions in individuals hypersensitive to nickel. Contact Dermatitis 50:298–303
19. Motolese A, Ferdani G, Manzini BM, Seidenari S (1995) Echographic evaluation of patch test inhibition by oral antihistamine. Contact Dermatitis 32:251
20. Elston D, Licata A, Rudner E, Trotter K (2000) Pitfalls in patch testing. Am J Contact Dermat 11:184–188
21. Lachapelle JM, Naeyaert JM (2006) About the D.U.N. Survey. Dermatologie Actualité (Brussels) N 94:5–7
22. Sjövall P (1988) Ultraviolet radiation and allergic contact dermatitis. An experimental and clinical study. Thesis, University of Lund, Sweden
23. Lindelöf B, Lidén S, Lagerholml B (1985) The effect of grenz rays on the expression of allergic contact dermatitis in man. Scand J Immunol 21:463–469
24. Goossens A, Morren M (2006) Contact allergy in children. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 811–830
25. Vigan M (2006) La batterie standard de l'enfant. In: Progrès en Dermato-Allergologie, Toulouse 2006, John Libbey Eurotext, Montrouge (France) pp 15–25
26. Fischer T, Maibach HI (1986) Patch testing in allergic contact dermatitis: an update. Semin Dermatol 5:214–224
27. Wahlberg JE, Wahlberg ENG (1987) Quantification of skin blood flow at patch test sites. Contact Dermatitis 17:229–233
28. Uter WJC, Geier J, Schnuch A (1996) Good clinical practice in patch testing: readings beyond day 2 are necessary: a confirmatory analysis. Am J Contact Dermatitis 7:231–237
29. Saino M, Rinara P, Guarrera M (1995) Reading patch tests on day 7. Contact Dermatitis 32:312
30. Hellinckx K, Goossens A (2008) Late reactions to paraphenylenediamine are not always an indication of active sensitization: an example. Contact Dermatitis 58:110
31. Todd DJ, Handley J, Metwali M, Allen GE, Burrows D (1996) Day 4 is better than day 3 for a single patch test reading. Contact Dermatitis 34:402–404
32. Geier J, Gefeller O, Wiechmann K, Fuchs T (1999) Patch test reactions at D4, D5 and D6. Contact Dermatitis 40:119–126
33. Manuskiatti W, Maibach HI (1996) 1 versus 2– and 3-day diagnostic patch testing. Contact Dermatitis 35:197–200
34. Goh CL, Wong WK, Ng SK (1994) Comparison between 1-day and 2-day occlusion times in patch testing. Contact Dermatitis 31:48–49
35. Le Coz CJ, Muller B (2002) A practical sparkling and durable way to mark patch test sites. Contact Dermatitis 46(Suppl 4):552–553
36. Wilkinson DS, Fregert S, Magnusson B, Bandmann HJ, Calnan CD, Cronin E, Hjorth N, Maibach HI, Malten KE, Meneghini CL, Pirilä V (1970) Terminology of contact dermatitis. Acta Dermato-Venereologica 50:287
37. Uter W, Becker D, Schnuch A, Gefeller O, Frosch PJ (2007) The validity of rating patch test reactions based on digital images. Contact Dermatitis 57:337–342
38. Lachapelle JM, Tennstedt D, Fyad A, Masmoudi ML, Nouaigui H (1988) Ring-shaped positive patch test reactions to allergens in liquid vehicles. Contact Dermatitis 18:234–236
39. Fyad A, Masmoudi ML, Lachapelle JM (1987) The "edge effect" with patch test materials. Contact Dermatitis 16:147–151
40. Isaksson M, Brandao FM, Bruze M, Goossens A (2000) Recommendations to include budesonide and tixocortol pivalate in the European standard series. Contact Dermatitis 43:1–42

41. Lachapelle JM (1989) A left versus right side comparison study of Epiquick® patch test reactions in 100 consecutive patients. Contact Dermatitis 20:51–56
42. Foussereau J, Benezra C, Maibach HI (1982) Occupational contact dermatitis. Clinical and chemical aspects. Munksgaard, Copenhagen, pp 26–27
43. Bruynzeel DP, Maibach HI (1990) Excited skin syndrome and the hyporeactive state: current status. In: Menné T, Maibach HI (eds), Exogenous Dermatoses: Environmental Dermatitis. CRC Press, Boca Raton, pp 141–150
44. Kelett JK, Ring CH, Beck MH (1986) Compound allergy to medicaments. Contact Dermatitis 14:45–48
45. Bashir SJ, Maibach HI (1997) Compound allergy. An overview. Contact Dermatitis 36:179–183
46. Smeenk G, Kerckhoffs HP, Schreurs PH (1987) Contact allergy to a reaction product in Hirudoid cream: an example of compound allergy. Br J Dermatol 116:223–231
47. Marks JG Jr, Elsner P, DeLeo V (2002) Contact and Occupational Dermatology, 3rd edn. Mosby, St.Louis
48. Fregert S (1985) Publication of allergens. Contact Dermatitis 12:123–124
49. Le Coz CJ, Bottlaender A, Scrivener JN, Santinelli F, Cribier B, Heid E, Grosshans E (1998) Photocontact dermatitis from ketoprofen and tiaprofenic acid: cross-reactivity study in 12 consecutive patients. Contact Dermatitis 38:245–252
50. Pigatto PD, Bigardi A, Legori A, Valsecchi R, Picardo M (1996) Cross-reactions in patch testing and photopatchtesting with ketoprofen, tiaprofenic acid and cinnamic aldehyde. Am J Contact Dermat 7:220–223
51. Le Coz CJ, Ducombs G (2006) Plant and plant products. In: Frosch PJ, Menné T, Lepoittevin JP (eds). Contact Dermatitis, 4th edn. Springer, Berlin, pp 751–800
52. Lisi P, Brunelli L, Stingeni L (2003) Co-sensitivity between cobalt and other transition metals. Contact Dermatitis 48:172–173
53. Fregert S, Bandmann HJ (1975) Patch Testing. Published on behalf of the International Contact Dermatitis Research Group. Spinger-Verlag, Berlin
54. Carlsen BC, Andersen KE, Menné T, Johansen JD (2008) Patients with multiple contact allergies: a review. Contact Dermatitis 58:1–8
55. Schnuch A, Brasch J, Uter W (2008) Polysensitization and increased susceptibility in contact allergy: a review. Allergy 63:156–167
56. Rietschel RL, Fowler JF Jr (2001) Fisher's Contact Dermatitis. 5th edn. Lippincott Williams & Wilkins, Philadelphia, pp 13–18
57. Weiss G, Shemer A, Trau H (2002) The Koebner phenomenon: review of the literature. JEADV 16:241–248
58. Deleuran M, Clemmensen O, Andersen KE (2000) Contact lupus erythematosus. Contact Dermatitis 43:169–185
59. Bahillo-Monné C, Heras-Mendaza F, Casado-Farinas I, Gatica-Ortega M, Conde-Salazar L (2007) Jessner's lymphocytic infiltrate as a Koebner response to patch test. Contact Dermatitis 57:197–199
60. Björkner B, Bruze M, Dahlquist I, Fregert S, Gruvberger B, Persson K (1986) Contact allergy to the preservative Kathon® CG. Contact Dermatitis 14:85–90
61. Kanerva L, ESstlander T, Jolanki R (1988) Sensitization to patch test acrylates. Contact Dermatitis 18:10–15
62. Gawkrodger DJ, English JCS (2006) How safe is patch testing to PPD? Br J Dermatol 154:1025–1027
63. Hellinckx K, Goossens A (2008) Late reaction to *para*-phenylenediamine are not always an indication of active sensitization: an example. Contact Dermatitis 58:110
64. Mitchell JC (1975) The angry back syndrome. Eczema creates eczema. Contact Dermatitis 1:193–194

65. Maibach HI (1981) The ESS-excited skin syndrome (alias the “angry back”). In: Ring J, Burg G (eds). New Trends in Allergy. Springer, Berlin, pp 208–221
66. Mitchell JC, Maibach HI (1982) The angry back syndrome – the excited skin syndrome. Semin Dermatol 1:9
67. Bruynzeel DP, Maibach HI (1986) Excited skin syndrome (angry back). Arch Dermatol 122:323–328
68. Modjahedi SP, Maibach HI (2006) Ethnicity. In: Chew AL, Maibach HI (eds). Irritant Dermatitis. Springer, Berlin, pp 177–183
69. Nakayama H (2006) Pigmented contact dermatitis and chemical depigmentation. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 319–333
70. Weigand DA, Haygood C, Gaylor JR (1974) Cell layers and density of Negro and Caucasian stratum corneum. J Invest Dermatol 62:563–568
71. Okoro A, Rook AJ, Canizares O (1992) Eczemas in the tropics. Plant dermatitis, Chapter 23. In: Canizares O, Harman R (eds). Clinical Tropical Dermatology, 2nd edn. pp 449–481
72. Goh CL (2006) Irritant contact dermatitis on the tropics. In: Chew AL, Maibach HI (eds) Irritant Dermatitis. Springer, Berlin, pp 105–108
73. Menné T, White I (2008) Standardization in contact dermatitis. Contact Dermatitis 58:321

The Standard Series of Patch Tests

4

J.-M. Lachapelle

4.1 Historical Background

The use of a standard series in all tested patients was adopted worldwide in the 1980s. Formerly, many authors refused to adhere to its systematic use and championed the concept of "selected patch tests." Werner Jadassohn (at Geneva) had a strong influence on many colleagues in this respect. The principle of "choice" or "selection" was based on a careful recording of anamnestic data, especially in the field of occupational dermatology [1]. A similar view was shared in France by Foussereau [2]. Their opinion was that "testing systematically" with a standard series led unavoidably to a lazy clinical attitude. They argued that by doing so, clinicians were tempted to neglect the medical history of each individual patient.

Conversely, the standard series found enthusiastic defenders among renowned pioneers in the field of allergic contact dermatitis.

Bruno Bloch acted as a group leader for promoting and disseminating the idea of applying a limited standard series on each patient [3]. This was made in close connection with Jozef Jadassohn in Breslau (Bloch's former teacher when he was in Bern), Blumenthal and Jaffé in Berlin, and later Sulzberger in New York.

Poul Bonnevie, Professor of Occupational Medicine in Copenhagen, expanded Bloch's embryonic standard series of tests and published it in his famous textbook of environmental dermatology [4]. The list (21 allergens) can be considered as the prototype of the standard series of patch tests. Later, this list of allergens was modified and updated by the founding members of the ICDRG group. The changes were based on the experience of the members in their own countries and mirrored the findings and current situation in different parts of Europe and the United States.

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4.2 Advantages and Disadvantages of Using a Standard Series of Patch Tests

4.2.1 Advantages

- The standard series corresponds to an allergological check-up of each individual patient, as regards the most common allergens encountered in the environment. Positive and negative patch test results map out the allergological profile of the patient.
- The standard series compensates for anamnestic failures. Even when the clinician tries to record carefully the history of each individual patient, he may omit important events in some cases, despite using a detailed standardized questionnaire. Positive patch test results lead the clinician to ask some additional (retrospective) questions.
- The systematic use of the standard series permits to conduct comparative studies in different countries, thus increasing our knowledge in terms of geographic variations.

4.2.2 Disadvantages

- The standard series can produce a "sleeping" effect on the clinician's attitude. This perverse result is avoided when the standard series is considered as a limited technical tool, representing one of the pieces of a puzzle, to be combined with other means of diagnosis. The general principle to be kept in mind is that the standard series cannot replace a detailed anamnestic (and catamnestic) investigation.
- Theoretically, application of the standard series could induce an active sensitization to some allergens (see Sect. 3.14.1). Common examples are *p*-phenylenediamine, primin, or isothiazolinone [5]. The risk, however, is extremely low when testing is performed according to internationally accepted guidelines.

In conclusion, taking into account all these considerations, it must be emphasized that the overall risk-benefit equation of patch testing patients with the allergens of the standard series is much in favor of the benefit [5].

4.3 The Three Major Standard Series Used Throughout the World

There is no unanimity worldwide as regards the contents of a standard series. There are three major options in building a standard series, in relation with regional potential variations:

1. The revised 2008 European standard series, on behalf of the European Society of Contact Dermatitis (ESCD) and the European Environmental and Contact Dermatitis Research Group (EECDRG) [6].

2. The revised 2008 North American standard series according to the North American Contact Dermatitis Research Group (NACDG) [7].
3. The revised 2008 Japanese standard series according to the Japanese Society for Contact Dermatitis (JCDS) [8].

A comparison of the three lists (Table 4.1) suggests that 40 allergens are potentially considered in the international standardization process.

The discrepancies in comparing lists (a), (b), and (c) are due to two main factors:

1. There are regional variations, related to either the natural occurrence of allergens (e.g., urushiol) or to a significant variability in the use of some allergens in various regions, due to different medical, cosmetic, industrial, or environmental habits.
2. A different approach of the three research groups (or societies) regarding each individual allergen, thus reaching dissimilar conclusions. The three groups are working independently and have not shared their opinions so as to reach a worldwide consensus.

Most decisions reached by each group are partly based on multicenter studies and/or thorough literature reviews.

4.4 Some Remarks About the "Mixes" of the Standard Series

Using mixes instead of single allergens saves time and space. In this respect, patients are tested with a number of closely related substances. The screening capacity of the standard series is thereby greatly increased. Nevertheless, the value of these mixes is sometimes questioned. It is difficult to find an optimal concentration for each allergen in a common vehicle (usually petrolatum) and to determine whether the allergens metabolize or interact to potentiate or quench a reaction [9].

It is recommended that patients positive for a mix be retested with the individual ingredients. Infrequently, the latter results are negative, and in that case it is questioned whether the initial reaction was an expression of irritancy or whether the ingredients have interacted. The opposite has also been noticed. The patient may be negative to a particular mix, but reacts when retested with its ingredients.

The composition of the various mixes of the standard series is detailed in Table 4.2 [6].

4.5 Proposal for an ICDRG Revised International Series of Patch Tests

Considering the current status of the standard series throughout the world, the members of the ICDRG group discussed the possibility of using a shortened list of common allergens, which could be used internationally as a "minimal international standard series" [10].

Table 4.1 Comparative lists of allergens in three different standard series

	Compound	ECDS and EECDRG[a] (%)	NACDG[b] (%)	JCDS[c](%)
1.	Potassium dichromate	0.5	0.25	0.5
2.	Neomycin sulfate	20	20	20
3.	Thiuram mix	1	1	1.25
4.	*p*-Phenylenediamine base	1	1	1
5.	Cobalt chloride ($CoCl_2$ $6H_2O$)	1	–	1
6.	Benzocaine	5	5	–
7.	Formaldehyde	1(aq)	1(aq)	1(aq)
8.	Colophony	20	20	20
9.	Clioquinol	5	–	–
10.	*Myroxylon Pereirae* (Balsams of Peru)	25	25	25
11.	*N*-Isopropyl-*N*-phenyl paraphenylen-ediamine (IPPD)	0.1	–	–
12.	Wool (lanolin) alcohols	30	30	30
13.	Mercapto mix	2	1	2
14.	Epoxy resin	1	1	1
15.	Paraben mix	16	12	15
16.	*p-tert*-Butylphenol formaldehyde resin (PTBP resin)	1	1	1
17.	Fragrance mix 1	8	8	8
18.	Quaternium 15	1	2	–
19.	Nickel sulfate ($NiSo_4$ $6H_2O$)	5	2.5	2.5
20.	Cl + Me-isothiazolinone	0.01(aq)	0.01(aq)	0.01 (aq)
21.	Mercaptobenzothiazole	2	1	–
22.	Sesquiterpene lactone mix	0.1	0.1	0.1
23.	Budesonide	0.01	0.1	
24.	Tixocortol pivalate	0.1	1	–
25.	Methyldibromoglutaronitrile	0.5	2(+phenox-yethanol)	–
26.	Fragrance mix 2	14	14	–
27.	Hydroxyisohexyl 3-cyclohexene carboxaldehyde (Lyral)	5.0	–	–
28.	Primin	0.01	–	0.01
29.	Imidazolidinyl urea	–	2	–
30.	Cinnamic aldehyde	–	1	–
31.	Carba mix	–	3	–
32.	Ethylenediamine dihydrochloride	–	1	1
33.	Black Rubber mix	–	0.6	0.6
34.	Gold sodium thiosulfate	–		0.5
35.	Caine mix	–	–	7
36.	Dithiocarbamate mix	–	–	2
37.	Urushiol	–	–	0.002
38.	Thimerosal (thiomersal)	–	–	0.05

(continued)

Table 4.1 (continued)

	Compound	ECDS and EECDRG[a] (%)	NACDG[b] (%)	JCDS[c](%)
39.	Mercuric chloride	–	–	0.05(aq)
40.	Petrolatum	–	–	as is

[a]The revised 2008 European standard series. The concentrations quoted refer to petrolatum except where otherwise stated
[b]The revised 2008 North American standard series. The concentrations quoted refer to petrolatum except where otherwise stated
[c]The revised 2008 Japanese standard series. The concentrations quoted refer to petrolatum except where otherwise stated

Table 4.2 The composition of the mixes of the European Standard Series

Thiuram mix	1% pet
Dipentamethylenethiuram disulfide (0.25%)	
Tetramethylthiuram disulfide (0.25%)	
Tetraethylthiuram disulfide (0.25%)	
Tetramethylthiuram monosulfide (0.25%)	
Mercapto mix	2% pet.
N-Cyclohexylbenzothiazyl sulphenamide (0.5%)	
Dibenzothiazyl disulfide (0.5%)	
Mercaptobenzothiazole (0.5%)	
Morpholinylmercaptobenzothiazole (0.5%)	
Fragrance mix 1 (incl. 5% sorbitan sesquioleate as emulsifier)	8% pet.
α-Amylcinnamaldehyde (1%)	
Cinnamic aldehyde (1%)	
Cinnamyl alcohol (1%)	
Eugenol (1%)	
Geraniol (1%)	
Hydroxycitronellal (1%)	
Isoeugenol (1%)	
Evernia Prunastri (oakmoss absolute) (1%)	
Fragrance mix 2	14% pet.
a-Hexyl cinnamaldehyde (5%)	
Citral (1%)	
Citronellol (0.5%)	
Farnesol (2.5%)	
Coumarin (2.5%)	
Hydroxyisohexyl 3-cyclohexene carboxaldehyde (2,5%)	
Paraben mix	16% pet.
Methyl-4-hydroxybenzoate (4%)	
Ethyl-4-hydroxybenzoate (4%)	
Propyl-4-hydroxybenzoate (4%)	
Butyl-4-hydroxybenzoate (4%)	
Sesquiterpene lactone mix	0.1% pet.
Alantolactone (0.033%)	
Dehydrocostus lactone and costunolide (0.067%)	

The list is primarily aimed to help dermatologists working in countries where patch testing is not commonly performed for different reasons, mainly related to the limited availability or cost of allergens. It is of course flexible and can be adapted, taking into account recent advances in epidemiological studies conducted in ACD patients [11].

Table 4.3 shows allergens that have been considered as eligible candidates for such a list.

Table 4.3 Proposed allergens for a modified "shortened" international standard series (concentrations refer to petrolatum unless otherwise stated)

		%
1.	Potassium dichromate	0.5
2.	Neomycin sulfate	20
3.	Thiuram mix[a]	1
4.	*p*-Phenylenediamine base[b]	1
5.	Formaldehyde	1 (aq)
6.	Colophony	20
7.	*Myroxylon pereirae* (Balsams of Peru)	25
8.	Wool (lanolin) alcohols	30
9.	Mercapto mix	2
10.	Epoxy resin	1
11.	*p-tert*-Butylphenol formaldehyde resin (PTBP resin)	1
12.	Fragrance mix 1[c]	8
13.	Nickel sulfate ($NiSO_4$ $6H_20$)	2.5
14.	Mercaptobenzothiazole (MBT)[d]	1
15.	Budesonide[e]	0.01
16.	Quaternium 15[f]	2
17.	Cl + Me-isothiazolinone[g]	0.01 (aq)
18.	Imidazolidinyl urea	2
19.	Tixocortol pivalate	0.1
20.	Fragrance mix 2[h]	14

[a]Thiuram mix lacks high specificity and sensitivity
[b]Some cases of hair dye dermatitis could be missed with the use of PPD alone
[c]If positive, breakdown is needed
[d]MBT can identify cases of allergic contact dermatitis negative to mercapto mix, and vice versa
[e]Budesonide is highly recommended in an international standard series, since it is considered an important marker for corticosteroid allergy [12]
[f]It is an important allergen in the United Kingdom, while it is not used in Japan
[g]Mainly used in Japan for cosmetics. It remains important worldwide, since it is used as a preservative in many industrial products
[h]If positive, breakdown is needed

4.6 List of Allergens Proposed for an Extended ICDRG Series, Which May be Required According to Each Individual Situation

Some allergens present in one (or more) of the three lists of Table 4.1 are not considered eligible candidates for the revised international standard series presented in Table 4.3.

On the other hand, they are listed in an "extended series" (Table 4.4). Other allergens are also proposed in the extended series, as they are considered useful in the literature.

4.7 List of Allergens Proposed to Be Deleted from the Revised and Extended ICDRG Series

Some of the allergens recorded in Table 4.1 lack general interest for different reasons. Therefore, they are not proposed as candidates for an extended international series. Nevertheless, they could be used in specific circumstances.

Table 4.4 Proposed allergens for an extended international standard series (concentrations refer to petrolatum unless otherwise stated)

		%
A. Allergens present in one (or more) of the three lists of Table 4.1		
1.	Cobalt chloride ($CoCl_2$ $6H_20$)[a]	1
2.	Benzocaïne	5
3.	Clioquinol	5
4.	Paraben mix	16
5.	Primin	0.01
6.	Ethylenediamine dihydrochloride	1
7.	Urushiol	0.002
8.	Thimerosal (thiomersal)	0.05
9.	Sesquiterpene lactone mix	0.1
B. Additional useful allergens		
10.	Hydrocortisone 17-butyrate	1 (alc)
11.	2,5-Diazolidinylurea[b]	2
12.	Cetylstearylalcohol	20
13.	Toluenesulphonamide formaldehyde resin	10
14.	Propylene glycol	30 (aq)
15	Disperse Blue mix (124/106)	1

[a]Cobalt is not traced as relevant in many cases. Petechial reactions should not be read as positive
[b]It is not used in Japan

The allergens dispersed in petrolatum are the following: *N*-isopropyl-*N* -phenylparaphenylenediamine (IPPD) 0.1%; cinnamic aldehyde 1%; carba mix 3% (often lacks clinical relevance); black rubber (PPD) mix 0.6%; dithiocarbamate mix 2%; and ammoniated mercuric chloride 1%.

4.8 Succinct Information about Allergens

Some basic information about allergens proposed for an ICDRG revised international series of patch tests (see Sect. 4.5) as well as for an extended ICDRG series (see Sect. 4.6) are given here. More details are available in the textbooks of contact dermatitis listed in the general references section at the end of this book. We have illustrated in Fig. 4.1 numerous positive patch test reactions in a multisensitized patient.

4.8.1 Allergens Listed in Sect. 4.5

1. *Potassium dichromate*: Hexavalent form of chromium. Present in cement, tanning of leather, textile dyes, wood preservatives, alloys in metallurgy, safety matches, photography, electroplating, anticorrosives, ceramics, tattoos, paints, glues, pigments, detergents, and other materials. Spot Test: diphenylcarbazide (see Sect. 7.7.2).
2. *Neomycin sulfate*: Broad-spectrum antibiotic in topical creams, powders, ointments, eye and ear drops. Growth promoter in veterinary use.
3. *Thiuram mix*: Mixture of thiurams, used as rubber accelerators and vulcanizers, fungicides, disinfectants for seed, animal repellents, etc. (see Table 4.2).
4. *p-Phenylenediamine (PPD)*: Primary intermediate in permanent hair dyes and fur dyes. Also used in photographic developers, lithography, photocopying, oils, greases, gasoline,

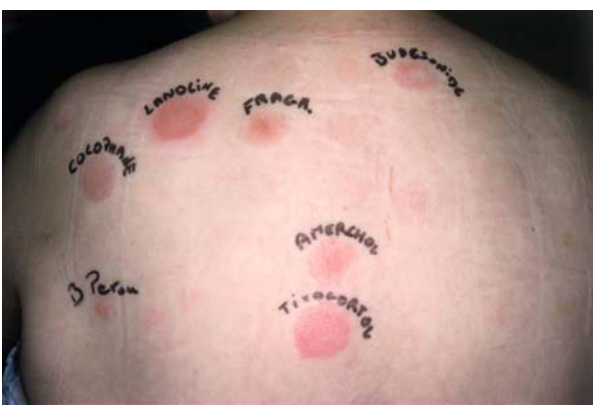

Fig. 4.1 Multisensitized patient. Multiple positive allergic patch test reactions

and as antioxidant/accelerator in the rubber and plastic industries. PPD may be present at high concentration in henna tattoos [13].

5. *Formaldehyde*: Ubiquitous allergen. Used as astringent, disinfectant, preservative in cosmetics, metalworking fluids, shampoos, etc. Widespread use in several industrial procedures. There are many formaldehyde releasers. Spot Test: chromotropic acid (see Sect. 7.7.2).
6. *Colophony*: Yellow resin in the production of varnishes, printing inks, paper, soldering fluxes, cutting fluids, glue tackifiers, adhesives, surface coatings, polish, waxes, cosmetics, topical medicaments, etc. Modified colophony used in hydrocolloid wound dressings is also allergenic [14].
7. *Myroxylon pereirae (Balsams of Peru)*: Flavor in tobacco, drinks, pastries, cakes, wines, liquors, and spices. Fixative and fragrance in perfumery; in topical medicaments, dentistry, etc.
8. *Wool alcohols*: Different types of alcohols (aliphatic, steroid, triterpenoid) present in wool fat (lanolin). As ointment base in cosmetic and pharmaceutical products. Amerchol L101 is another marker of lanolin allergy. It contains lanolin alcohols obtained from the hydrolysis of lanolin.
9. *Mercapto mix*: Mixture of mercaptothiazoles (see mercaptobenzothiazole and Table 4.2).
10. *Epoxy resin*: Resin based on epichlorhydrin and bisphenol A for use in adhesives, surface coatings, electrical insulation, plasticizers, polymer stabilizers, laminates, surface coatings, paints and inks, product finishers, PVC products, and vinyl gloves. Oligomers may vary in molecular weight from 340 and higher. The higher the molecular weight, the less sensitizing the compound.
11. *p-tert-Butylphenol formaldehyde (PTBF resin)*: Resin used in adhesives for shoes and watch straps and for many other uses in various industrial procedures.
12. *Fragrance mix 1*: Fragrance mix 1 is an invaluable tool to detect some (but not all!) contact allergies to perfumes, scented cosmetics, and detergents. It was developed by Larsen [15] as a mixture of eight ingredients. Its interest was confirmed by several studies, but its limitations were obvious due to the countless ingredients present in some perfumes. It was implemented recently by an additional fragrance mix, called "fragrance mix 2."
13. *Nickel sulfate*: Nickel metal: a common allergen present in various alloys, electroplated metal, earrings, watches, buttons, zippers, rings, utensils, tools, instruments, batteries, machinery parts, working solutions of metal cutting fluids, nickel plating for alloys, coins, pigments, orthopedic plates, keys, scissors, razors, spectacle frames, kitchenware, etc. The release of nickel by coins is well-documented [16, 17]. Spot Test: dimethylglyoxime (see Sect. 7.7.2).
14. *Mercaptobenzothiazole*: Accelerator, retarder, and peptizer for natural and other rubber products. Fungicide, Corrosion inhibitor in soluble cutting oils and antifreeze mixtures. Also used in many other industrial procedures.
15. *Budesonide*: Non-halogenated corticosteroid for use in topical preparations and for the treatment of rhinitis and asthma. Belongs to the group B (triamcinolone acetonide) type of corticosteroids. One of the markers of corticosteroid allergy (see Appendix, Table A.3).

16. *Quaternium 15*: Formaldehyde-releaser used chiefly as a cosmetic preservative. Also in widespread usage in industry and household products. Marketed under different trade names.
17. *Cl + Me-isothiazolinone*: Used as a preservative in oils and cooling fluids, soaps, latex emulsions, slime control in paper mills, jet fluids, printing inks, detergents, shampoos, hair conditioners, and bubble baths. Also known under the trade name Kathon CG. Many other trade names are indexed.
18. *Imidazolidinyl urea*: Formaldehyde-releaser used as a cosmetic preservative (lotions, creams, hair conditioners, shampoos, deodorants) and also in topical drugs. Also known under the trade name Germall 115 (not exclusive).
19. *Tixocortol pivalate*: Topical corticosteroid belonging to the group A (hydrocortisone) type of steroids used in nasal sprays for the treatment of rhinitis. Good marker for group A corticosteroid contact allergy (see Appendix, Table A.3).
20. *Fragrance mix 2*: Fragrance mix 2 was developed in Europe [18] as a mixture of six ingredients. It was demonstrated to be a useful additional marker of fragrance allergy, particularly in cases of allergic contact dermatitis "missed" by fragrance mix 1 [18]. It is recommended for inclusion in the standard series [6].

4.8.2 Allergens Listed in Sect. 4.6

1. *Cobalt chloride*: Component in paints for glass and porcelain. Siccative in paints. Present in various alloys. Concomitant sensitization (see Sect. 3.13.2) occurs with nickel and chromates.
2. *Benzocaïne*: Topical anesthetic used in many over-the-counter preparations and topical drugs.
3. *Clioquinol*: Synthetic anti-infective (antibacterial and to a lesser extent antifungal) agent. Present in topical drugs (i.e., Vioform). Occasionally used as a systemic drug.
4. *Paraben mix*: Mixture of parabens (esters of parahydroxybenzoic acid) very widely used as preservatives in foods, drugs, and cosmetics (see Table 4.2).
5. *Primin*: Primin (or 2-methoxy-6 pentylbenzoquinone) is the major allergen in Primula dermatitis.
6. *Ethylenediamine dihydrochloride*: Stabilizer in some steroid creams and rubber latex. Inhibitor in antifreeze solutions and cooling fluids. Component in aminophylline. One of the allergens in Mycolog cream.
7. *Urushiol*: Oleoresin of the sap of the *Toxicodendron* plants. It contains catechols, which are the sensitizing chemicals. A very useful allergen in some parts of the world: the United States (poison ivy/oak dermatitis), South America (*Lithrea* dermatitis), and Eastern Asia, mainly Japan and China (lacquer's tree dermatitis).
8. *Thimerosal (thiomersal)*: Organic mercury salt used as a disinfectant and as a preservative agent, but less commonly than previously, especially in contact lens fluids, eyedrops, and vaccines.
9. *Sesquiterpene lactone mix*: A mixture of three sesquiterpene lactones: alantolactone, dehydrocostus lactone, and costunolide; contact allergens present in Compositae plants (Syn. Asteraceae), which constitute one of the largest plant families in the world.

10. *Hydrocortisone-17-butyrate*: Used as a topical corticosteroid with anti-inflammatory properties. Marker for some cases of topical corticosteroid allergy (see Appendix, Table A.3).
11. *2,5-Diazolidinylurea*: Formaldehyde-releaser used as a cosmetic preservative in, for example, lotions, creams, shampoos, and hair gels. Known also under the trade name Germall II.
12. *Cetylstearylalcohol*: A combination of cetyl (C16) and stearyl (C18) alcohols 50/50 used as emulsifier and emollient in cosmetic lotions, creams, ointments, and pharmaceutical preparations.
13. *Toluenesulphonamide formaldehyde resin*: Modifier and adhesion promoter for film natural and synthetic resins. Occurs in vinyl lacquers, nitrocellulose compositions (e.g., nail lacquers), PVA adhesives, acrylics.
14. *Propylene glycol*: Vehicle in pharmaceutical and cosmetic bases. In food as solvent for colors and flavors and to prevent growth of moulds. Present in cooling fluids.
15. *Disperse Blue mix*: Disperse Blue mix is a mixture of two disperse dyes (partially soluble in water): Disperse Blue 106 and Disperse Blue 124. These dyes are chiefly used in the textile industry to color synthetic fibers such as polyester, acrylic, and acetate, and sometimes nylon. They are not used for natural fibers. When suspecting textile contact dermatitis, Disperse Blue mix is considered a good marker, but investigation has to be completed by the Textile Colors and Finish Series.

4.9 Additional Series of Patch Tests

The (extended) standard series of patch tests has some limitations. Cohorts of allergens are present in our environment. In each patient, additional allergens have to be considered according to the personal history; it is sometimes needed to test with unknown products (see Sect. 7.5). To improve the performance of the patch testing procedure, several groups of research have proposed additional series of patch tests, suitable in well-defined environmental and/or work exposures. Such series are available from companies (see Sect. 3.4.1). The clinician has to adapt his (her) choice to each individual patient [19]. Additional series of patch tests are presented in Appendix, in alphabetical order.

References

1. Jadassohn W (1951) A propos des tests épicutanés “dirigés” dans l’eczéma professionnel. Praxis 40:1–4
2. Foussereau J, Benezra C (1970) Les eczémas allergiques professionnels. Masson, Paris
3. Bloch B (1929) The role of idiosyncrasy and allergy in dermatology. Arch Dermatol Syphilis 19:175–197
4. Bonnevie P (1939) Aetiologie und Pathogenese der Ekzemkrankheiten. Klinische Studien über die Ursachen der Ekzeme unter besonderer Berüksichtigung des Diagnostischen Wertes der Ekzemproben. Busch, Copenhagen/Barth, Leipzig

5. Wahlberg JE, Lindberg M (2006) Patch testing. In: Frosch PJ, Menné T, Lepoittevin JP (eds). Contact Dermatitis, 4th edn. Springer, Berlin, pp 365–390
6. Bruze M, Andersen KE, Goossens A. on behalf of the ESCD and EECDRG (2008) Recommendation to include fragrance mix 2 and hydroxyisohexyl 3-cyclohexene carboxaldehyde (Lyral) in the European baseline patch test series. Contact Dermatitis 58:129–133
7. Sasseville D (2008) Personal communication, on behalf of the NACDG
8. Matsunaga K (2008) Personal communication, on behalf of the JCDS
9. Hansson C, Agrup G (1993) Stability of the mercaptobenzothiazole compounds. Contact Dermatitis 28:29–34
10. Lachapelle JM, Ale SI, Freeman S, Frosch PJ, Goh CL, Hannuksela M, Hayakawa R, Maibach HI, Wahlberg JE (1997) Proposal for a revised international standard series of patch tests. Contact Dermatitis 36:121–123
11. The ESSCA Working Group (2008) The European Surveillance System of Contact Allergies (ESSCA): results of patch testing the standard series, 2004. JEADV 22:174–181
12. Dooms-Goossens A, Andersen KE, Brandao FM, Bruynzeel D, Burrows D, Camarasa J, Ducombs G, Frosch P, Hannuksela M, Lachapelle JM, Lahti A, Menné T, Wahlberg JE, Wilkinson JD (1996) Corticosteroid contact allergy: EECDRG Multicentre Study. Contact Dermatitis 35:40–44
13. Brancaccio RR, Brown LH, Chang YT, Fogelman JP, Mafong EA, Cohen DE (2002) Identification and quantification of para-phenylenediamine in a temporary black henna tattoo. Am J Contact Dermat 13:15–18
14. Pereira TM, Flour M, Goossens A (2007) Allergic contact dermatitis from modified colophonium in wound dressings. Contact Dermatitis 56:5–9
15. Larsen W, Nakayama H, Lindberg M, Fischer T, Elsner P, Burrows D, Jordan W, Shaw S, Wilkinson J, Marks J Jr, Sugawara M, Nethercott J (1996) Fragrance contact dermatitis: a worldwide multicenter investigation. Part I. Am J Contact Dermatitis 7:77–83
16. Lachapelle JM, Marot L (2004) High nickel release from 1- and 2- euro coins: are there practical implications? Dermatology 209:288–290
17. Lidén C, Skare L, Vahter M (2008) Release of nickel from coins and deposition onto skin from coin handling – comparing euro coins and SEK. Contact Dermatitis 59:31–37
18. Frosch PJ, Pirker C, Rastogi SC, Andersen KE, Bruze M, Svedman C, Goossens A, White IR, Uter W, Arnau EG, Lepoittevin JP, Menné T, Johanson JD (2005) Patch testing with a new fragrance mix detects additional patients sensitive to perfumes and missed by the current fragrance mix. Contact Dermatitis 52:207–215
19. de Groot AC, Frosch PJ (2006) Patch test concentrations and vehicles for testing contact allergens. In: Frosch PJ, Menné T, Lepoittevin JP (eds). Contact Dermatitis, 4th edn. Springer, Berlin pp 907–928

Photopatch Testing

5

J.-M. Lachapelle and A. Goossens

5.1 Definition and Aims

Photopatch testing (PPT), simply stated, is patch testing with the addition of UV radiation to induce formation of the photoallergen. Application of allergens and scoring criteria are the same as those described for plain patch testing (see Chap. 3). The only additional equipment that is necessary is an appropriate light source and opaque shielding for the period after removal of the patch test material before readings [1].

PPT is intended to detect the responsible photoallergen(s) in two clinical situations, namely photoallergic contact dermatitis and photoallergic drug eruptions. Nevertheless, these two conditions cannot always be easily diagnosed from other dermatoses, induced and/or worsened by exposure to light, that is, chronic actinic dermatitis (CAD), polymorphic light eruption (PLE) and other variants of photosensitivity. Therefore, some authors recommend that all photosensitive patients should be photopatch tested [1]. Photoallergic contact dermatitis (PACD) can in fact be superimposed on PLE.

The strategies for assessing the relevance of positive photopatch testing results are similar to those used for plain patch testing (see Chap. 8).

5.2 Photoallergic Contact Dermatitis

PACD is produced when sensitization occurs from the combination of skin contact with a compound together with ultraviolet light (UVL) exposure. In these cases, the hapten requires UVL to be fully activated. Such patients develop dermatitis on light-exposed sites. This typically involves the face, neck, dorsal hands, and forearms, but spares shaded sites such as the upper eyelids, submental area, and post-auricular areas (Fig. 5.1).

However, PACD has become less common because of the withdrawal from the market of many photocontact sensitizers. In the past 30 years, several notorious photoallergens were identified. Musk ambrette and six methyl coumarin were found to be potent photosensitizers present in fragrances. Their use has now been banned by the International Fragrance

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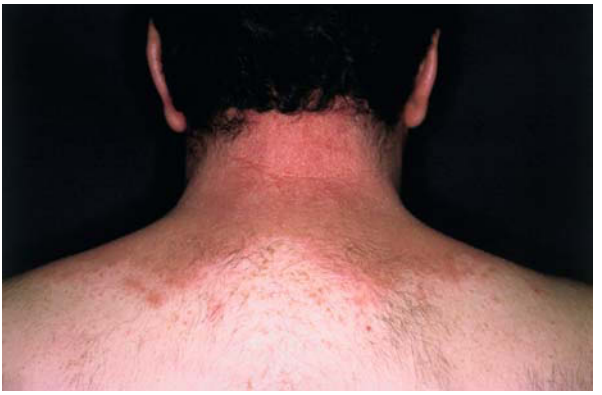

Fig. 5.1 Photoallergic contact dermatitis to a sunscreen. Covered sites are spared

Association (IFRA). Halogenated salicylanilides and chlorinated phenols, e.g., bithionol, fenticlor, and tribromosalicylanilide (TBS) were popular antiseptic and antifungal agents. These have also been withdrawn. However, it is always possible that these photoallergens may creep in from unregulated sources. They were particularly troublesome in the past as they were capable of producing persistent light reactions (PLR). In such cases the patient continued to react to UVL even after withdrawal of the contact allergen.

PLR (chronic actinic dermatitis, actinic reticuloid) is an idiopathic, severe, chronic photodermatosis, which occurs most often in men, middle-aged, or older (Fig. 5.2). It is characterized by infiltrated, erythematous, shiny plaques on an eczematous background on exposed areas, often with involvement of covered sites. The patients react to UVA, UVB, and visible light. Contact dermatitis plays a major role.

However, with the ever-increasing number of new products coming on the market, there is always the possibility of the appearance of new photoallergens. An important example is the increasing use of sunscreens, which are now often incorporated into cosmetic products where their use may not be so obvious. All the sunscreen chemicals that absorb UVL are capable of producing PACD. These include the *p*-aminobenzoic acid (PABA) products, the cinnamates, the benzophenones, oxybenzones, and dibenzoyl methanes (Table 5.1). The reflectant sunscreens that act as a physical barrier are not photosensitizers (i.e., zinc oxide and titanium dioxide). Sunscreens are now the most common photocontact allergens seen [2]. However, the benefits of sunscreens still greatly outweigh the risks. Sunscreens form the basis of any PPT series (Table 5.1).

Another example of a photocontact allergen identified in recent years is olaquindox [3]. This is a chemotherapeutic growth-promotor used in food for pigs. It was marketed in 1975 as a 10% premix with vitamins and minerals. It forms a dusty mixture to which pig farmers are easily exposed when they add it to their pig's food. As the work is usually outdoors, it can be a potent photocontact allergen for these pig farmers. It can also produce persistent light reactors. Withdrawal of olaquindox and its substitution by an alternative growth promoter has been recommended and has already been instituted in some countries.

The non-steroidal anti-inflammatory drugs (NSAIDs) are increasingly used as topical preparations. These, too, are another reported source of PACD, as well as of allergic contact

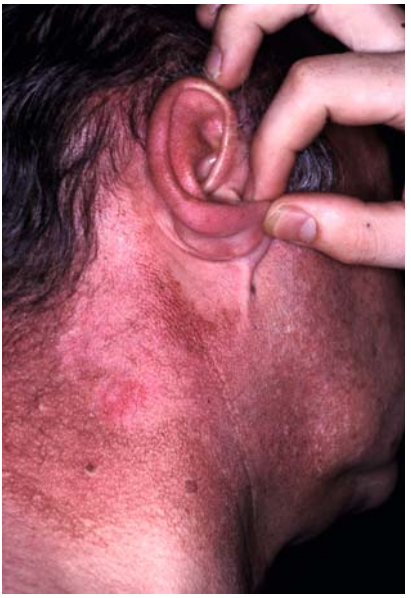

Fig. 5.2 Chronic actinic dermatitis. Large, variably oedematous and extremely pruritic erythematous plaque over the exposed parts of the face and neck. The retroauricular region is spared

Table 5.1 Criteria of differential diagnosis between photoallergic contact dermatitis and airborne allergic contact dermatitis

	Photoallergic contact dermatitis	Airborne allergic contact dermatitis
Clinical signs	Acute dermatitis	Acute dermatitis (most often)
	Affecting the whole face and neck	Affecting the whole face and neck
	Sparing to some extent the so-called shadow areas	Not sparing the so-called shadow areas
	i.e., eyelids	i.e., eyelids (oedematous)
	retroauricular folds	retroauricular folds
	V-shaped area of the anterior aspect of the neck	V-shaped area of the anterior aspect of the neck
Patch testing	Conventional patch tests are negative	Some of the conventional patch tests to suspected allergens are positive
Photopatch testing	Some photopatch tests are positive	Photopatch tests are negative, but some positive patch test reactions can be worsened by UV-light (when photopatch tested)

dermatitis and drug photosensitivity [4]. Since many of these compounds may also been used systemically, the possibility of development of systemic (photo-or non-photo) contact dermatitis, in patients topically sensitized, must always be borne in mind. Among NSAIDs, ketoprofen is of prime importance. In a recent study [4], 42 patients were investigated; 38

showed PACD, 1 photoaggravated reaction, and 3 ACD to ketoprofen. One-third of the patients reported PLR. Simultaneous PACD were frequently observed not only to structurally related but also to nonstructurally related NSAIDs and sunscreens. The authors conclude that routine PPT with ketoprofen might be indicated.

It must be emphasized that in CAD there are often many positive patch tests (including the compositae plants) and they are usually of doubtful relevance. There is no convincing evidence that the compositae plants are photoallergens, although they may produce an airborne dermatitis distinct from a photosensitive dermatitis.

However, once again, when the history and the physical examination suggest the possibility of PACD, PPT can in fact be superimposed on an endogenous photosensitivity such as PLE.

5.3 Photoallergic Contact Dermatitis vs. Airborne Allergic Contact Dermatitis: Criteria for Differential Diagnosis

Differential diagnosis between PACD and airborne allergic contact dermatitis can be difficult in clinical practice, mainly when lesions occur on the face and neck (see Sect. 2.2). The approach of such cases requires detailed information about the onset of the disease, thorough checking of the environment, careful examination, and extensive patch testing and photopatch testing investigation.

Criteria for differential diagnosis are summarized in Table 5.1.

5.4 Photoallergic Drug Eruptions

As explained elsewhere (see Chap. 12), the use of patch tests in some varieties of drug eruptions has been expanded in recent years and more experience has been gained in the field. This also applies to PPT in PACD. Similar principles of caution when interpreting positive and negative PPT results can be used in this respect. The main drugs for which a positive PPT has been observed are the following: phenothiazines, NSAIDs, thiazides, fluoroquinolones, captopril, fenofibrate, thioureas, etc.

5.5 Photopatch Testing Methodology

The methodology of PPT was first standardized in 1982 by the Scandinavian Photodermatology Research Group [5]. A European Taskforce for PPT was created in 2002 [6]. A panel representing Contact Dermatology/Photobiology/Photophysics with a special interest in PPT (on behalf of the European Society of Contact Dermatitis and European Photodermatology Society) met in Amsterdam. They came together to discuss and, if feasible,

to establish a consensus methodology, a list of recommended test chemicals, and interpretation guidelines for PPT [6].

The following recommendations were proposed:

- Allergens are applied to the upper back in duplicate and covered by an opaque material. In addition to the photoallergens series (Table 5.1), any products that the patient uses on exposed sites, or is exposed to, should also be applied in duplicate. The Finn Chamber is the proposed Patch Test Unit.
- One set is removed after 24 h (or preferably 48 h) and irradiated with 5 J cm^{-2} of ultraviolet A (UVA). If the patient shows signs of a persistent photosensitivity, the minimum erythema dose (MED) must first be determined. If the MED is found to be reduced to 1/2 of the MED, it is used for PPT. The normal MED for UVA is over 20 J cm^{-2}.
- Readings should be recorded using the ICDRG scoring system (see Sect. 3.8.1) pre-irradiation, immediately post-irradiation, and 48 h post-irradiation. Further readings at 72 and 96 h post-irradiation are recommended to enable detection of crescendo and decrescendo scoring patterns, suggesting allergic and nonallergic mechanisms.

A true positive photopatch test persists or increases between the first and the second readings. Phototoxic, i.e., false positive reactions, are common. These are weak, macular reactions that fade in 24 h. An erythema occurring immediately after irradiation with UVA is also common. This is also a phototoxic response that fades in 24–48 h.

A product can be both a contact allergen and a photocontact allergen. To make a diagnosis of PACD, the photopatch test reaction should be greater than the patch test reaction.

5.6 Light Sources

The action spectrum for most photoallergens lies in the UVA range (315–400 nm). Hence, UVA is used for PPT. Any artificial source of light with a broad spectrum output of UVA is suitable for PPT. This is the case with the UVA lamps used in PUVA treatment units. If significant amounts of UVB are emitted, a window glass filter must be used, as UVB is far more erythemogenic than UVA.

The energy output of the light source must be known and monitored at intervals, as there may be fluctuation. The Waldmann Lichttechnik UV meter may serve as a standard monitoring device.

5.7 Proposal for a Photopatch Test Series

The proposal of the Taskforce is to include the following in the PPT series: (a) sunscreen agents; (b) some NSAIDs; and (c) optional allergens, added with reference to patient's medical history. Allergens are dispersed in petrolatum (Table 5.2). The series can be used worldwide. It requires to be adapted at regular intervals to fit in with environmental changes.

Table 5.2 Proposed allergens for PPT series

	Concentration (%)
A. Sunscreen series	
Octyl methoxycinnamate (2-Ethylhexyl-*p*-methoxycinnamate, Parsol MCX, Eusolex 2292)	10
Benzophenone-3 (2-Hydroxy-4-methoxy benzophenone, Oxybenzone, Eusolex 4360)	10
Octyl dimethyl PABA (2-Ethylhexyl-*p*-dimethyl-aminobenzoate, Escalol 507, Eusolex 6007)	10
PABA (4 Aminobenzoic acid)	10
Butyl methoxydibenzoylmethane (Parsol 1789, Eusolex 9020)	10
4-Methylbenzylidene camphor (Eusolex 6300, Mexoryl SD)	10
Benzophenone-4 (2-Hydroxy-4-methoxy-benzophenone-5-sulphonic acid, (Uvenyl MS-40)	10
Isoamyl *p*-methoxycinnamate (Neoheliopan, E1000)	10
Phenylbenzimidazole sulphonic acid (2-Phenyl-5-benzimidazolsulphonic acid, Eusolex 232)	10
B. NSAIDs: Non-steroidal anti-inflammatory agents (require to be prepared extemporaneously)	
Naproxen	5
Ibuprofen	5
Diclofenac	1
Ketoprofen	2.5
C. Additional tests (optional, according to patient's history)	
Cosmetic products	as is
Drugs (check irritation)	10–30
Occupational: olaquindox	1
Patient's own products, as appropriate	Suitable dilution

Concentrations quoted refer to petrolatum

References

1. Marks JG Jr, Elsner P, De Leo VA (2002) Contact and Occupational Dermatology, 3rd edn., Mosby, St.Louis
2. British Photodermatology Group (1997) Workshop report on photopatch testing methods and indications. Br J Dermatol 136:371–376
3. Schauder S, Schroder W, Geier J (1996) Olaquindox-induced airborne photoallergic contact dermatitis followed by transient or persistent light reactions in 15 pig breeders. Contact Dermatitis 35:344–354
4. Devleeschouwer V, Roelandts R, Garmyn M, Goossens A (2008) Allergic and photoallergic contact dermatitis from ketoprofen: results of (photo)patch testing and follow-up of 42 patients. Contact Dermatitis 58:159–166
5. Jansén CT, Wennersten G, Rystedt I, Thune P, Brodthagen H (1982) The Scandinavian standard photopatch test procedure. Contact Dermatitis 8:155–158
6. Bruynzeel DP et al. (2004) Photopatch testing: a consensus methodology for Europe. JEADV 18:679–682

The TRUE Test System

6

J.-M. Lachapelle and H.I. Maibach

6.1 Introduction

Conventional patch testing (as described and evaluated in other chapters of this book) is extensively used by the dermatological community throughout the world. Pitfalls of conventional patch testing are that allergens are not evenly dispersed in petrolatum and the dosage will vary between tests, as the allergens are manually dispensed.

TRUE Test represents an alternative way of patch testing [1], which intends to avoid variations of the allergens applied on the skin.

6.2 The TRUE Test System

The TRUE Test is a ready-to-use patch test system (Fig. 6.1). It represents a more sophisticated approach in the technology of patch testing, taking into account the parameter of optimal penetration and delivery of allergens through the skin [2]. The allergens are incorporated in hydrophilic gels. The gel (hydroxypropylcellulose and polyvinylpyrrolidone or syn.: polyvidone) is adapted to each individual allergen. The patches measure 0.81 cm^2 (9 mm^2) and the gel is coated on a polyester sheet. For protection against light and air, the strips are contained in airtight and opaque aluminum poaches.

Upon application of TRUE Test, perspiration and transepidermal water loss quickly rehydrate the dried gel layer, thereby releasing the allergens onto the skin [3, 4]. The homogeneous distribution of allergens helps to minimize the potential for false-positive and irritant reactions. To help ensure accurate testing and interpretation, the placement of allergens is standardized. Allergen stability has been confirmed by in vitro studies, and an optimized allergen dose level has been determined by in vivo dose–response studies [5].

TRUE Test is produced according to cGMP standard procedures used in manufacturing and quality control, which guarantees uniform quality and consistent performance [6].

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Fig. 6.1 The TRUE Test patch test system

The main advantages of TRUE Test are (a) the consistency and reproducibility of the test; (b) it is ready to apply and is time-saving. Its limitations are fourfold: (a) the cost, as compared with conventional patch testing; (b) the limited number of allergens available nowadays; (c) the fact that most epidemiological studies on patch test results are based on the use of conventional patch testing; and (d) the current series is out of date.

A reevaluation of comparative cost/benefit implications [7] of both systems could speed the move towards the widespread use of TRUE Test in the years to come.

6.3 The Standard TRUE Test Series

The standard TRUE Test series consists of 24 patches (two panels), with 12 allergen/allergen mixes on each of two panels (Table 6.1). Each patch is coated with a thin dry film that incorporates a specific allergen or allergen mixture in a calibrated dose.

The amount of allergen incorporated in each test is not expressed in terms of concentrations, but in terms of micrograms per centimeter square.

The list of TRUE Test standard series of allergens/allergen mixes differs slightly from lists proposed in conventional patch testing (see Chap. 4).

A concurrent right-vs.-left study using the TRUE Test system consisting of the two panels, each one containing 12 standard allergens, was performed on 500 consecutive patients [10]. This investigation showed that only 5% of patch test results were discordant. The authors explained the potential causes responsible for such discrepancies. They conclude that the technique is a reasonably reproducible procedure as long as methodological errors are minimized.

A panel 3 is under current evaluation in several centers and will include 12 new allergens (Table 6.2). The first 5 allergens on panel 3 are already commercially available in several markets. In US the first 4 allergens are today available. Panel 3 is aimed to extend the scope of investigation of allergic patients.

Table 6.1 The standard TRUE test series

Allergens	μg cm^{-2}
Panel 1	
1. Nickel sulphate	200
2. Wool alcohols	1,000
3. Neomycin sulphate	230
4. Potassium dichromate	23
5. Caine mix	630
6. Fragrance mix	430
7. Colophony	850
8. Epoxy resin	50
9. Quinoline mix	190
10. Balsams of Peru	800
11. Ethylenediamine dihydrochloride	50
12. Cobalt chloride	20
Panel 2	
13. *p-tert*-Butylphenol formaldehyde resin	50
14. Paraben mix	1,000
15. Carba mix	250
16. Black rubber mix	75
17. Cl + Me-Isothiazolinone (Kathon CG)	4
18. Quaternium 15	100
19. Mercaptobenzothiazole	75
20. *p*-Phenylenediamine	90
21. Formaldehyde (*N*-hydroxymethyl succinimide)	180
22. Mercapto mix	75
23. Thiomerosal (thiomersal)	8
24. Thiuram mix	25

Investigations conducted with Panel 1 [8] and Panel 2 [9] show that results are highly reproducible.

6.4 Methodology of Use

Application of TRUE Test® is as follows [11]:

1. The envelope is opened, the panel is removed, and the backing is removed from the series.
2. The adhesive backing is numbered (1–12 for panel 1 and 13–24 for panel 2). The panel 1 is applied to the left back by first laying the lower edge of the adhesive backing to the back skin and then slowly applying gentle pressure with the fingers as the rest of the panel is smoothed upward.

Table 6.2 The standard TRUE test panel 3

Allergens	µg cm^{-2}
Panel 3	
25. Diazolidinyl urea (Germall II)	0.600
26. Imidazolidinyl urea (Germall 115)	0.600
27. Budesonide	0.001
28. Tixocortol-21-pivalate	0.032
29. Hydrocortisone-17-butyrate	0.020
30. Gold sodium thiosulfate	0.075
31. Methyldibromoglutaronitrile	0.0055
32. Bacitracin	0.60
33. Parthenolide	0.0030
34. Disperse blue 106	0.050
35. 2-Bromo-2-nitropropane-1,3-diol (Bronopol)	0.25
36. Lyral	[a]

[a]Under current investigation

3. Panel 2 is applied in a similar fashion to the right upper back. The marker pen is used to mark the position of the notches on the panels.
4. A reading template is supplied with each test kit to ease the reading of the test results.

When using TRUE Test, the reading scores (Fig. 6.2a–c) are identical to those adopted for conventional patch testing (see Sect. 3.8).

6.5 Regulatory Information

Importantly, from a regulatory viewpoint, patch test preparations are regarded as an immunological medicinal product (Directive 2001/83/EC) and have to be approved by the responsible federal authorities. No medicinal product may be put on the market of a Member State unless a marketing authorization has been issued by the competent authorities of this Member State in accordance with this directive. This approval guarantees the pharmaceutical and analytical quality, toxic safety, and clinical relevance. In most European markets, TRUE Test is the only product with such a market authorization. In the US, the TRUE Test is currently the only allergen patch test that has received marketing approval from the US Food and Drug Administration (FDA) [12].

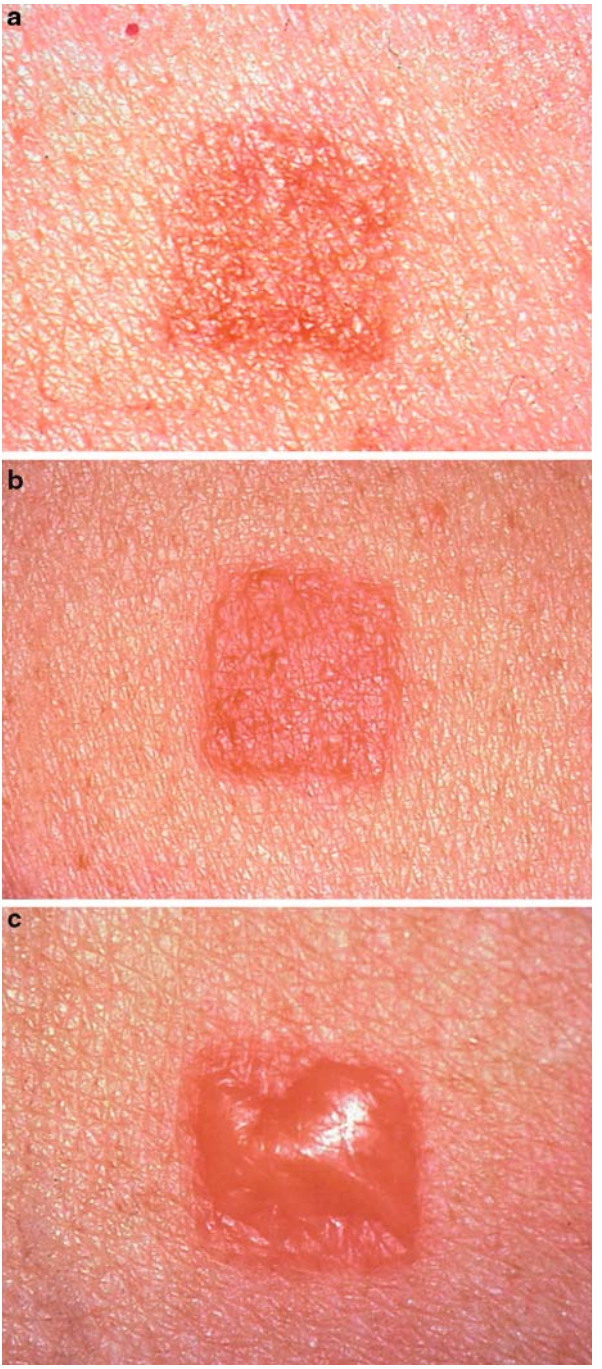

Fig. 6.2 TRUE test. Scoring positive allergic patch test reactions. (**a**) + reaction; (**b**) ++ reaction; (**c**) +++ reaction (see explanations in text)

6.6 Additional Practical Information

TRUE Test is supplied in boxes of 10 standard tests (2 panels × 10). Advice is given to store it at +2 to +8°C. The shelf live under the above conditions for 24 months. The expiry date is stated on the package.

To assist when interpreting the results and advising the patient, TRUE Test system includes the following:

1. Templates to identify each allergen
2. Patient information leaflets, which answer the most commonly asked questions about the test procedure and assist the physician.

TRUE Test is manufactured by Mekos Laboratories ApS, Herredsvejen 2, DK-3400 Hillerod, Denmark, Phone: +45 48 20 71 00, Fax: +45 48 20 71 01, e-mail: www.mekos.dk, info@mekos.dk.

TRUE Test is available through various distributors throughout the world (a complete list given on www.mekos.dk). New name of the company scheduled for beginning in 2009: SmartPractice® Denmark ApS.

Additional information on allergen components are given in Table 6.3.

6.7 Conventional Patch Testing vs. TRUE Test: The Current Situation

In European countries, many dermatologists working in University (and non-University) hospitals are using the conventional patch testing methodology (see Chap. 3). Nevertheless, some of them have switched over to the TRUE Test system for routine daily practice. It has to be pointed out that the two methods produce different results when the two methods are used simultaneously in the same subjects. Some patch tests are positive with one method, whereas they are negative with the other and vice versa. Where is the truth, when the percentage of reproducibility is 60–70% [13, 14]? No relevant explanation does exist to date. Practically, when dermatologists use a defined methodology, they are committed to note it in their files.

Morevover, when results of positive patch tests are crucial for the patient, it is of prime importance to repeat patch testing with the alternative technique.

Another point of concern refers to the self-application and the self-reading by patients of TRUE Test, related to its easy mode of application. The practice must be condemned; patch test reading is a medical deed, leading to research for relevance (see Chap. 8).

Table 6.3 TRUE test: allergen concentration, allergen component per patch, vehicle

Allergen	Concentration (mg cm^{-1})	Allergen component per patch	Vehicle
1. Nickel sulphate	0.20	Nickel, 0.036 mg	Hydroxypropylcellulose
2. Wool alcohols (lanolin)	1.00	Cholesterol, lanosterol, agnosterol (and dihydro derivatives); straight- and branched-chain aliphatic alcohols; 0.81 mg total (the active allergenic component has not been identified)	Polyvidone
3. Neomycin sulphate	0.023	Neomycin sulphate, USP, 0.19 mg	Methylcellulose
4. Potassium dichromate	0.023	Chromium, 0.0067 mg	Hydroxypropylcellulose
5. Caine mix	0.63	Benzocaine, USP, 0.364 mg; tetracaine HCl, USP, 0.063 mg; dibucaine HCl, USP, 0.064 mg	Polyvidone
6. Fragrance mix	0.43	Geraniol, 0.070 mg; cinnamaldehyde, 0.034 mg; hydroxycitronellal, 0.054 mg; cinnamyl alcohol, 0.054 mg; eugenol, 0.034 mg; isoeugenol, 0.015 mg; amylcinnamaldehyde, 0.015 mg; oak moss, 0.070 mg	Hydroxypropylcellulose, cyclodextrin
7. Colophony	0.85	Colophony, 0.69 mg	Polyvidone
8. Epoxy resin	0.050	Diglycidylether of bisphenol A, 0.032 mg	Hydroxypropylcellulose
9. Quinoline mix	0.190	Clioquinol: 0.077 mg, Chlorquinaldol: 0.077 mg	Polyvidone
10. Balsams of Peru	0.80	Balsams of Peru, 0.65 mg total	Polyvidone
11. Ethylenediamine dihydrochloride	0.050	Ethylenediamine, 0.018 mg	Methylcellulose
12. Cobalt dichloride	0.020	Cobalt, 0.0040 mg	Hydroxypropylcellulose
13. *p-tert*-butylphenol formaldehyde resin	0.050	*p-tert*-Butylphenol formaldehyde, 0.041 mg	Hydroxypropylcellulose
14. Paraben mix	1.00	Methyl *p*-hydroxybenzoate, 0.162 mg; ethyl *p* -hydroxybenzoate, 0.162 mg; propyl *p* -hydroxybenzoate, 0.162 mg; butyl *p* -hydroxybenzoate, 0.162 mg; benzyl *p* -hydroxybenzoate, 0.162 mg	Polyvidone
15. Carba mix	0.25	Diphenylguanidine, 0.067 mg; zinc dibutyldithiocarbamate, 0.067 mg; zinc diethyldithiocarbamate, 0.067 mg	Hydroxypropylcellulose

(continued)

Table 6.3 (continued)

Allergen	Concentration (mg cm^{-1})	Allergen component per patch	Vehicle
16. Black rubber mix	0.075	*N*-Isopropyl-*N* -phenyl *p*-phenylenediamine, 0.0102 mg; *N*-cyclohexyl-*N*′-phenyl *p* -phenylenediamine, 0.0255 mg; *N*,*N*′ -diphenyl *p*-phenylene-diamine, 0.0255 mg	Polyvidone
17. Cl + Me- Isothiazolinone	0.0040	5-Chloro-2-methyl-4-isothiazolin-3-one, 0.0024 mg; 2-methyl-4-isothiazolin-3-one, 0.0008 mg	Polyvidone
18. Quaternium-15	0.100	Quaternium-15, 0.081 mg	Hydroxypropylcellulose
19. Mercaptobenzothiazole	0.075	Mercaptobenzothiazole, 0.061 mg	Polyvidone
20. *p* -Phenylenediamine	0.090	*p*-Phenylenediamine, 0.073 mg	Polyvidone
21. Formaldehyde	0.18	Formaldehyde, 0.15	Polyvidone
22. Mercapto mix	0.075	*N*-Cyclohexyl benzothiazyl-sulfenamide, 0.0203 mg; dibenzothiazyl disulfide, 0.0203 mg; morpholinylmercaptobenzothiazole, 0.0203 mg	Polyvidone
23. Thiomersal	0.0080	Thiomersal 0.0065 mg	Hydroxypropylcellulose
24. Thiuram mix	0.025	Tetramethylthiuram monosulfide, 0.0051 mg; tetramethylthiuram disulfide, 0.0051 mg; disulfiram, USP, 0.0051 mg; dipentamethylene thiuram disulfide, 0.0051 mg	Polyvidone
25. Diazolidinyl urea	0.55	Diazolidinyl urea, 0.45 mg	Polyvidone
26. Imidazolidinyl urea	0.60	Imidazolidinyl urea, 0.49 mg	Hydroxypropylcellulose
27. Budesonide	0.0010	Budesonide, 0.00081 mg	Polyvidone
28. Tixocortol pivalate	0.00300	Tixocortol pivalate, 0.0024 mg	Polyvidone
29. Hydrocortisone-17-butyrate	0.020	Hydrocortisone-17-butyrate, 0.016 mg	Polyvidone

References

1. Andersen KE (2002) The interest of the True Test in patch testing. Ann Dermatol Venereol 129:1S148
2. Fischer T, Maibach HI (1985) The thin layer rapid use epicutaneous test (TRUE-Test), a new patch test method with high accuracy. Br J Dermatol 112:63–68
3. Jensen CD, Andersen KE (2005) Course of contact allergy in consecutive eczema patients patch tested with TRUE Test panels 1 and 2 at least twice over a 12-year period. Contact Dermatitis 52:242–246
4. Woo DK, Militello G, James WD (2004) Neoprene. Dermatitis 15:206–209
5. Lazarov A (2004) Textile dermatitis in patients with contact sensitization in Israel: a 4-year prospective study. JEADV 18:531–537
6. Fischer T, Kreilgard B, Maibach HI (2001) The True value of the True Test for allergic contact dermatitis. Curr Allergy Asthma Reports 1:316–322
7. Rajagopalan R, Anderson RT, Sama S (1998) An economic evaluation of patch testing in the diagnosis and management of allergic contact dermatitis. Am J Contact Dermatitis 9:149–154
8. Lachapelle JM, Bruynzeel DP, Ducombs G, Hannuksela M, Ring J, White IR, Wilkinson JD, Fischer T, Bilberg K (1988) European multicenter study of the True Test®. Contact Dermatitis 19:91–97
9. Wilkinson JD, Bruynzeel DP, Ducombs G, Frosch PJ, Gunnarsson Y, Hannuksela M, Lachapelle JM, Ring J, Shaw S, White IR (1990) European multicenter study of True Test®, Panel 2. Contact Dermatitis 22:218–225
10. Ale SI, Maibach HI (2004) Reproducibility of patch test results: a concurrent right–*versus*–left study using TRUE Test. Contact Dermatitis 50:304–312
11. Marks JG Jr, Elsner P, De Leo V (2002) Contact and Occupational Dermatology. 3rd edn., Mosby, St.Louis, p 47
12. Belsito DV (2004) Patch testing with a standard allergen ("screening") tray: rewards and risks. Dermatol Ther 17:231–239
13. Goh CL (1992) Comparative study of TRUE Test and Finn Chamber patch test techniques in Singapore. Contact Dermatitis 27:84–89
14. Lazarov A, David M, Abraham D, Trattner A (2007) Comparison of reactivity to allergens using the TRUE Test and IQ chamber system. Contact Dermatitis 56:140–145

Additional Testing Procedures

7

J.-M. Lachapelle and H.I. Maibach

7.1 Stripping Test

The stripping test, proposed by Spier [1], is a variant of patch testing (PT). It consists of "stripping" the stratum corneum before applying the allergens in the usual way. The aim of the technique is to remove most layers of the stratum corneum and to consequently suppress the skin barrier. This technique is theoretically useful for allergens with poor penetration through the skin, e.g., neomycin. It is easily performed by stripping the skin 8–12 times with a cellophane tape. Its main drawback is the fact that it provokes by itself skin irritation [2] that interferes with the reading; nevertheless, it can be performed in well-defined conditions parallel to conventional PT. Reading of results needs caution and expertise. The method has passed into disuse, due to its time-consuming limitations. It has been reevaluated by Brazilian dermatologists [3]. They concluded that the stripping test provides more positive relevant PT reactions than conventional PT. Obviously, it cannot be used routinely (time limitations), but it can be advised for testing allergens with a low penetration potential.

7.2 Open Test

Open test means that a product, as is or dissolved in water or some solvent (e.g., ethanol, acetone, methylethylketone, etc.), is dropped onto the skin and allowed to spread freely. No occlusion is used. The usual test site is the volar forearm, and the surface of spreading is usually limited to $5 \times 5\,cm^2$ (Fig. 7.1).

An open test is recommended as the first step when testing poorly defined or unknown substances or products, such as those brought by the patient (paints, glues, oils, cleansing agents, etc.). Readings are similar to those adopted for conventional PT (see Sect. 3.8).

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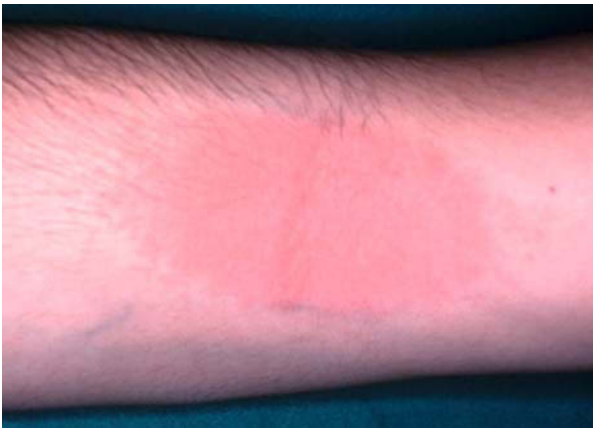

Fig. 7.1 Open test. Positive allergic reaction to a perfume, after one single application. Read at 48 h

A negative open test does not preclude that allergy is not present, since it can be explained by insufficient penetration. With unknown substances, it indicates that one may go on with an occlusive patch test.

Another application of the open test is to "trap" eventual immediate (urticarial) reactions from well-known allergens, such as balsam of Peru or cinnamic aldehyde (see Sect. 3.7). The technique to be applied is similar to that described earlier.

7.3 Semi-Open Test

The semi-open test is an interesting variant of the open test, following the same principle of non-occlusion. The only difference from the open test is that the products, applied on the skin, are covered by a non-occlusive tape (e.g., Micropore, Fixomull) when they have dried off (about 5–10 min).

The semi-open test is thus "half-way" between open testing and conventional PT, and is particularly useful when testing is carried out with industrial and/or domestic products (Fig. 7.2a–c). Therefore, it is extensively used in some countries, mainly in units of occupational dermatology. Various sites can be used, such as the upper back, the extensor aspect of the arm, or the volar aspect of the forearm. It is mandatory to check the pH of household and industrial products [4].

Its main advantage compared to conventional PT is avoidance (or reduction) of skin irritation when unknown products are applied onto the skin. It is therefore easier to make the distinction between contact allergy and irritation, but false-negative reactions do occur due to insufficient penetration of products.

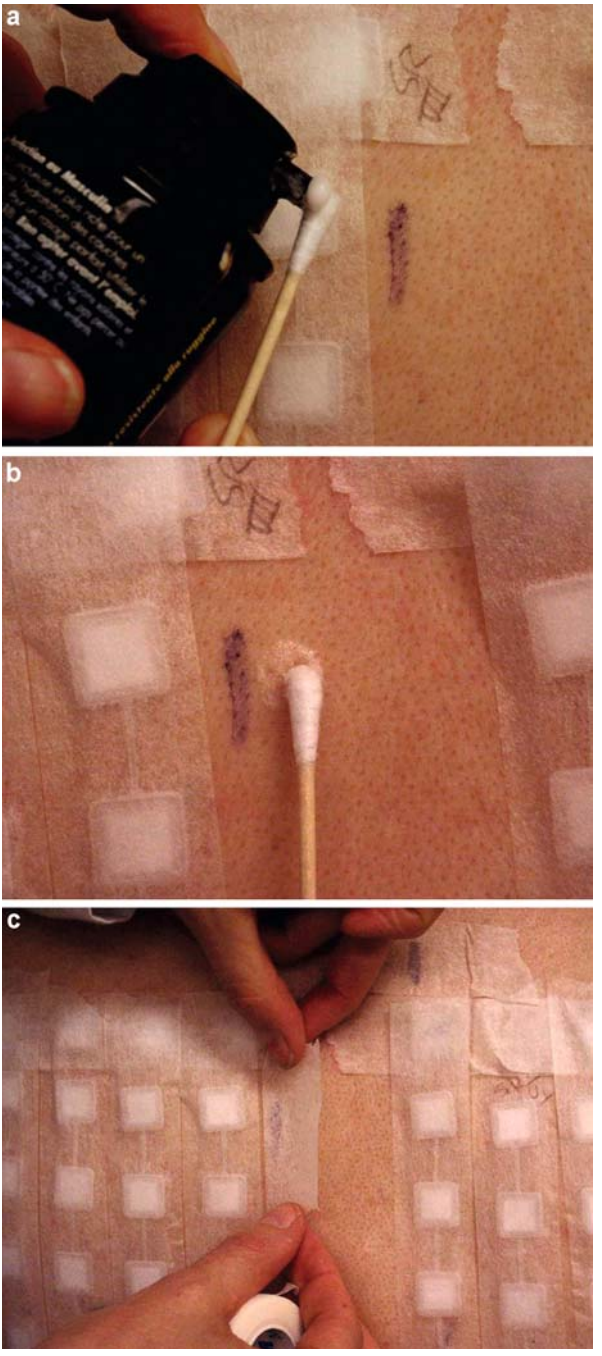

Fig. 7.2a–c Semi-open test. Three-step procedure. (**a**) Spreading a glue sample (as is) on a swab. (**b**) Smearing the glue on a marked skin site. (**c**) Covering the skin site with a non-occlusive tape (by courtesy of A. Goossens)

7.4 Repeated Open Application Test

The repeated open application test (ROAT) was standardized by Hannuksela and Salo [5]. Test substances, either commercial products, as is, or special test substances (e.g., patch test allergens) are applied twice daily for 7 days to the outer aspect of the upper arm, antecubital fossa, or back skin (scapular area). The size of the test area is not crucial: a positive result may appear 1–2 days later on a 1 × 1-cm^2 area than on a larger area. The amount of test substance should be approximately 0.1 ml at a 5 × 5-cm^2 area and 0.5 ml at a 10 × 10-cm^2 area [6, 7]. A positive-response eczematous dermatitis usually appears on days 2–4, but it is recommended to extend the applications beyond 7 days so as not to miss late-appearing reactions. It is our experience that reactions (as late as 28 days, i.e., 56 applications) may occur, e.g., with scented cosmetics (such as deodorants, creams, lotions, etc.). It is worthwhile to test at the three sites concomitantly, because one test area can react in an unpredictable way sooner than the other two. The patient is asked to stop the application of the test substance(s) when he or she notices a reaction [5]. The clinical features of positive ROAT reactions may be surprising for the dermatologist, compared to those observed in conventional PT.

Erythema (diffuse or spotted) and follicular elevations (Fig. 7.3) looking like tiny papules are commonly observed. When these symptoms appear after the first applications, irritation cannot be ruled out, and similar applications in control subjects are needed. Oedematous and/or vesicular reactions are rare. Therefore, the technique requires correct interpretation. When carefully conducted, it provides good information (Figs. 7.4 and 7.5) and is particularly useful for comparative studies (e.g., the application of a scented cosmetic product on the three sites of the left side, compared with the application of the same product, but unscented on the right side). A refined scheme for the scoring of ROAT reactions was presented [8].

The value of ROAT has been verified in cases with positive, negative, or questionable reactions at initial PT and in animal studies.

The morphology of ROAT on arm, neck, and face in formaldehyde- and diazolidinyl urea-sensitive individuals was studied recently [9]. On the arm and neck, the dominant

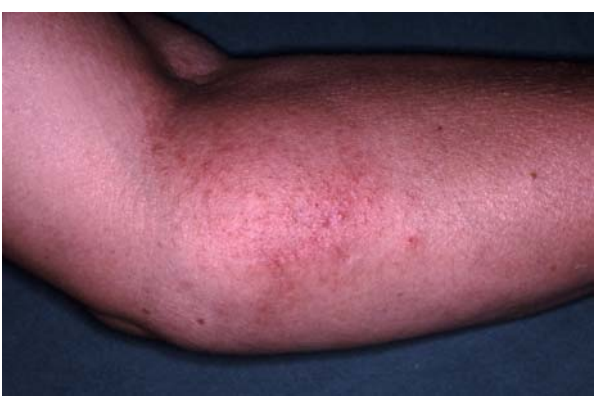

Fig. 7.3 ROAT test to a body lotion. Positive allergic erythematous and vesicular (mainly follicular) reaction after 10 applications

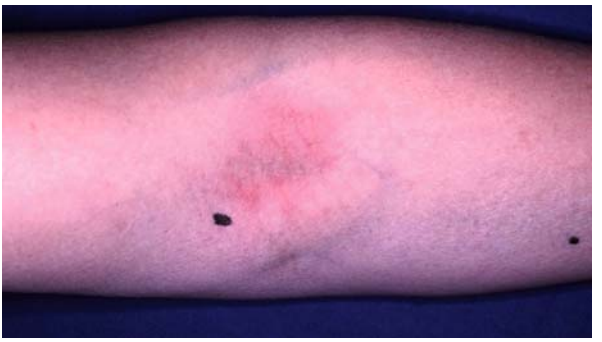

Fig. 7.4 ROAT test to a shaving foam. Positive allergic reaction after 14 applications

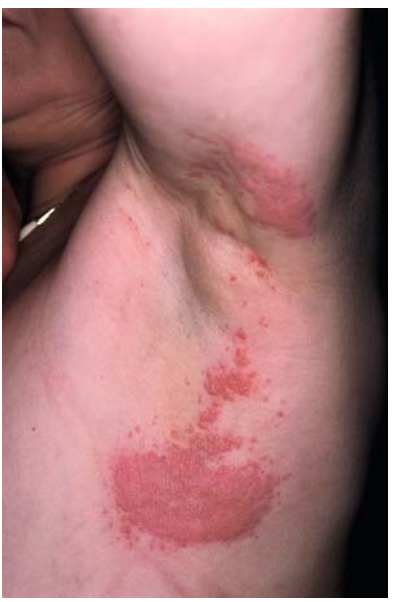

Fig. 7.5 ROAT test to a deodorant stick. Positive reaction after three applications

initial morphology was an eczematous papular eruption. In the face, the initial skin changes were more homogeneous and infiltrated erythema.

The provocative use test (PUT) is synonymous with the ROAT test.

7.5 Testing Procedures with Unknown Substances

"Wild" uncontrolled testing with totally unknown products is prohibited. Necrosis, scarring, keloids, pigmentation, depigmentation, and any other complications listed earlier (see Sect. 3.14) can appear and the dermatologist may be accused of malpractice.

7.5.1 Strategy

When patients bring suspected products or materials from their (work) environment, we recommend that adequate product safety data sheets, lists of ingredients etc. are requested from the manufacturer so that a general impression of the product, ingredients, concentrations, and intended use etc. can be formed. There are usually one or two ingredients that are of interest as suspected allergens, while the rest are well-known substances of proven innocuousness and/or known irritancy for which detailed information is available. For substances or products where skin contact is unintentional and the dermatitis is a result of misuse or accident, detailed information from the manufacturer is required before any tests are initiated [10].

7.5.2 Steps Required Prior to Any Testing Procedure

The next step is to look for the suspected allergens. If they are available from suppliers of patch test allergens, one can rely on the choice of vehicle and concentration. If one suspects that impurities or contaminants caused the dermatitis, this can only be discovered via samples of the ingredient from the manufacturer.

If it is an entirely new substance, where no data on toxicity are available, the patient and the dermatologist must decide how to find an optimal test concentration and vehicle, and must discuss the risk of complications. To minimize the risk, one can start with an open test and, if this is negative, continue with occlusive patch testing. Most allergens are tested in the concentration range 0.01%–10% and we usually start with the lowest and raise the concentration when the preceding test is negative. A practical method is to apply 0.01% and 0.1% for 1 day in a region where the patient can easily remove the patch her- or himself (upper back or upper arm). If severe stinging of burning occurs, the patient should be instructed to remove the patch immediately. If the test is negative, the concentration can be raised to 1%. Occasionally, the likely irritant or sensitization potential of a chemical may be such that starting with concentrations of 0.001% and 0.01% is advisable, increasing to 0.1% if negative. An alternative is to start with a higher concentration, but with reduced exposure time (5 h), but this procedure is not sufficiently standardized.

An important check point is the pH of the product to be tested. It is unwise to test with a product whose pH is below 4 or above 9 (see Sect. 7.7.1).

If the patient's test is positive, the clinician must demonstrate in unexposed controls that the actual test preparation is non-irritant. Otherwise the observed reaction in the particular patient does not prove allergenicity.

When testing products are brought by the patient, it is essential to use samples from the actual batch to which the patient was exposed, but also when testing, for example, cutting fluids, unused products must be tested for comparison. When testing with dilutions, one runs the risk of overlooking true allergens by using over-diluted materials.

7.5.3
Testing Procedures with Solid Products and Extracts

When a solid product is suspected (e.g., textiles, rubber, plants, wood, paper), this can usually be applied as it is. Rycroft [11] recommends that the material be tested as wafer-thin, regular-sided, smooth sheets, e.g., rubber (Figs. 7.6a,b and 7.7), or as finely divided

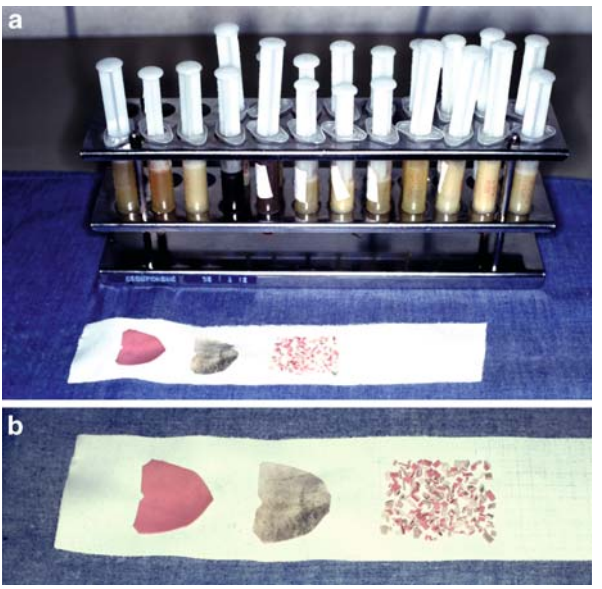

Fig. 7.6 a,**b** Testing procedure applied to rubber gloves: (**a**) rubber additives series and rubber pieces of gloves; (**b**) rubber pieces are cut into small fragments to increase contact with the skin

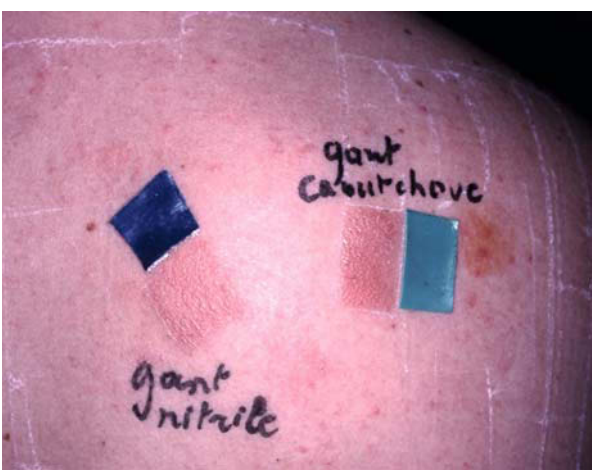

Fig. 7.7 Testing procedure applied to gloves. Positive allergic patch test reactions at 48 h to a nitrile glove (*left*) and to a rubber glove (*right*)

particulates (e.g., woods). Plants and woods and their extracts constitute special problems, due to variations in the quantity of allergens produced and their availability on the surface. Extracts for testing can be obtained by placing the product or sample in water, synthetic sweat, ethanol, acetone or ether, and heating to 40–50°C. False reactions to non-standardized patch tests have been reviewed by Rycroft [11].

When patch testing with solid materials, a classic unwanted reaction is the pressure effect (see Sect. 3.14).

7.5.4 Testing Procedures with Cosmetics and Other Related Products

For most products with intended use on normal or damaged skin (e.g. cosmetics, skin care products, soaps, shampoos, detergents, topical medicaments), detailed predictive testing and clinical and consumer trials have been performed. The results can usually be obtained from the manufacturer. For this category of products, open tests (see Sect. 7.2), semi-open tests (see Sect. 7.3) and ROAT tests (see Sect. 7.4) probably give more information on the pathogenesis of the patient's dermatitis than an occlusive patch test does. Suggestions on concentrations and vehicles can be found in textbooks.

7.6 Oral Provocation Test (Oral Challenge)

The oral provocative test is rarely conducted in the field of allergic contact dermatitis. It has been mainly used in cases of recurrent vesicular palmar eczema (pompholyx), in which systemic administration of allergens is considered significant in provoking recurrences of the disease. Nickel is the most often incriminated culprit [12].

The assumption that there is an association between nickel allergy and recurrent vesicular hand eczema is supported by several trials of placebo-controlled oral challenge with doses of nickel ranging from 0.5 to 5.6 mg. These studies indicate that an oral dose of nickel may reactivate vesicular hand eczema in nickel-sensitive patients and that the response is dose-dependent. A dose of 0.5 mg nickel will reactivate vesicular hand eczema in only a small proportion of nickel-sensitive patients. Oral challenge with 2.5 mg nickel will cause a flare of dermatitis in approximately 50% of such patients, and a majority of nickel-sensitive patients will experience a flare-up reaction after a dose of 5.6 mg nickel [13]. Foods rich in nickel content may cause flares of vesicular hand eczema.

Cobalt and chromates have also been suspected, but oral challenge with these metals is not of common use.

Other investigations are related to balsam of Peru and spices. These are sparse. Veien et al [14] challenged 17 balsam-sensitive patients with 1 g balsam of Peru. Four of four patients with recurrent pompholyx had flare-up reactions after oral challenge with balsam but not after challenge with a placebo. Dooms-Goossens et al. [15] studied reactions to spices and described three patients who had pompholyx that flared after ingestion of various spices.

Oral challenge is of utmost importance when investigating some drug eruptions (see Sect. 12.3).

7.7 Other Investigations

Some in vitro investigations are focused on the characteristics and the detection of irritants and/or allergens in "end-products," susceptible to be tested at the clinic.

7.7.1 pH Measurement

Acidic and, particularly, alkaline products play a significant role in the development of irritant contact dermatitis. It is important to determine the degree of acidity or alkalinity in a product suspected of causing skin problems to avoid false-positive diagnoses of ACD. As mentioned earlier (see Sect. 7.5.2), it is not wise to test with a product whose pH is below 4 or above 9.

pH determinations are relevant only in water-based products/solutions. A universal pH paper is usually satisfactory for clinical use. A few drops of the solution or the emulsion are applied on the pH paper. The resulting color is compared with the color stage of the pH paper. A pH paper moistened with water can be applied to solid objects to demonstrate residual acidic or alkaline solution on the object. For accurate determination of the pH in a solution, a pH meter is necessary [16].

7.7.2 Spot Tests

Spot tests can be used to demonstrate both inorganic and organic compounds in several items [16]. A specific reagent may react with a specific substance to give a specific color and thus indicate the occurrence of the specific substance. A few spot tests can be used routinely by dermatologists.

7.7.2.1 Dimethylglyoxime Test for Nickel

Nickel is most commonly detected by using the dimethylglyoxime test. A few drops each of dimethylglyoxime 1% in ethanol and ammonium hydroxide 10% in water are applied to a cotton-tipped applicator, which is rubbed against the metal object to be investigated [16]. Dimethylglyoxime reacts with nickel ions in the presence of ammonia, giving a red salt. Coins known to contain nickel can be used to test the reagent and to observe the pink

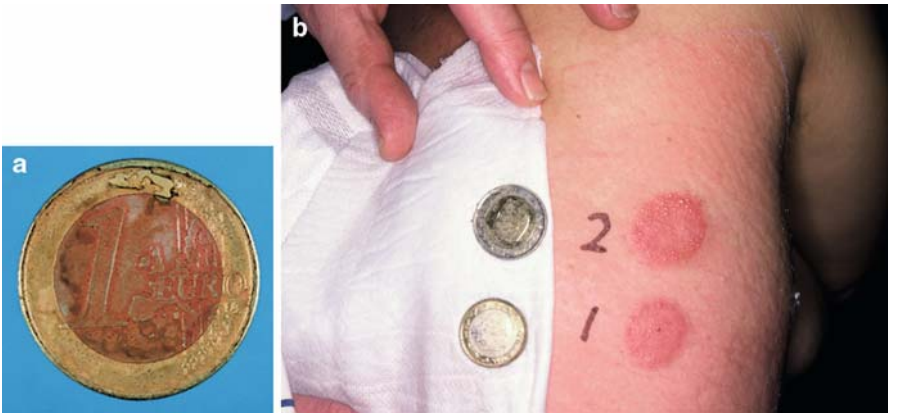

Fig. 7.8a,b Dimethylglyoxime spot test for nickel. (**a**) Positive spot test. One-euro coin. (**b**) Positive patch test to one- and two-euro coins in a patient sensitized to nickel

color. The solutions can also be applied directly on the metallic objects. Chemotechnique has developed a nickel spot test that consists of an ammoniacal solution of dimethylglyoxime (thus, only one solution is used). The test detects free nickel down to a limit of 10 ppm. The sensitivity of the test can be enhanced by pretreatment of the surface of the object with a solution of synthetic sweat and by heating. The method is very simple and can be used by dermatologists and nickel-allergic patients to detect nickel release from various metallic objects (Fig. 7.8).

7.7.2.2 Diphenylcarbazide Test for Hexavalent Chromium (Chromate)

The chromium spot test is valid only for hexavalent chromium. *Sym*-diphenylcarbazide reacts with chromate and dichromate ions in the presence of sulfuric acid, giving a red-violet color. Reagents: (I) *Sym*-diphenylcarbazide 1% w/v in ethanol (must be prepared immediately before the investigation). (II) Sulfuric acid 1 ml/L. Reference: Solutions of potassium chromate 2.0, 1.0, 0.5, and 0.25 μg chromate/ml [16].

- *Chromate on the surface of a solid object*. A few drops each of the reagents I and II are applied to a cotton swab. The cotton swab is, thereafter, rubbed against the surface of the object for 1 min. If chromate is present, a red-violet color appears.
- *Chromate in solutions*. To a sample of approximately 10 ml, a few drops each of the reagents I and II are added. If chromate is present, a red-violet color appears.
- *Chromate in powders insoluble in water (e.g. cement)*. Five grams of cement is mixed with 10 ml of water for few minutes. The mixture is then filtered and the filtrate is handled as for chromate in solutions (Fig. 7.9). Iron ions can interfere with the reagent and give discolored solutions.

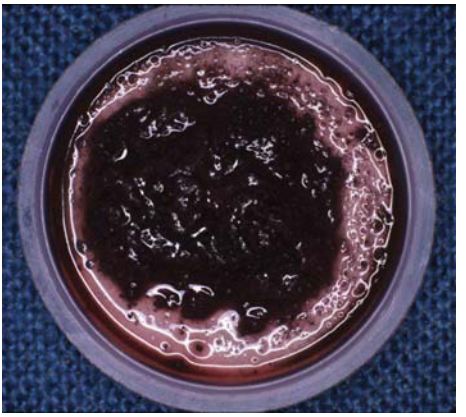

Fig. 7.9 Diphenylcarbazide spot test for chromate in cement. Positive reaction (see explanations in text)

7.7.2.3
Chromotropic Acid Test for Formaldehyde

Forty milligrams of chromotropic acid is dissolved in 10 ml of concentrated sulfuric acid (freshly prepared). Standard solutions: a concentrated water solution of formaldehyde (35%) is diluted to 100 μg/ml and refrigerated (stock solution). Standard solutions containing 2.5, 10, 20, and 40 μg formaldehyde/ml are prepared. The standard solutions should be refrigerated and freshly prepared every week [16].

Approximately, 0.5 g of the sample is placed in a 25-ml glass jar with a ground-glass stopper. Then 1 ml of each standard solution and 1 ml water (blank) is placed in separate glass jars. Then, 0.5 ml of the reagent is added to small glass tubes and placed individually in the glass jars containing the sample, the standards, and the blank, respectively. The jars are kept in dark and observed after 1 and 2 days. A violet color indicates the presence of formaldehyde (Fig. 7.10).

This method is based on chemical reaction of chromotropic acid and free formaldehyde evaporated from the sample/standards [17]. However, other aldehydes and ketones can also react with chromotropic acid, giving colors that can interfere with the violet reagent.

With the chromotropic acid method, a rough estimation of the concentration of formaldehyde can be obtained by comparing the intensity of the sample color with those of the standards.

In occupational medicine, detection of formaldehyde in air is performed with the Bio-Check Dräger technique. It is a very sensitive method (0.05, 1 > 1 ppm).

7.7.2.4
Other Spot Tests

Other spot tests are available; but they are too elaborate for use in clinical practice. They can detect, for example, epoxy resin based on bisphenol A [16] or dyes from textiles [18].

Fig. 7.10 Chromotropic acid spot test for formaldehyde in shampoos. Negative (*left*) and positive reactions (see explanations in text)

7.7.3 Chemical Analysis

To detect the presence of allergens in products or items brought by patients, chemical analysis can be performed in specialized laboratories (Table 7.1).

Table 7.1 Methods of chemical analysis

Thin-layer chromatography
Gas chromatography
Atomic absorption spectrophotometry
UV–vis spectrophotometry
Infrared spectrophotometry
Mass spectrometry
Inductively coupled plasma-mass spectrometry
Nuclear magnetic resonance spectroscopy

References

1. Spier HW, Sixt I (1955) Untersuchungen über die Abhangigkeit des Ausfalles der Ekzem – Lappchenprobes van der Hornschichtdieke (Quantitativer Abrisss-Epicutantest) Hautarzt 6:152–159
2. Schaefer H, Redelmeier TE (1996) Skin barrier. Principles of percutaneus absorption. Karger, Basel, p172
3. Fernandes MFM, de Mello JF, Pires MC, Vizeu MCM (2007) Comparative study of patch test using traditional method ***versus*** prior skin abrading. JEADV 21:1351–1359
4. Goossens A (2003) Le test semi-ouvert. Dermatologie Actualité (Bruxelles) 79:16–17

5. Hannuksela M, Salo H (1986) The repeated open application test (ROAT). Contact Dermatitis 14:221–227
6. Hannuksela M (1991) Sensitivity of various skin sites in the repeated open application test. Am J Contact Dermatitis 2:102–104
7. Hannuksela A, Ninimäki A, Hannuksela M (1993) Size of the test area does not affect the result of the repeated open application test. Contact Dermatitis 28:299–300
8. Johansen JD, Bruze M, Andersen KE, Frosch PJ, Dreier B, White IR, RastogiI S, Lepoittevin JP, Menné T (1998) The repeated open application test: Suggestions for a scale of evaluation. Contact Dermatitis 39:95–96
9. Zachariae C, Hall B, Cupferman S, Andersen KE, Menné T (2006) ROAT: morphology of ROAT on arm, neck and face in formaldehyde and diazolidinyl urea sensitive individuals. Contact Dermatitis 54:21–24
10. Wahlberg JE, Lindberg M (2006) Patch Testing. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis. 4th edn. Springer, Berlin, pp 365–390
11. Rycroft RJG (1986) False reactions to nonstandard patch tests. Semin Dermatol 5:225–230
12. Veien NK, Menné T (2000) Acute and recurrent vesicular hand dermatitis (pompholyx), Chapter 15. In: Menné T, Maibach HI (eds) Hand Eczema, 2nd edn. CRC Press, Boca Raton, pp 147–164
13. Veien NK, Hattel T, Justesen O, Norholm A (1987) Oral challenge with nickel and cobalt in patients with positive patch tests to nickel and/or cobalt. Acta Derm Venereol 67:321–325
14. Veien NK, Hattel T, Justesen O, Norholm A (1985) Oral challenge with balsam of Peru. Contact Dermatitis 12:104–107
15. Dooms-Goossens A, Dubelloy R, Degreef H (1990) Contact and systemic contact-type dermatitis to spices. Contact Dermatitis 8:89–92
16. Gruvberger B, Bruze M, Fregert S, Lidén C (2006) Allergens exposure assessment. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 413–427
17. Dahlquist I, Fregert S, Gruvberger B (1980) Reliability of the chromotropic acid method for qualitative formaldehyde determination. Contact Dermatitis 6:357–358
18. Le Coz CJ (2006) Clothing. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 679–702

Clinical Relevance of Patch Test Reactions

8

J.-M. Lachapelle and H.I. Maibach

8.1 Introduction

Reading patch test results cannot be limited to scoring as positive or negative. Scoring in itself has no meaning if it is not linked in some way with the medical history of the patient. In other words, a positive patch test (and to some extent a negative patch test) has no interest if it is not labeled as relevant or nonrelevant. Incidentally, this concept is valid also for all laboratory investigations [1].

8.2 General Principles

To diagnose allergic contact dermatitis, two significant steps should be considered:

1. Demonstrating the existence of contact allergy to one or several allergens
2. Demonstrating their clinical relevance

The first step is fulfilled when a positive patch test reaction deemed to reveal the presence of a genuine contact hypersensitivity is obtained. This involves assessing the morphology of the reaction and deciding whether it represents a true-positive allergic reaction as opposed to a false-positive one. Accurate reading and interpretation of patch test reactions are difficult tasks. Different variables, that is, type of patch test system, sources of patch test allergens, amount of allergen applied, criteria of patient's selection, application and reading times, skin area, and variations in biological responsiveness, may influence the test result [2]. Other notorious disadvantage of patch testing is that reading is eminently subjective, based on inspection and palpation of the test sites. Even if the ICDRG criteria concerning an uniform scoring system for patch test readings and a quantitative scale for positive reactions (from + to +++) are generally accepted, the exact definition of the morphological criteria of this scale is still not uniform and there are also slight variations in the categorization between the different research groups [3].

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After arriving – not without difficulty – at an interpretation indicating contact sensitivity to a defined allergen, there is still one more issue to overcome, that is, demonstrating its relevance to the clinical situation. We will not herein consider the assessment of the relevance of the negative reactions, undoubtedly of significance to address the issue of false-negative responses. Moreover, doubtful reactions may be clinically relevant according to undeniable clinical criteria or follow-up testing. It could be worthwhile to ascertain whether doubtful (?) or weak (+) patch test reactions yield a significantly different relevance score than stronger and presumably more reliable positive patch test reactions.

Assessing the relevance of a positive patch test reaction is complex and involves many confounding factors. Evaluating the relevance of a reaction is the most difficult and intricate part of the patch test procedure, and is a challenge to both dermatologist and patient. The dermatologist's skill, experience, and curiosity are crucial factors. Little or no data on clinical relevance are provided in many clinical studies. Moreover, there is no consensus as to the definition of clinical relevance, how it should be scored, and how it should be assessed [4].

8.3 Past and Current Relevance

According to the ICDRG criteria, we consider that a positive patch test reaction is "relevant" if the allergen is traced. If the source of a positive patch test is not traced, we consider it as an "unexplained positive." We refer to as "current" or "present" relevance if the positive patch test putatively explains the patient's present dermatitis. Similarly, when the positive patch test explains a past clinical disease, not directly related to the current symptoms, we refer to this as past relevance. However, recurrent but discontinuous contact with an allergen can occur in some patients, making it difficult to discriminate between current and past relevance [5].

8.4 Scoring System

A modified relevance scoring system was proposed by Lachapelle [5] (Tables 8.1 and 8.2) for categorizing present and past relevance of positive patch tests reactions. The system codifies relevance scores from 0 to 3: 0 = not traced, 1 = doubtful, 2 = possible, and 3 = likely. Therefore, 16 combinations can be pondered for each individual case. The NACDG utilizes a similar scoring system using the terms "relevance possible," "relevance probable," and "relevance definite" [6].

Our goal in assessing relevance is to ascertain the putative responsibility of a particular allergen to the clinical circumstance. In this sense, the exposure to the incriminated allergen may explain the dermatitis entirely, that is, "complete relevance," but dermatitis with a multifactorial background frequently occurs. Contact sensitization may complicate

Table 8.1 The relevance scoring system of positive patch test reactions (from [5])

Past relevance (PR)	
PR 0	Not traced
PR 1	Doubtful
PR 2	Possible
PR 3	Likely
Current relevance (CR)	
CR 0	Not traced
CR 1	Doubtful
CR 2	Possible
CR 3	Likely

Table 8.2 Concomitant recording of past relevance (PR) and current relevance (CR) scores of positive patch test reactions: the 16 potential combinations (from [5])

PR0 CR3	PR0 CR2
PR1 CR3	PR1 CR2
PR2 CR3	PR2 CR2
PR3 CR3	PR3 CR2
PR0 CR1	PR0 CR0
PR1 CR1	PR1 CR0
PR2 CR1	PR2 CR0
PR3 CR1	PR3 CR0

dermatitis with an endogenous background, and other exogenous factors, such as irritants, may also play a significant role. Hence, we use the term "partial relevance" when the patch test-positive allergen contributed to or aggravated the dermatitis. It may be complicated, and often unattainable, to assess the relative influence of the different exogenous and endogenous factors on a given case of dermatitis.

8.5 Strategies

Therefore, determining the relevance of a positive patch test reaction principally relies upon the judicious interpretation of the clinical facts [7]. An allergen is clinically relevant if

1. We can establish the existence of an exposure
2. The patient's dermatitis is explainable (totally or partially) with regard to that exposure.

Establishing exposure involves appropriate knowledge of the patient's chemical environment and perseverance in pursuing lines of investigation. Relevance can be defined as the capability of an information retrieval system to select and redeem data appropriate to a patient's need [5].

8.5.1 Clinical History

The assessment starts with a comprehensive clinical history (Table 8.3). The patient should be questioned about occupational exposure, homework, and hobbies. Use of skin care products, topical medications, and protective measures should be covered. Emphasis should be made on possible exposures to the responsible environmental allergen or chemically related substances. Frequently it proves worthwhile to inform the patient in writing about the allergen producing the reaction, different names under which it is present, sources of

Table 8.3 Clinical data for the assessment of relevance

1. History of exposure to the sensitizer (present or past), specially seeking for intolerance
Occupational exposure
– Complete job description and materials
– Personal protective measures at work (gloves, safety shoes, garments, masks, barrier creams, after work creams)
– Other materials present in the working environment
Nonoccupational exposure
– Homework, hobbies
– Skin care products, nail and hair products, fragrances
– Pharmaceutical products (by prescription and over the counter)
– Personal protective measures. Use of gloves, detergents, etc.
– Jewelry and clothing
Indirect contact (skin care and other products of partner, fomites, etc.)
Seasonal related contact (plants and other environmental agents)
Photoexposure
Type of exposure: dose, frequency, site
Environmental conditions: humidity, temperature, occlusion, vapors, powders, mechanical trauma, friction, etc.
2. Clinical characteristics of the present dermatitis
Dermatitis area corresponding to the exposure site. Time of onset and characteristics of the initial lesions
Some morphologies suggest specific allergen
Clinical course (caused or aggravated by the exposure)
Time relationship to work. Effect of holidays and time-off work
3. History of previous dermatitis and other clinical events
– Past exogenous dermatitis with similar or different characteristics
– Previous patch testing
– Other endogenous skin diseases (atopic dermatitis, psoriasis, stasis, etc.)
4. Personal and family atopy and history of other family skin diseases

exposure, and chemical relatives. A complete review of the patient's history should provide insight into differentiating allergic contact dermatitis from other exogenous or endogenous dermatitis. This is crucial when dealing with multifactorial dermatitis.

8.5.2 Environmental Evaluation

Historical data should be confirmed and supplemented by a rigorous environmental evaluation, including research into the composition of products to which the patient has been exposed [8]. Identifying all possible sources of exposure in the subject's environment is an indispensable yet troublesome procedure involving many qualitative and quantitative estimations (Table 8.4). The intrinsic allergenic potential of the suspected agent as well as other physicochemical properties should be considered.

In addition, other exposure characteristics such as route of exposure, specific cutaneous site of contact, total contact area, dose, duration, and frequency of exposure are crucial factors in both the sensitization and elicitation phases of allergic contact dermatitis. Relevance scores and accuracy of the assessment are significantly improved by a comprehensive knowledge of the patient's chemical environment. Visiting the patient's workplace enables the physician to obtain a comprehensive picture of the real conditions at the working environment, bringing many details into clinical significance. Useful information about sources of allergens may be obtained from textbooks, "lists" of allergens, material safety data sheets, and manufacturers. Sometimes, chemical analysis of the supposedly causative product(s) is necessary. Simple

Table 8.4 Evaluation of exposure for the assessment of relevance

1. Clinical history (looking for all possible sources of exposure)
2. Workplace visit
3. Assessment of intrinsic sensitization potential of the substance
Data from predictive tests
Data from epidemiological studies
Structure/activity analysis
4. Additional physicochemical properties of the substance
Solvent properties, hygroscopicity, substantivity, wash and rub
Resistance to removal, etc.
5. Assessment of exposure parameters
Route of exposure
Specific site of contact and surface area
Dose
Duration
Frequency (periodicity) of exposure
Simultaneous exposure factors: humidity, occlusion, temperature, mechanical trauma
6. Look for cross-reacting and concomitant allergens
7. Information from "lists" of allergens, databases, product's manufacturer, etc.
8. Chemical analysis of suspected products

qualitative chemical spot tests performed by the clinician may orient the laboratory work [9]. Specialized techniques for allergen isolation and quantitative microanalysis are required in many cases. In some circumstances, it may be difficult to substantiate the presence of the allergen in the patient's environment. This may be due to the complexity in detecting certain allergens or to the insufficient knowledge about the composition of different products. As a consequence, the relevance scores for different allergens will vary; the easier the identification of the source of an allergen, the higher the relevance scores. Absolute proof of relevance is often unattainable, as it is frequently not known whether suspected products actually contain the implicated allergen in sufficient amount to elicit the dermatitis.

8.5.3 Further Correlations

The history of exposure to the sensitizer is essential, but not sufficient to establish the clinical relevance. To ascertain whether the exposure is relevant to the clinical dermatitis, the following factors should be considered:

1. Existence of a temporal relationship between the exposure and the clinical course of the dermatitis
2. Correspondence between the exposure and the clinical pattern (anatomical distribution) of the dermatitis

When actually present, these two conditions provide crucial diagnostic clues. Different confounding factors should be considered, that is, the contact with the allergen is not direct (e.g., airborne, ectopic, or connubial dermatitis), the clinical pattern of the dermatitis is nonspecific or has been modified (e.g., previous treatment, secondary infection, etc.); the dermatitis is multifactorial and factors other than contact allergy must also be considered as a cause (e.g., irritation, atopy, stasis, eczematous psoriasis) [7]. Often the clinical situation is intricate, demanding a systematic and critical approach.

8.5.4 Additional Investigations

Additional tests may prove valuable in establishing a definite causative relationship (Table 8.5). Tests with products to which the patients refer exposure and which supposedly contain the putative allergen should be performed. Patch testing with the unmodified product frequently produces negative results. This may be due to the following:

1. The concentration of the allergen in the final product is too low to elicit a positive patch test reaction, but sufficient to produce a clinical dermatitis through multiple exposures or special anatomic site exposure.
2. Certain environmental factors cannot be reproduced by the test procedure (e.g., humidity, friction, temperature, etc.)

Table 8.5 Testing procedures for the assessment of relevance

1. Testing with the suspected allergen(s)
Sequential patch testing
ROATs
– On normal skin
– On slightly damaged or previously dermatitic skin
2. Testing with products suspected to contain the responsible allergen
Patch testing (using suitable vehicle and appropriate concentration, frequently starting with highly diluted substances)
ROAT (similar as stated above, using proper vehicle and adequate concentration)
Use test (typical product use)
Testing in normal controls (if necessary) Testing in normal controls (if necessary)
3. Testing with product's extracts
Similar to 2. Testing with products suspected to contain the responsible allergen
4. Testing with cross-reacting allergens and products suspected to contain them.
Similar to 1. Testing with the suspected allergen(s)

Therefore, performing special tests, such as, tests with product's extracts, ROATs, PUTs may be indicated.

The positive patch test reactions for which clinical relevance cannot be established may represent false-positive results. But, much too frequently they represent true positive reactions wherein the patient fails to recall a significant exposure or the clinician does not retrieve the pertinent historical data, trace the responsible environmental exposure, or perform the appropriate tests.

8.6 Suggestions for Improved Evidence-Based Diagnosis of Relevance

As mentioned in the preceding sections, assessing relevance is not easy. Nevertheless, efforts should be undertaken to overcome those difficulties. Suggestions for improved evidence-based diagnosis of relevance are listed in Table 8.6.

Table 8.6 Suggestions for improved evidence-based diagnosis of relevance

1. Re-question the patient in light of the test results
2. Perform a worksite or home visit
3. Seek cross-reacting substances
4. Consider concomitant (and/or simultaneous) sensitization
5. Consider indirect, accidental, or seasonal contact
6. Obtain information about environmental allergens from lists and textbooks
7. Obtain information from the product's manufacturer
8. Perform chemical analysis of products
9. Perform sequential tests with the allergen and the suspected products (tests with extracts, ROATs, etc.)

In conclusion, "The relevance of a reaction is whether it explains any dermatitis in the patient. This is a pragmatic decision strongly influenced by the knowledge, inquisitiveness and determination of the dermatologist, and the time and resources available to him or her. In difficult cases, it is an interactive process of follow-up and reassessment" [10].

References

1. Ale SI, Maibach HI (2002) Scientific basis of patch testing. Dermatol Beruf Umwelt 50:43–50, 91–96, 131–133
2. Fischer TI, Hansen J, Kreilgård B, Maibach HI (1989) The science of patch test standarization. Immun Allergy Clin 9:417–434
3. Bruze M, Isaksson M, Edman B, Björkner B, Fregert S, Möller H (1995) A study on expert reading of patch test reactions: inter-individual accordance. Contact Dermatitis 32:331–337
4. de Groot AC (1999) Clinical relevance of positive patch test reactions to preservatives and fragrances. Contact Dermatitis 41:224–226
5. Lachapelle JM (1997) A proposed relevance scoring system for positive allergic patch test reactions: practical implications and limitations. Contact Dermatitis 36:39–43
6. Marks JG Jr, Belsito DV, De Leo VA, et al. (1998) North American Contact Dermatitis Group patch test results for the detection of delayed-type hypersensitivity to topical allergens J Am Acad Dermatol 38:911–918
7. Ale SI, Maibach HI (1995) Clinical relevance in allergic contact dermatitis. Dermatosen 43:119–121
8. Ale SI, Maibach HI (2001) Operational definition of occupational allergic contact dermatitis. In: Kanerva L, Menné T, Wahlberg J, Maibach HI (eds) Handbook of Occupational dermatology. Springer, Berlin, pp 344–350
9. Fregert S (1988) Physicochemical methods for detection of contact allergens. Dermatol Clin 6:97–104
10. Rycroft RJG (2002) Relevance in contact dermatitis. Contact Dermatitis 46(Suppl 4):39

The Atopy Patch Test in Atopic Dermatitis

9

U. Darsow and J. Ring

9.1 Introduction

Atopic dermatitis (AD) is a chronic inflammatory skin disease; its diagnosis is made by a combination of clinical features. AD is characterized by recurrent intense pruritus and a typically age-related distribution and skin morphology [1, 2]. The role of allergy in eliciting and maintaining the eczematous skin lesions has been controversial, partially due to the lack of specificity of the classic tests for IgE-mediated hypersensitivity. On the other hand, the most recent encouraging results of specific immunotherapy studies in AD patients with IgE-mediated allergies represent another line of evidence for the role of aeroallergens in AD. Among the allergens found to be relevant in AD, aeroallergens and food allergens (in children) are most important. As therapeutical consequences of the diagnosis of an allergy are based upon avoidance strategies, the relevance of (often multiple) IgE-mediated sensitizations in patients with AD must be evaluated.

Environmental substances such as aeroallergens produce flares in some patients with AD. Also, aeroallergen avoidance, especially with regard to house dust mites, can result in marked improvement of skin lesions [3]. Patients with AD often have elevated serum levels of IgE, which may correlate with the disease severity. The hypothesis is that Langerhans' cells bind and present "immediate type" allergens [4], which penetrate the impaired epidermal barrier in AD patients [5]. This concept is derived from studies showing IgE and IgE-binding structures on the surface of epidermal Langerhans' cells [5] together with mite allergen [6]. From atopy patch test biopsies, allergen-specific T cells have been cloned [7]. These T cells showed a characteristic TH2 (T-helper cell subpopulation) secretion pattern initially, while after 48 h a TH1 pattern was predominant. This same pattern is also found in chronic lesions of AD.

An epicutaneous patch test with allergens known to elicit IgE-mediated reactions and the evaluation of eczematous skin lesions after 48–72 h (Atopy Patch Test, APT) can be used as diagnostic tool in characterizing patients with aeroallergen-triggered AD. Several groups demonstrated that eczematous skin lesions can be induced in patients with AD by patch testing with aeroallergens. Patch testing of aeroallergens especially in patients with AD was first documented in 1982 by Mitchell et al. [8]. Because of variations in the applied methodology such as skin abrasion, tape stripping, and sodium laurylsulfate application for the enhancement of allergen penetration, differing percentages of positive APT results were obtained. No clear-cut correlations to skin prick test or specific IgE

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measurements could be obtained, and the sensitivity and specificity of experimental atopy patch tests with regard to clinical history remained unclear. We performed several studies to standardize the methods of APT on nonabraded skin and investigated the relation to clinical covariates.

9.2 Atopy Patch Test Technique

As a result of methodological studies [9–12], APTs with significant correlations to clinical parameters such as allergen-specific IgE or patients history are today performed with a very similar technique to conventional patch tests for the diagnosis of classic contact allergy. The standardization of aeroallergen APT is currently more advanced than that of food patch testing. In Europe, these efforts are coordinated by the European Task Force on AD (ETFAD)/EADV Eczema Task Force. Patch tests with lyophilized allergens, for example, from house dust mite (*Dermatophagoides pteronyssinus, D. pter*.), cat dander, grass pollen, are performed with a petrolatum vehicle (including a vehicle control). Patients should be in a state of remission of their dermatitis; the patch test is applied in large (12 mm) Finn Chambers (see Sect. 3.3.1) for 48 h on their back on nonabraded and uninvolved skin. We prefer to avoid any potentially irritating methods of skin barrier disruption such as tape stripping of the skin. Exclusion criteria and the possibility of contact urticaria should be considered (Table 9.1). Nonatopic volunteers and patients suffering from only allergic rhinoconjunctivitis presented no positive APT reactions with our methods in several studies. Allergens in petrolatum elicited twice as many APT reactions as allergens in a hydrophilic vehicle. Thirty-six percent of patients reacted to house dust mite *D. pter*., 22% to cat dander, and 16% to grass pollen. High allergen-specific IgE in serum is not a prerequisite for a positive APT, but 62% of patients with *D. pter*.-positive APT showed a corresponding positive skin prick test and 77% showed a corresponding elevated specific IgE. In other allergens, the concordance was even higher. Allergen concentrations of 500, 3,000, 5,000, and 10,000 PNU (protein nitrogen units) g in petrolatum were comparatively used in 57 patients [11]. In this study, the percentage of patients with clear-cut positive reactions was significantly higher in subjects with eczematous skin lesions in air-exposed areas (69%) compared to those without this predictive pattern (39%; $p = 0.02$). In the first

Table 9.1 Proposed exclusion criteria for atopy patch test

Antihistamines except astemizole	1 week
Systemic steroids	4 weeks
Topical steroids (test area)	1 week
UV-radiation	1 week
Acute eczema flare	3 weeks

Most of these criteria must be confirmed by further clinical studies

Table 9.2 Comparison of biological and PNU-based standardization of APT preparations: comparable concordance of APT with clinical allergen-specific history[a]

Standardization	7,000 (PNU/g)		200 (IR/g)	
Corresponding history	Yes	No	Yes	No
Dermatophagoides pteronyssinus	25	25	31	19
Cat dander	36	14	37	13
Grass pollen	39	11	39	11
Birch pollen	41	9	38	12
Total (%)	71	29	73	27

[a]*PNU* protein nitrogen units, *IR* index réactif (biological unit)
400 APTs in 50 patients with atopic dermatitis

group, the maximum reactivity was nearly reached with 5,000PNU/g. The data from a randomized, double-blind multicenter trial, involving 253 adult patients and 30 children with atopic eczema, were used to calculate a suitable APT allergen dosage [9–12]. Adults were tested with four concentrations, 3,000–10,000 PNU/g of *D. pter*., cat dander, grass pollen, and (in two study centers only; $n = 88$) with birch and mugwort pollen. A dose response for the APT could be obtained by McNemar statistics comparing with only questionable, only erythematous, or irritative reactions. The optimal allergen doses are in the range of 5,000–7,000 PNU/g. Simultaneously tested, the allergen doses of 7,000 PNU/g and 200 IR/g (biological unit; Index Réactif) of the most important aeroallergens in Europe showed comparable concordance with the patients' history, suggesting clinical relevance in another study on 50 patients with AD (Table 9.2).

9.3 Atopy Patch Test Reading

APT reactions are read after 48 and 72 h. Most reactions are seen after 48 h (Fig. 9.1), sometimes with decrescendo to 72 h. Using our methods, only very few reactions were seen as early as 24 h, but after tape stripping (see Sect. 7.1) followed by allergen application, there are more early reactions visible. APT was shown to give clinically relevant results with the ICDRG reading key for conventional patch testing [12, 13]. Consensus meetings of most groups performing APT for clinical use in Europe were held in Munich in 1997 and 1998. One result of these meetings was a consensus APT reading key for describing the intensity of APT reactions. This key has more options to describe the different morphology of positive APT reactions. After use in a multicenter trial in six European countries [14], it was modified to its actual version, 2003 (Table 9.3). A more important point in our opinion is to distinguish clear-cut positive reactions from negative or questionable ones, because only reactions showing papules or at least some degree of infiltration seem to be of clinical relevance.

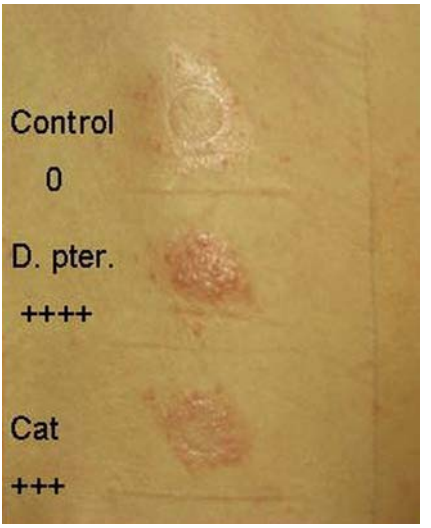

Fig. 9.1 APT reactions to different allergens after removal of Finn Chambers after 48 h. Clear-cut eczematous appearance with infiltration and spreading papules, partially with a follicular pattern. Control: petrolatum

Table 9.3 APT reaction grading key (European Task Force on Atopic Dermatitis ETFAD consensus), 2003

–	Negative
?	Only erythema, questionable
+	Erythema, infiltration
++	Erythema, few papules
+++	Erythema, many or spreading papules
++++	Erythema, vesicles

9.4 Atopy Patch Test Relevance, Patient Subgroups, and Pitfalls

To date, no "gold standard" of provocation for allergy diagnosis in AD exists. Thus, the history of allergen-specific exacerbation may be used as a parameter for clinical relevance. With regard to the clinically known phenomenon of "summer eruption" of AD, the validity of the APT was investigated [13]. Seventy-nine patients were tested with 10,000 PNU/g grass pollen allergen mixture in petrolatum and simultaneously with 10 mg dry, unprocessed pollen of *Dactylis glomerata* grass. The APT results were compared with history, skin prick tests, and specific corresponding IgE and the eczema pattern. This study showed significantly higher frequencies of positive APT reactions (with both methods used) in patients with a corresponding history of exacerbation of skin lesions during the grass pollen season of the previous year, or in direct contact with grass (75% with positive APT). Patients without this history showed significantly lower APT reactivity (16% with positive APT; $p < 0.001$). Depending on the APT procedure, the sensitivity referred to history of exacerbations during grass pollen season was 0.67–0.75, and the specificity was

0.84–0.90. The APT specificity exceeded the specificity of the classic tests of IgE-mediated hypersensitivity, which was 0.33 for skin prick test and radioallergosorbent test (RAST). On the other hand, the sensitivity of the classical methods was higher (0.92 for RAST and 1.0 for skin prick test). The results of unprocessed pollen leading to eczematous lesions on non-pretreated skin of AD patients with good correlation to history demonstrate that pollen may be involved in eczema flares in some patients. In the multicenter study with five aeroallergens, 10–52% of patients reported previous eczema flares after contact with at least one of the allergens. Again, APT results were significantly correlated with history, skin prick test, and specific corresponding IgE for *D. pter*., cat dander, and grass pollen ($p < 0.001$). Sensitivity and specificity of the APT were calculated for every allergen with regard to the corresponding history of eczema flares (Table 9.4) [12].

The APT with aeroallergens may provide an important diagnostic tool, as has been shown in two patient subgroups. In patients with an air-exposed eczema distribution pattern, positive APT reactions occurred at lower allergen doses compared with other patients with AD. Patients with an aeroallergen-specific history had significantly more positive APT reactions. The lower sensitivity but higher specificity of the APT compared to skin prick test or RAST favors the notion that the classical tests may have some value as screening tests, and specificity may be added by the APT. The APT does not replace the classical methods of diagnosis of IgE-mediated allergy. Questions remain open concerning the clinical relevance of positive APT results in patients with a negative history and discordant negative skin prick tests or RAST, as no gold standard exists for the provocation of eczematous skin lesions in aeroallergen-triggered AD. These questions may be answered only by conducting controlled studies using specific provocation and elimination procedures in patients with positive and negative APT results. However, this does not argue against the clinical use by dermato-allergists at this time point, since one must keep in mind that in many classic contact allergens the standardization and evaluation efforts have been less systematic. Still, these allergens are used for routine diagnosis in patch test clinics. Appropriate allergen-specific avoidance strategies are recommended in patients showing positive APT reactions. The diagnostic validity of APT in routine diagnosis of aeroallergen-triggered AD will be investigated in further controlled studies. The current status is summarized in a recent position paper [15].

Table 9.4 Sensitivity and specificity of different diagnostic methods with regard to patients history in 253 patients with AD[a]

Test	Sensitivity[b] (%)	Specificity[b] (%)
Skin prick	69–82	44–52
Specific IgE	65–94	42–64
APT	42–56	69–92
Data from [12]		

[a]Allergens: house dust mite *D. pter*., cat dander, grass pollen. APT shows a higher specificity, but lower sensitivity compared to skin prick test and measurement of specific IgE

[b]Referring to predictive history of eczema exacerbations in pollen season or in direct contact with allergen, excluding questionable cases, depending on allergen

Problems such as irrelevant positive or spreading APT reactions may occur in patients undergoing APT during an eczema flare, or if methods of abrasion of the stratum corneum are used (see Sect. 7.1). The issue of pharmacological influence on APT still holds many unanswered questions. An effect of pretreatment with 1% pimecrolimus cream on the APT was seen in a randomized, controlled, double-blind study enrolling 20 patients with AD and positive APT screening reaction to house dust mite *D. pter*., cat dander, grass or birch pollen [16]. For two weeks, patients applied twice daily pimecrolimus and vehicle control to marked fields on their backs. Then, APT was performed on both fields on the back. Including only patients with different readings ($n = 13$), stronger APT suppression of at least one ETFAD grade in the pimecrolimus area vs. intra-individual control was observed in 10 of these patients after 48 and 72 h ($p < 0.05$). Including all 20 subjects still showed a borderline significance compared with vehicle ($p = 0.0564$). Immunohistochemical analysis in two patients revealed an induction of interferon-γ in pimecrolimus-pretreated skin.

It was concluded that pimecrolimus treatment has a potential to suppress the development of lesions induced by aeroallergen exposure in patients with AD.

As the standardization of the high-molecular-weight allergens has some specific problems, a commercial provider of test substances with reproducible quality and major allergen content is desirable. However, to date such allergen preparations are not easily available. For research purposes, APT preparations in the preferred petrolatum base and 200 IR/g concentrations as listed in Table 9.2 have been provided by Stallergènes France (Stallergènes S.A., 6 Rue Alexis de Tocqueville, 92183 Antony Cedex, France, Tel.:+33 155 59 2000, Fax: +33 155 59 2068). Even more problems with allergen standardization are known for food APTs. APTs should be applied and read by dermato-allergologists.

References

1. Rajka G (1989) Essential aspects of atopic dermatitis. Springer, Berlin
2. Ring J (1991) Angewandte Allergologie, 2. Aufl. MMV Medizin Verlag, München
3. Tan B, Weald D, Strickland I, Friedman P (1996) Double-blind controlled trial of effect of housedust-mite allergen avoidance on atopic dermatitis. Lancet 347:15–18
4. Maurer D, Ebner C, Reininger B, Fiebiger E, Kraft D, Kinet JP, Stingl G (1995) The high affinity IgE receptor mediates IgE-dependent allergen presentation. J Immunol 154:6285–6290
5. Bieber T (1994) FCeRI on human Langerhans cells: a receptor in search of new functions. Immunol Today 15:52–53
6. Tanaka Y, Anan S, Yoshida H (1990) Immunohistochemical studies in mite antigen-induced patch test sites in atopic dermatitis. J Derm Science 1:361–368
7. van Reijsen FC, Bruijnzeel-Koomen CAFM, Kalthoff FS, Maggi E, Romagnani S, Westland JKT, Mudde GC (1992) Skin-derived aeroallergen-specific T-cell clones of TH2 phenotype in patiens with atopic dermatitis. J Allergy Clin Immunol 90:184–192
8. Mitchell E, Chapman M, Pope F, Crow J, Jouhal S, Platts-Mills T (1982) Basophils in allergen-induced patch test sites in atopic dermatitis. Lancet 1:127–130
9. Darsow U, Vieluf D, Berg B, Berger J, Busse A, Czech W, Heese A, Heidelbach U, Peters KP, Przybilla B, Richter G, Rueff F, Werfel T, Wistokat-Wülfing A, Ring J (1999) Dose response study of atopy patch test in children with atopic eczema. Pediatr Asthma Allergy Immunol 13:115–122

10. Darsow U, Vieluf D, Ring J (1995) Atopy patch test with different vehicles and allergen concentrations – an approach to standardization. J Allergy Clin Immunol 95:677–684
11. Darsow U, Vieluf D, Ring J (1996) The atopy patch test: an increased rate of reactivity in patients who have an air-exposed pattern of atopic eczema. Br J Dermatol 135:182–186
12. Darsow U, Vieluf D, Ring J for the APT study group (1999) Evaluating the relevance of aeroallergen sensitization in atopic eczema using the tool "atopy patch test": a randomized, double-blind multicenter study. J Am Acad Dermatol 40:187–193
13. Darsow U, Behrendt H, Ring J. (1997) Gramineae pollen as trigger factors of atopic eczema: evaluation of diagnostic measures using the atopy patch test. Br J Dermatol 137:201–207
14. Darsow U, Laifaoui J, Bolhaar S, Bruijnzeel-Koomen CAFM, Breuer K, Wulf A, Werfel T, Brönnimann M, Braathen LR, Dangoisse C, Blondeel A, Song M, Didierlaurent A, André C, Drzimalla K, Simon D, Disch R, Borelli S, Elst L, Devilliers A, Oranje AP, de Raeve L, Reiser K, Wollenberg A, Przybilla B, Roul S, Taieb A, Seidenari S, Wüthrich B, Ring J (2004) Atopy patch test with aeroallergens and food allergens in petrolatum: a European multicenter study. Allergy 59:1318–1325
15. Turjanmaa K, Darsow U, Niggemann B, Rancé F, Vanto T, Werfel T (2006) EAACI/GA2LEN position paper: present status of the atopy patch test – position paper of the section on dermatology and the section on pediatrics of the EAACI. Allergy 61:1377–1384
16. Weissenbacher S, Traidl-Hoffmann C, Eyerich K, Katzer K, Bräutigam M, Loeffler H, Hofmann H, Behrendt H, Ring J, Darsow U (2006) Modulation of atopy patch test and skin prick test by pretreatment with 1% pimecrolimus cream. Int Arch Allergy Immunol 140:239–244

Part II

Prick Testing

The Spectrum of Diseases for Which Prick Testing and Open (Non-Prick) Testing are Recommended

10

Patients Who Should be Investigated

J.-M. Lachapelle and H.I. Maibach

10.1 The Contact Urticaria Syndrome

The contact urticaria syndrome (CUS), first defined as a biological entity by Maibach and Johnson [1], comprises a heterogeneous group of inflammatory reactions that usually appear within minutes after cutaneous or mucosal contact with the eliciting agent and disappear within 24 h, usually within a few hours [2, 3]. The term 'syndrome' clearly illustrates the biological and clinical polymorphism of this entity, which may be either localized or generalized and may involve organs other than the skin, such as the respiratory or the gastrointestinal tract, as well as the vascular system, displaying a wide spectrum of clinical manifestations, ranging from mild erythema or itching to death.

Protein contact dermatitis (PCD), which could be considered a part of CUS, is described separately (see Sect. 10.2) for didactic (clinically related) reasons.

10.1.1 Clinical Symptoms and Stages of CUS

The symptoms can be classified according to morphology and severity (Table 10.1). In the mildest cases, there are only subjective symptoms (invisible contact urticaria). These are reported as itching, tingling or burning sensations, without any objective change, or just a discrete erythema occurs. In daily practice, these reactions are seen from cosmetics [4] and from fruits and vegetables.

Wheal and flare at the contact area is the prototype of contact urticaria (Fig. 10.1–10.3), while generalized urticaria following a local contact is less common (Fig. 10.4).

Extracutaneous symptoms may also occur as part of a more severe reaction and may include rhinoconjunctivitis (Fig. 10.4), asthmatic attack and orolaryngeal or gastrointestinal manifestations. Finally, anaphylaxis may occur as the most severe manifestation of CUS.

Urticarial lesions of CUS do not differ clinically from those observed in common urticaria. Itching erythematous macules develop (at the site of contact) into wheals consisting of pale-pink, oedematous, raised skin areas often with a surrounding flare (Fig. 10.3).

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Table 10.1 The contact urticaria syndrome (CUS): staging by symptomatology (from [3])

Stage 1	Localized urticaria (Fig. 10.1–10.3)
	Dermatitis
	Nonspecific symptoms (itching, tingling, burning, etc.)
Stage 2	Generalized urticaria
	Cutaneous and extracutaneous reactions
Stage 3	Rhinoconjunctivitis (Fig. 10.4)
	Orolaryngeal symptoms
	Bronchial asthma
	Gastrointestinal symptoms
Stage 4	Anaphylactic symptoms

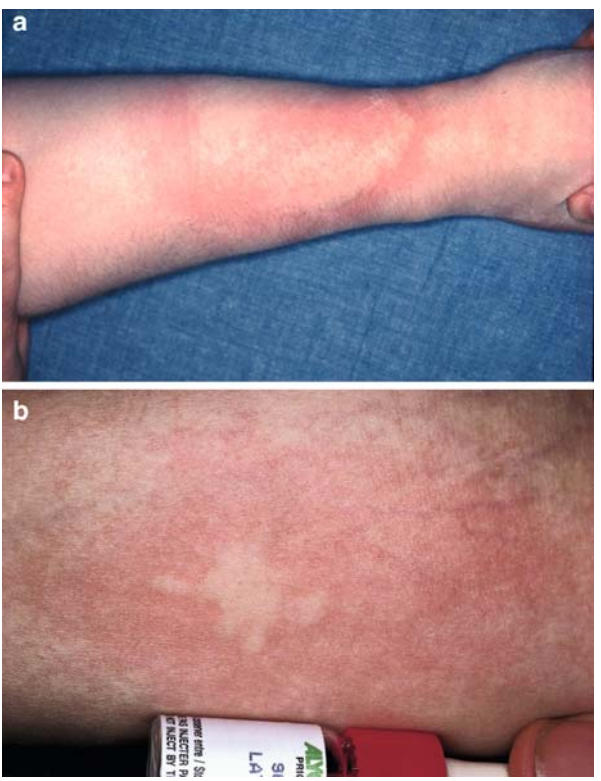

Fig. 10.1 Immunological contact urticaria. (**a**) To latex proteins (from a latex glove); (**b**) Positive prick test reaction to latex

They appear in various numbers and sizes, ranging from a few millimeters (Fig. 10.2) to lesions covering a large area, corresponding to the site of contact (Fig. 10.1a). These clinical variants are well illustrated in contact urticaria to rubber latex, a clinical entity that has exploded (in terms of numbers of cases) during the last two decades.

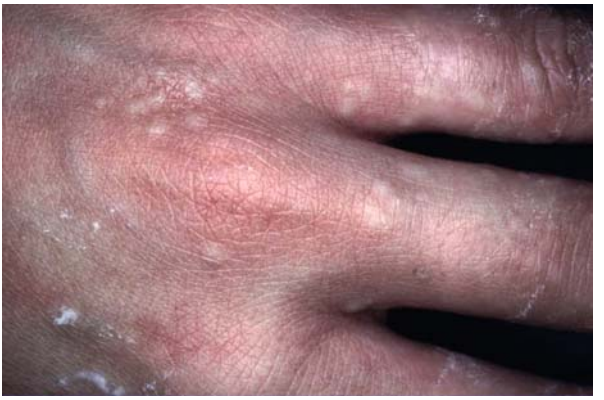

Fig. 10.2 Immunological contact urticaria of the hands from internally powdered latex gloves. The dorsa are dotted with small urticarial papules

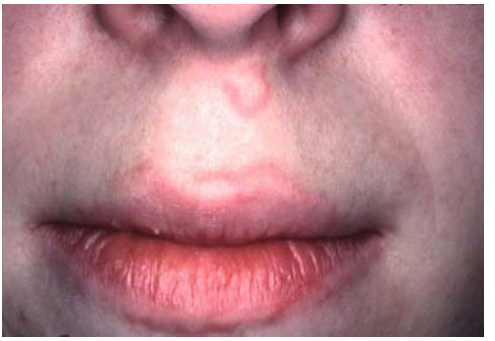

Fig. 10.3 Immunological contact urticaria to vanilla in a child sucking ice cream. The lesions extend not only on the lips, but also to the perioral area

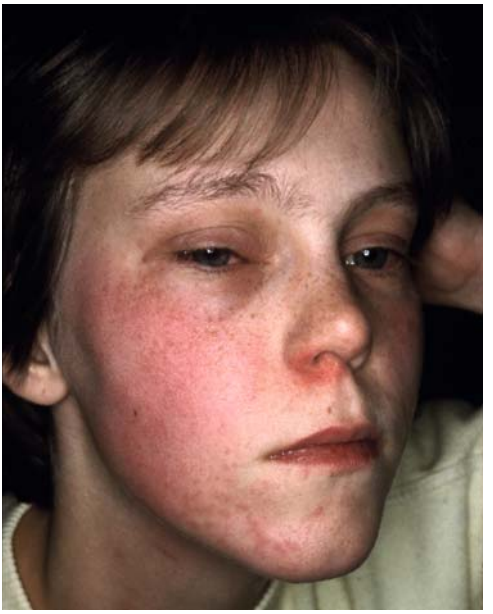

Fig. 10.4 Airborne immunological contact urticaria of the face caused by the dispersion of cornstarch particles with a high latex protein content in a female operating theatre nurse presensitized by latex gloves. Urticarial plaques on the cheeks, eyelids and nostril areas, associated with conjunctivitis and allergic rhinitis

10.1.2 Etiology and Mechanisms of CUS

The mechanisms underlying immediate-contact reactions are divided into two main types: immunological and non-immunological. However, there are substances that cause immediate contact reactions whose mechanisms (immunological or not) remain unknown [5, 6].

10.1.2.1 Immunological Contact Urticaria

Immunological contact urticaria (ICU) is a type I hypersensitivity immunological reaction in individuals who have previously contacted the causative agent and synthesized specific immunoglobulin E (IgE) antibodies against this agent. IgE molecules react with IgE receptors on the mast cells, basophils, eosinophils, Langerhans' cells and other cells. Eventually, allergen penetrating through the skin or mucosal membrane will react with the two adjacent IgE molecules bound to the cell membranes of the mast cells. Within minutes, histamine, neutral proteases and proteoglycans are released from the mast cells, resulting in an immediate skin response. The allergen–IgE reaction also leads to the synthesis of leucotrienes, prostaglandins and platelet-activating factors in the cell membranes of the activated mast cells. The mast cells also release chemotactic factors attracting eosinophils and T cells from the vessels into the dermis.

Immunological-type agents are confirmed by specific positive radioallergosorbent tests (RASTs) and by negative tests on control subjects.

The number of substances that have been reported to produce ICU is protean. Most examples refer to proteins (also responsible for protein contact dermatitis; see Sect. 10.2). Proteins can penetrate through normal human skin; any disorder in skin barrier function enhances protein penetration. This is particularly true in atopic dermatitis [7]. Proteins are of vegetal or animal origin. The list has no limitation, as recent reports from the literature regularly provide additional urticariogens. An extensive repertoire of most common animal, plant or other derivates (natural products) proteins has been proposed recently [3]. Rubber latex is by far the commonest cause of ICU; several proteins has been incriminated. Because of its major importance, a special section has been devoted to latex contact urticaria (see Sect. 10.1.3).

Apart from proteins, several non-protein allergens are able to provoke ICU. Among others, food-derived and food-associated materials such as preservatives, flavourings, stabilizers, emulsifiers and antioxidants also responsible for allergic contact dermatitis are often quoted [3]. Ammonium persulphate and other persulphates used in hair bleaches [8] represent the most common cause of ICU in hairdressers (ammonium persulphate could also act as a non-immunological urticariogen).

Special attention must be paid to the occurrence of ICU related to a vast number of cosmetic products [9]. Very common are the reactions induced by cinnamic aldehyde or balsams of Peru. Many cases are underdiagnosed or misdiagnosed due to the lack of knowledge in the matter.

Another field of interest is ICU related to topical drugs. Chlorhexidine is often quoted as a major urticariogen, leading in some cases to an anaphylactic shock [10]. Other examples include PVP-I, ethanol, Emla cream (lidocaine plus prilocaine), cephalosporins, rifampicin, aminosides, diphenylcyclopropenone (diphencyprone), penicillins and many others.

In all these circumstances, prick testing is the investigation tool to be used in order to trace aetiological factors responsible for ICU.

10.1.2.2 Non-Immunological Contact Urticaria (NICU)

Non-immunological contact urticaria (NICU) occurs in subjects not sensitized to the contactant, that is almost any normal subject. The mechanism of action is the result of a direct release of vasoactive substances, which causes a localized response. Prostaglandins are mediators in the reaction (to at least benzoic acid, sorbic acid and methylnicotinate). The NICU is often (but not always) limited to erythematous macules without oedema rather than a real wheal-and-flare reaction. In practice, the intensity of reactions depends mainly on the duration of exposure, the concentration of the contactant and other factors, such as rubbing or scratching. The reaction usually remains localized, and systemic reactions are probably not evoked. Substances capable of producing NICU are not proteins, but low-molecular-weight molecules that easily cross the skin barrier. Responsible agents include plants, animals or chemical substances. Many of the chemical substances involved are used as flavourings, fragrances and preservatives used in the cosmetic, pharmaceutical and food industries.

NICU from various plants is not uncommon. In many cases, it is linked with the release of calcium oxalate and saponins into the skin. The most common example is NICU related to the sting of a nettle (*Urtica dioica*). Another typical example is NICU provoked by *Agave americana* ('mal de agaveros' in Mexico), coexisting sometimes with purpuric dermatitis [11].

As mentioned previously, prick testing reproduces experimentally NICU reactions at the site of application. Nevertheless, prick testing is not primarily aimed to trace NICU contactants, which are well-known urticariogens. Prick testing provides positive results in almost all normal individuals.

10.1.2.3 Contact Urticaria of Uncertain Mechanism

This category is considered provisional [3], as it implies uncertain mechanism(s). It will be probably more precisely defined when adequate research will be conducted in this field. In some instances, the reaction resembles that of ICU, but no specific IgE can be demonstrated in the patient's serum or in the tissues. It is possible that there are other immunological mechanisms in addition to the IgE-mediated ones. Specific IgG and IgM might activate the complement cascade through the classical pathway. A classic example is provided by ammonium persulphate. There have been several reports of both localized and generalized contact urticaria, as well as respiratory symptoms and even anaphylactoid reactions. Although the clinical symptoms correspond to an IgE-mediated reaction, IgE

antibodies against ammonium persulphate have been demonstrated only in rare cases [8]. Similar considerations are applicable to formaldehyde.

Prick testing also detects the etiological agent(s) in cases of contact urticaria of uncertain mechanism. In such cases, the result of prick testing may also be positive in some control subjects.

10.1.3 Contact Urticaria to Natural Rubber Latex

Natural rubber latex refers to products derived from or containing the milky fluid, or natural latex, produced by the tropical rubber tree, *Hevea brasiliensis*, a tree originating from the Amazon basin.

IgE-mediated natural rubber latex hypersensitivity to the constituent proteins of natural rubber latex is now recognized as a health problem of growing importance [12–14]. While the prevalence of natural-rubber latex sensitization among the general population is estimated less than 1%, 3–17% of health care workers and up to 50% of spina bifida patients are sensitized. Other high risk groups have also been identified: patients with a history of multiple surgical interventions, atopic individuals, people working in factories when natural rubber latex are manufactured, patients suffering from hand dermatitis and patients presenting allergies to certain plant-derived food, especially 'tropical' fruit. Natural rubber latex gloves (mainly but not exclusively surgical ones) represent the most common source of skin contact allergy, but many other rubber items (e.g. rubber balloons) can also be incriminated.

Natural latex is a complex mixture for which allergenicity depends on botanical, chemical, immunological and epidemiological variables. Today, several natural latex allergens have been identified and characterized at both the molecular and the immunological level. Most of these proteins are present in the laticifer cells. In addition, several structural proteins have been described as allergens. Among these numerous proteins recognized as allergenic contactants, some are considered more important, for example rubber elongation factor (Hev b1), rubber elongation factor homologue (Hev b3), Hev b5, Hev b6.01, Hev b6.02, Hev b6.03 and Hev b13, but many others may be of interest. Special attention is paid nowadays to recombinent latex allergens [15].

Diagnosis of IgE-mediated hypersensitivity to natural rubber latex is based on (a) a clinical history of CUS (see Sect. 10.1.1) and (b) the confirmation of IgE-mediated reaction by appropriate reactions. Skin prick testing (see Chap. 11) is extensively used throughout the world and provides reasonably good sensitivity and specificity. The alternative (usually considered less performant) is the assessment of specific IgE antibodies to latex (RAST). The sensitivity of CAP RAST has recently been improved by adding Hev b5 to the solid phase. False-positive results may be due to cross reactivity between the major allergen hevein (Hev b6.02) and class I chitinases present in various fruits like avocado and banana [16].

Natural rubber latex hypersensitivity has become so important nowadays that, in some clinics, prick testing with natural rubber latex extract is recommended as a routine additional test to the international standard series of patch tests; however, some authors reserve its use only to well-defined circumstances, for example when clinical history is evocative or before surgery or other medical interventions when increased risk of contact is evident.

Although prevention is sufficient to reduce sensitization, prolonged avoidance is needed to prevent resensitization or adverse reactions on re-exposure.

In one study [17], sublingual immunotherapy seems to offer promising results.

10.2 Protein Contact Dermatitis

Protein contact dermatitis (PCD) is a complex entity, originally described by Hjorth and Roed-Petersen [18] and accepted as a well-defined syndrome [3, 19]. Its most usual clinical presentation is hand dermatitis (described first among food handlers) that may resemble an ordinary chronic or recurrent contact dermatitis, either of the delayed allergic variety or of the chronic irritation. However, redness, wheals and sometimes microvesicles appear as symptoms of contact urticaria, usually within an hour after skin contact with the causative agent. These immediate changes usually appear only in skin sites previously affected by eczematous dermatitis.

Most often, it is not possible to depict the presence of an immediate component in hand dermatitis on the basis of the clinical examination; therefore, a detailed clinical history is essential. A distinction feature from classic allergic contact dermatitis is the fact that the patient complains of immediate symptoms such as burning, itching or stinging accompanied by redness, swelling or vesiculation when handling the allergen. To a large extent, these symptoms resemble those of skin irritation and can be misinterpreted if the patient is not questioned properly. Lesions of PCD are mainly located on hands and forearms. It has been advocated that PCD could represent a mixed situation, including both immediate (type I) and delayed (type IV) hypersensitivity reactions to allergenic proteins. Moreover, skin irritation by contactants could intervene as an additional cause.

It appears clearly from recent studies that PCD occurs more frequently in patients suffering from atopic dermatitis than in non-atopics. The impairment of the barrier function in atopics (see Chap. 9) plays an important role for an increased penetration of proteins into the skin. Some authors have coined the term ‘extrinsic atopic dermatitis’, which means that atopic dermatitis of the hands is mainly related to proteins in contact with the skin [20]. Nevertheless, it must be kept in mind that, despite these advances in our knowledge of AD, an atopic background is not a prerequisite in PCD. In other words, PCD may occur without any personal or family history of atopy.

Clinical variants do exist:

- *Fingertip dermatitis.* Mainly but not exclusively of the ‘gripping type’ (see Sect. 2.5.2). Itching is often present and may be distinctive.
- *Chronic paronychia.* This is a common variant (Fig. 10.5 and 10.6a) mainly observed in patients who have chronically wet hands [21]. Wet foods are a combined source of factors, where the food may be an irritant or an allergic contactant. It is therefore predominantly a disease of domestic workers and fishmongers (Fig. 10.6b). Bacterial and/or *Candida albicans* infection may be associated in some cases (Fig. 10.2).

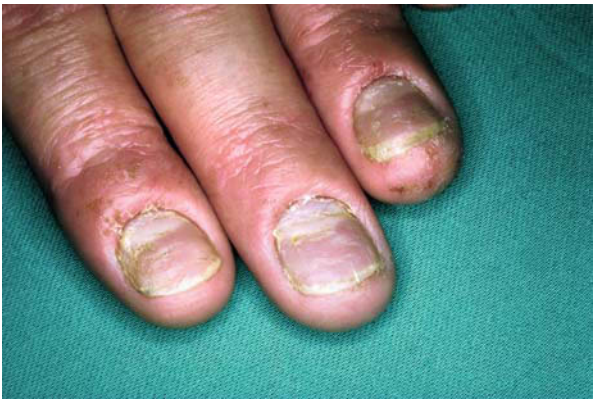

Fig. 10.5 Occupational immune protein contact dermatitis to food allergens in a food handler. Striking paronychia and nail changes (yellowish onycholysis)

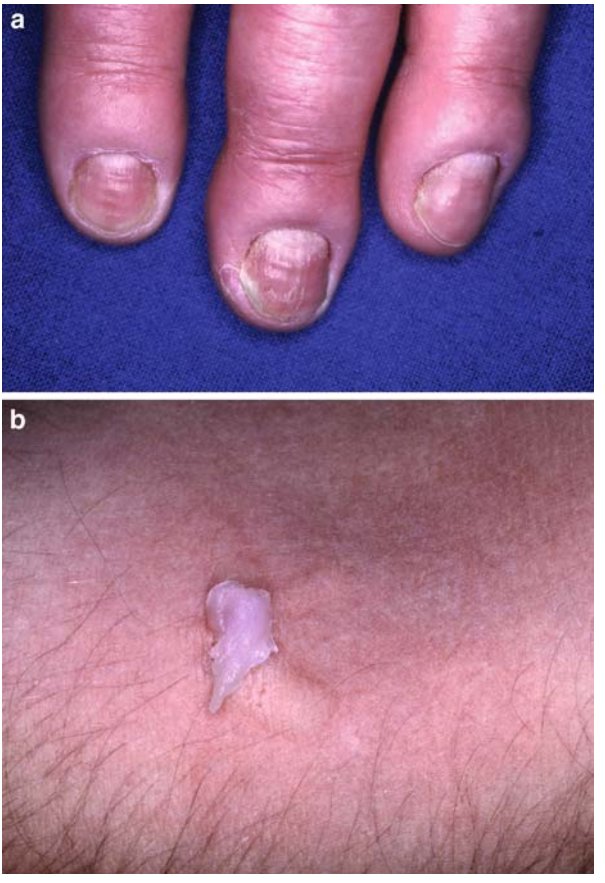

Fig. 10.6 Occupational immune protein contact dermatitis to monkfish in a fishmonger. (**a**) Striking paronychia and nail changes (irregular striae); (**b**) positive prick test to monkfish. Reading at 30 min

Table 10.2 Clinical facets of protein contact dermatitis (PCD)

Chronic dermatitis, mainly located on the hands and/or forearms, sharing common features with irritant and/or allergic contact dermatitis. Atopic background may be present. In that case, differential diagnosis with atopic dermatitis of the hands may be subtle and imprecise.
Urticarial symptoms (contact urticaria) are usually present, but they are often underestimated, since they are transient (acute onset after contact) and partly occulted by underlying dermatitis.
A variant of PCD is fingertip dermatitis, mainly the 'gripping' form (i.e. involving thumb, index and medius of one or both hands). Itching may be a distinctive feature.
Chronic paronychia (Figs. 10.5–10.6a)

The various clinical facets of PCD are listed in Table 10.2.

Prick testing (and its variants; see Chap. 11) is the key tool in the aetiological diagnosis of PCD. The atopy patch test (see Chap. 9) could be an additional diagnostic procedure. This approach has to be linked with conventional patch testing, meaningful for a complete evaluation of each individual case.

References

1. Maibach HI, Johnson HL (1975) Contact urticaria syndrome. Contact urticaria to diethyltoluamide (immediate type hypersensitivity) Arch Dermatol 111:726–730
2. von Krogh G, Maibach HI (1982) The contact urticaria syndrome. Semin Dermatol 1:59–66
3. Ale SI, Maibach HI (2000) Occupational contact urticaria, Chapter 24. In: Kanerva L, Elsner P, Wahlberg JE, Maibach HI (eds) Handbook of Occupational Dermatology. Springer, Berlin, pp 200–216
4. Collet E, Bonniaud P (2006) Hypersensibilité immédiate aux cosmétiques et aux médicaments topiques. Rev Fr Allergie Clin Immunol 46:248–251
5. Amin S, LahtiI A, Maibach HI (1997) Contact urticaria syndrome, CRC Press, Boca Raton, 316 p
6. Bashir SJ, Maibach HI (2006) Contact urticaria syndrome. In: Chew AL, Maibach HI (eds) Irritant Dermatitis. Springer, Berlin, pp 63–70
7. Weidinger S, Rodriguez E, Stahl C, Wagenpfeil S, Klopp N, Illig T, Novak N (2007) Filaggrin mutations strongly predispose to early-onset and extrinsic atopic dermatitis. J Invest Dermatol 127:724–726
8. Aalto-Korte K, Mäkinen-Kiljunen S (2003) Specific immunoglobulin E in patients with immediate persulfate hypersensitivity. Contact Dermatitis 49:22–25
9. Vigan M (2007) Urticaire de contact aux cosmétiques. In: Progrès en Dermato-Allergologie, Paris 2007, John Libbey Eurotext, Paris pp 17–34
10. Krautheim AB, Jermann THM, Bircher A (2004) Chlorhexidine anaphylaxis: a case report and review of the literature. Contact Dermatitis 50:113–116
11. Genillier-Foin N, Avenel-Audran M (2007) Dermatite purpurique de contact au suc d'Agave americana. Ann Dermatol Venereol 34:477–478
12. Ownby DR (2002) A history of latex allergy. J Allergy Clin Immunol 110(Suppl.2): S3–S14
13. Reunala T, Alenius H, Turjanmaa K, Palosuo T (2004) Latex allergy and skin. Curr Opin Allerg Clin Immunol 4:397–401

14. Tennstedt D, Baeck M (2007) Allergie au latex en 2007, faut-il encore y penser? In; Progrès en Dermato-Allergologie, Paris 2007, John Libbey Eurotext, Paris, pp 35–56
15. Raulf-Heimsoth M, Brüning T, Rihs HP (2007) Recombinant latex allergens. Rev Fr Allergol 47:123–125
16. Turjanmaa K, Palosuo T, Alenius H, et al (1997) Latex allergy diagnosis: in vivo and in vitro standardization of a natural rubber latex extract. Allergy 52:41–50
17. Nettis E, Colanardi AL, Soccio AL, Marcandrea M, Pinto L, Ferrannini A, Tursi A, Vacca A (2007) Double-blind, placebo-controlled study of sublingual immunotherapy in patients with latex-induced urticaria: a 12-month study. Br J Dermatol 156:674–681
18. Hjorth N, Roed-Petersen J (1976) Occupational protein contact dermatitis in food handlers. Contact Dermatitis 2:28–42
19. Janssens V, Morren M, Dooms-Goossens A, Degreef H (1995) Protein contact dermatitis: myth or reality ? Br J Dermatol 132:1–6
20. Hennino A, Vocanson M, Rozières A, Nosbaum A, Gunera-Saad N, Goujon C, Bérard F, Nicolas JF (2007) La dermatite atopique, un eczéma de contact aux allergènes protéiques? In: Progrès en Dermato-Allergologie, Paris 2007, John Libbey Eurotext, Paris, pp 7–15
21. Tosti A, Buerra L, Mozelli R (1992) Role of food in the pathogenesis of chronic paronychia. J Am Acad Dermatol 27:706–710

The Methodology of Open (Non-Prick) Testing, Prick Testing, and its Variants

11

J.-M. Lachapelle and H.I. Maibach

11.1 Open (Non-Prick) Testing

An introductory remark needs some explanation. When immunological contact urticaria (ICU) (see Sect. 10.1.2) or protein contact dermatitis (PCD) (see Sect. 10.2) is suspected, it is considered that prick testing is the "key" diagnostic tool to detect the incriminated allergens [1, 2]. Nevertheless, some dermatologists are reluctant to practice prick testing, particularly in cases of ICU when general symptoms have been mentioned by the patient (see Sect. 10.1.1). This attitude is fully justified and, in those cases, it is wise to start the investigation with an open test [3]. A first approach is to use the open test as such (see Sect. 7.2); another way to test is to use a Chamber Test, similar to that recommended in the atopy patch test (see Chap. 9). The results of such investigations need to be carefully interpreted, as they can lead to false-positive reactions.

Therefore, it is mandatory that, when in doubt, control subjects are tested in a similar way to avoid misinterpretation, due to irritant reactions [3].

Some variants can be adopted. In the rub test, the suspected substance is gently rubbed onto the slightly affected or irritant skin [3]. Rubbing may enhance the reactivity compared to the open application test. Here again, potential irritant reactions have to be taken into account.

Oranje et al. have developed a modified test to be used especially in cases of suspected food contact allergy, the so-called skin application food test (SAFT). 0.8 mL of liquid food or a solid piece of food is placed on a 4-cm^2 gauze and fixed onto the back skin with an acrylic tape [4]. The test can also be performed by using 12 mm Finn Chambers (see Sect. 3.3.1).

The results are followed up every 10 min, the maximal occlusion time being 30 min. According to the authors, the test results are highly reproducible [4].

11.2 Prick Test: Technical Modalities and Reading

The prick test is usually the most convenient test method for detecting immunoglobulin E (IgE)-mediated allergy. Large numbers of commercial prick test allergens are available; self-made allergens can also be used (see Sect. 11.7). They are kept in a refrigerator.

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11.2.1 Technique of Puncture

Drops of allergen solutions are applied to the volar aspect of the forearm or to the upper part of the back. The flexures of the elbows must be avoided, because this may give rise to not easily readable reactions, either positive or negative. Other skin sites are not convenient as well. An important point concerns the distance between the individual prick tests. These are applied ideally 3–5 cm apart to avoid overlapping of reactions at reading. If such a distance is not respected, difficulties in correct reading are obvious and no definite conclusions can be drawn. This mistake in technology happens too often, even among well-trained clinicians.

When drops of allergen solutions are applied to the skin, they are pierced with a special lancet (e.g., the Dome-Hollister-Stier prick test lancet, the plastic lancet Stallerpoint Stallergènes, the metallic lancet Allerbiopoint Allerbio Laboratories).

Stallerpoint and Allerbiopoint are used in many European clinics (Fig. 11.1a).

Stallerpoint is a polymethacrylate lancet (length: 1.1 mm; four microscopic furrows allow a progressive and reproducible penetration of allergens into epidermis; presenting itself as a blister of ten sterile disposable lancets). The lancet conforms to the European Directive N93/42/CEE.

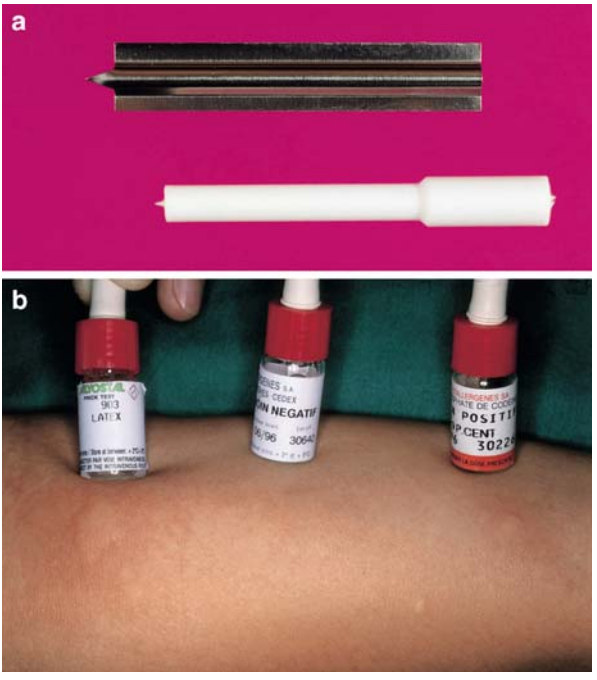

Fig. 11.1 Prick testing. (**a**) Prick test lancets; (**b**) Positive prick test to latex: positive and negative controls

Allerbiopoint is a stainless steel lancet (length: 1.1 mm; penetration angle 45°; presenting itself as a blister of ten sterile disposable lancets). The lancet conforms to the European Directive N93/42/CEE.

Puncture is made by gentle pressure; some authors, when puncturing, exert a slight rotation movement to ensure better penetration of the allergen. No bleeding may occur.

11.2.2 Control Solutions

Prick testing of allergens needs the concomitant use of controls, positive and negative.

11.2.2.1 Positive Controls

- Histamine chlorhydrate solution (10 mg ml^{-1}) to measure direct reactivity to histamine
- Codeine phosphate solution (9%) to verify in each individual the aptitude for mast cell degranulation

In the dermato-allergology unit, at Louvain University, Brussels, Belgium, both controls are always performed. It is our experience that positive prick tests to codeine phosphate are very uniform in all patients (with some exceptions), whereas positive prick tests to histamine chlorhydrate are more variable from patient to patient (within acceptable limits).

11.2.2.2 Negative Control

Saline and/or the vehicle of the allergens is used as a negative control.

11.2.3 Reading Time

After 15 min, the allergen and control droplets are wiped off with soft paper tissue. Conventional time reading is 15–20 min, as we are evaluating an immunological immediate-type I reaction (Fig. 11.1b).

11.2.4 Reading Prick Test Results

Reading prick test reactions (Fig. 11.1b) needs careful evaluation and interpretation, taking into account several parameters of prime importance.

- The negative control ought to be negative; if positive, it raises questions about the reading of allergen prick tests. Its main interest is therefore to detect false-positive reactions.
- Wheal and flare reactions to positive controls, which appear around the piercing usually in minutes, are measured in terms of diameters and surface areas (Fig. 11.1b).
- Allergen prick test results are usually expressed as the mean of the longest diameter of the wheal and the largest diameter perpendicular to it.
- Reactions greater than 3 mm and at least half of that produced by histamine are regarded as positive [5, 6]. Reactions smaller than those produced by histamine may not be clinically significant.
- If the patient has dermographism (factitious urticaria), skin piercing produces usually small (1–2 mm) wheals, which may make the interpretation of the results very difficult.

There is a clear-cut difference in terms of readings between patch testing and prick testing. Patch testing is a codified procedure that does not imply any control, whereas prick testing is invariably submitted to controls either positive or negative in order to achieve correct interpretation of the results.

The final goal in prick testing is to assess (either past or current) the relevance. The practical means to conclude "likely," "possible," "doubtful," or "not traced" relevance can be copied from those described in Chap. 8.

11.2.5 Medicaments and Prick Testing

Caution must be taken when prick testing patients treated with antihistamines. Antihistamines of the so-called third generation, extensively used nowadays, abolish the immediate reactivity of the skin usually for 1–3 days. This concerns cetirizine, loratadine, fexofenadine, ebastine, mizolastine, and the newcomers desloratadine and levocetirizine. Prick testing can be performed 3 days after stopping treatment. Longer wash-out periods are needed with ketotifen (15 days) and astemizole (4 weeks).

Oral methylprednisolone more than 8 mg daily and equivalent doses of other corticosteroids may also weaken the immediate reactivity of the skin. Other drugs such as non-steroidal anti-inflammatory drugs as well as topical application of corticosteroids do not affect prick test results significantly.

11.2.6 False-Negative Reactions

False-negative reactions may occur. Interpretation of results needs caution:

- When reactions to positive controls are weak or negative.
- When time reading is inadequate.
- When patients are treated with antihistamines or oral corticosteroids (see Sect. 11.2.5).

11.2.7
False-Positive Reactions

False-positive reactions may occur. Interpretation of results needs caution:

- When reactions to negative controls are positive.
- When patients have dermographism.
- When all prick test sites react positively in a similar way.

11.2.8
Prick Tests in Children and Babies

Prick tests can be performed, if suitable, in children and babies, whose skin reactivity is similar to that observed in adults.

11.3
Prick-by-Prick Test

A modification of the prick test is the prick-by-prick test, used especially for prick testing with fresh foodstuffs, for example, fruits and vegetables [7].

A piece of food is pricked with an ordinary prick test lancet, immediately after which the skin is pricked with the same lancet. This fresh food prick testing is handy and superior to prick testing with commercial food allergens.

11.4
Scratch Test

This previously common method for detecting immediate allergy is still used when only nonstandardized allergens are available. If the prick test is used for testing with nonstandardized allergens, for example, flours, edible roots, vegetables, and fruits, skin infections and other untoward inflammatory processes can be produced. A scratch of approximately 5 mm long is made with a blood lancet or venipuncture needle, and bleeding is avoided. The back and arms are the preferred test sites. Small amounts of allergen solution are applied to the scratches, and the results are read 15–20 min later (Fig. 11.2). Powdered allergens are mixed with a drop of physiological saline or 0.1N NaOH on the scratch. Histamine chlorohydrate (10 mg ml^{-1}) is the positive and saline or 0.1N NaOH is the negative control. Reactions equal to or greater than those from histamine are usually clinically significant.

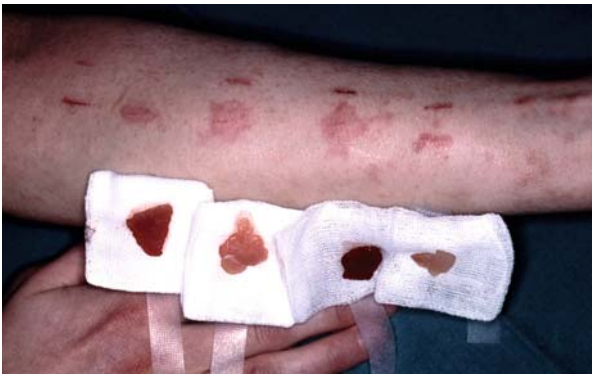

Fig. 11.2 Positive scratch tests to different meats in a butcher. Reading at 40 min

11.5 Scratch-Chamber Test

Certain foodstuffs, for example, edible roots, fruits, and vegetables, tend to dry out too quickly when applied to a scratch. Covering the scratch with a large (inner diameter: 12 mm) Finn Chamber (see Sect. 3.3.1) prevents drying out of the test material [8]. The positive and negative controls and the way results are read are the same as for the scratch test [9].

11.6 Comparative Indications of Open (Non-Prick) Testing, Prick Testing, and Other Related Tests

The indications for which the use of prick tests and other related tests [10] are advised are listed in Table 11.1.

11.7 Intradermal Testing

Nowadays, as far as the etiological diagnosis of CUS or PCD is concerned, prick testing and its variants do not have to be complemented by intradermal testing. Intradermal testing with rubber latex extracts has been practiced in some studies, but it is no longer advised. Therefore, in practice, the use of intradermal testing is limited to investigations in relation with drug eruptions (see Chap. 12).

Table 11.1 Comparative indications of prick tests and of other related tests

Test	Indications
Open (non-prick) testing	For IgE-mediated allergy; as a first step (see Sect. 11.1), when prick testing is not advisable, especially in patients at stages 2, 3, and 4 of CUS (see Sect. 10.1.1)
Prick test	For IgE-mediated allergy; especially for standardized allergen solutions
Prick-by-prick	Recommended for testing with fresh foods
Scratch test	For IgE-mediated immediate allergy; nonstandardized allergen can also be used
Scratch-chamber test	Especially for testing foodstuffs

11.8 Prick Testing: Allergens of Interest for Skin Problems

Many categories of standardized allergens are available for prick testing; there is no standard series (as compared with patch testing). Among the long list quoted in catalogues, some are of greater importance as far as skin problems are concerned. A few series are listed below.

11.8.1 Latex

Natural rubber latex glove extracts have been widely used as skin prick test allergens. However, since the allergen content of natural rubber latex gloves varies considerably, it is of extreme importance to dispose of the most suitable glove for test material. An updated list on the allergenicity of natural rubber latex gloves is available from the National Agency for Medicines, Medical Device Centre (P.O. Box 278, 00531 Helsinki, Finland). For the time being, only one standardized commercial natural rubber latex extract is available in Europe (Stallergènes, 6 rue Alexis de Tocqueville, F-92183 Antony Cedex, France) [11]. In addition, a few nonstandardized skin prick test extracts (ALK-Abello a/s, Hörsholm, Denmark; Bencard, Missisanga, Ontario, Canada) are commercialized in Europe and Canada. Turjanmaa et al. [12] studied Stallergènes, ALK, Bencard, and the home-made extract, and observed a sensitivity of 83%, 54%, 92%, and 92%, respectively.

No US Food and Drug Administration-approved commercial skin test extract allergen is currently available in the USA.

Cross-sensitization may occur with plant-derived food allergens, especially "tropical" fruits. Well-known cross-reactive foods include avocado, banana, chestnut, kiwi, papaya, potato, and peaches ("Latex-fruit syndrome"). There is also serologic cross-reactivity between natural rubber latex and aeroallergens, for example, pollen ("Latex-mould syndrome").

11.8.2 Airborne Environmental per Annum Allergens

The most common airborne environmental per annum allergens (the list is not limited) are quoted in Table 11.2

In terms of quality, this group is very heterogeneous. Allergens from mites and cockroaches have a good specificity and sensitivity. Sensitivity is less accurate for mould (except *Alternaria*)) and animal allergens.

In atopic patients, prick testing with mite allergens competes with the atopy patch test (see Chap. 9); further studies may reveal their complementarity.

11.8.3 Airborne Environmental Seasonal Allergens

The most common airborne environmental seasonal allergens (the list is not limited) are quoted in Table 11.3. These allergens are pollens from different plants and are of limited interest in dermato-allergology; nevertheless, they could prove useful in atopics. They are of no use in small children, since sensitization to pollens does occur significantly at the age of 5 years. They are chosen according to the geographical area, in relation with environmental variations.

Table 11.2 Airborne environmental per annum allergens

Mites	From house dust: *Dermatophagoides farinae, Dermatophagoides pteronyssinus, Euroglyphus maynei* From storage: *Acarus siro, Glyciphagus domesticus, Lepidoglyphus destructor, Tyrophagus putrescenciae*
Animals	Cat, dog, horse, guinea pig, hamster, rabbit, feathers
Domestic insects	Cockroaches
Moulds	*Alternaria, Aspergillus, Botrytis, Chaetomium, Cladosporium, Epicoccum, Merulius, Mucor, Penicillium, Pullularia, Rhizopus, Stemphyllium, Trichotecium*

Table 11.3 Airborne environmental seasonal allergens

Trees	Betulaceae: birch, hazel, elm, alder
	Fagaceae: chestnut, oak, beech
	Olacaceae: olive tree, ash, privet, forsythia, lilac
	Cupressaceae: cypress, juniper
	Salicaceae: poplar, willow
Graminaceae	Fodder crops: agrostis, creeping wheat-grass, dactylis, fescue, holcus, darnel, meadow (spear)grass, phleum
	Cereal crops: oat, corn, maize, barley, rye
Herbaceae	Compositae: artemisia, ambrosia
	Chinopodiaceae: cherropodium
	Urticaceae: pellitory

11.8.4 Food Allergens (Trophallergens)

The interest of prick testing with foodstuffs is primordial when protein contact dermatitis (see Sect. 10.2) is suspected in foodhandlers. It is of prime importance in occupational dermatology when patients are handling food repeatedly at work, for example, bakers, bartenders, butchers, cooks, fishermen, and fishmongers.

In some cases, positive reactions can lead to a change of job; nevertheless, it is advisable to take into consideration different points of discussion (see below) before drawing any definite conclusion.

The quality of food allergens in terms of sensitivity and specificity is variable. It is often advisable to prick test with fresh foodstuffs, for example, fruits and vegetables, which are handy and more reliable, compared to commercial food allergens. Prick-by-prick testing (see Sect. 11.3), scratch testing (see Sect. 11.4), and scratch-chamber testing (see Sect. 11.5) are highly recommended.

A pitfall when reading prick tests to foodstuffs is related to the fact that some of them may release histamine (or other vasoactive molecules).

When interpreting prick test results, cross-sensitization between foodstuffs is taken into account, but the relevance of cross-sensitization is sometimes doubtful; caution and moderation are needed when expressing our opinion to patients [13].

A positive prick test (or its variants) needs to be confirmed for assessment of relevance by additional procedures (anamnestic data, oral provocation test, eviction/reintroduction, etc.). This step is important prior to edict eviction measures.

Cross-sensitization reactions between food allergens (trophallergens) are listed in Table 11.4.

11.8.5 Occupational Allergens

Occupational allergens are extremely varied [14]. It is out of the scope of this book to include a list of all allergens quoted in recent years. Important ones are given in Table 11.5.
Most of these allergens are not marketed as such. Therefore, they are prepared extemporaneously at the proper concentrations (see textbooks) at the patch and prick test clinic.

11.8.6 Fungi

- *Malassezia furfur*
- *Candida albicans*
- *Epidermophyton*
- *Trichophyton*

Prick testing with these allergens is of very limited clinical interest. Its use is not routinely recommended.

Table 11.4 Cross-sensitization potential reactions to food allergens (trophallergens)

Cereals: corn, rye, barley, oat, maize, pollens of Graminaceae
Leguminosae: peanut, soya bean, peas, lentil, broad bean, kidney bean (bush bean)
Umbelliferae: celery, carrot, parsley, fennel, anise, coriander, cumin, green pepper
Cruciferae: mustard, cabbage, cress, broccoli, turnip, radish, horseradish
Solanaceae: tomato, sweet pepper, potato, paprika, coffee, aubergine
Liliaceae (*amaryllidaceae*): garlic, onion, asparagus, chives, shallot
Nuts: walnut, coconut, hazelnut, pistachio nut, almond, cashew nut
Rutaceae: orange, lemon, grapefruit, mandarin
Drupaceae: apple, hazel nut, peach, pear, apricot, plum, raspberry, strawberry, almond, cherry, birch and hazel tree pollens
Eggs, chicken, turkey, quail, goose, pigeon, feathers
Milk, cheese, beef
Fishes
Shellfish
Mollusca
Celery, carrot, spices, artemisia
Melon, banana
Celery, birch, water melon, cucumber, ambrosia
Honey, pollens
Pork, cat (epithelia)
Latex (see Sect. 11.8.1)
Snail, mites
Barm

11.8.7 Miscellaneous (Immunological and/or Non-Immunological) Urticariogens

A multitude of other (immunological and/or non-immunological) urticariogens is encountered in our environment. As examples, we name blood, caterpillars, corals, jellyfish, saliva, and seminal fluid.

Table 11.5 Occupational Allergens

Latex (see Sect. 11.8.1)
Per annum and seasonal (pollens) allergens (see Sects. 11.8.2 and 11.8.3)
Foodstuffs (see Sect. 11.8.4)
Enzymes: α amylase (bakers), cellulase, papain, xylanase
Brucella abortus, placenta (cow), amniotic fluid (veterinarians)
Silk
Pearl oysters
Urine (mice, rats)
Worms
Various plants (e.g., camomile, tulips)
Plants derivates: abietic acid, colophony, cornstarch, etc.
Tropical woods
Teak
Tobacco
Topical drugs (mainly antibiotics)
Ammonium persulfate and other persulfates
Paraphenylenediamine, para-aminophenol, paramethylaminophenol
Cosmetics, preservatives
Acrylic monomers
Carbamates
Carbonless copy paper
Diglycidyl ether of bisphenol
Formaldehyde resin
Metals (e.g., chromium salts, cobalt, nickel, platinum salts)
Epoxy resins, reactive diluents and hardeners

References

1. Lahti A, Basketter D (2006) Immediate contact reactions. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 83–95
2. Hannuksela M (2006) Protein contact dermatitis. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 345–348
3. Hannuksela M (2006) Skin tests for immediate hypersensitivity. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 429–432
4. Oranje AP, Van Gysel D, Mulder PGH, Dieges PH (1994) Food-induced contact urticaria in atopic dermatitis patients: reproducibility of repeated and duplicate testing with a skin provocation test, the skin application food test (SAFT). Contact Dermatitis 31:314–318
5. Basomba A, Sastre A, Palaez A, Romar A, Campos A, Garcia-Villamanzo A (1985) Standardization of the prick test. A comparative study of three methods. Allergy 40:395–399
6. Malling HJ (1985) Reproducibility of skin sensitivity using a quantitative skin prick test. Allergy 40:400–404
7. Dreborg S, Foucard T (1983) Allergy to apple, carrot and potato in children with birch pollen allergy. Allergy 38:167–172

8. Hannuksela M, Lathi A (1977) Immediate reactions to fruits and vegetables. Contact Dermatitis 3:79–84
9. Niinimaki A (1987) Scratch-chamber tests in food handler dermatitis. Contact Dermatitis 16:11–20
10. Osterballe M, Scheller R, Stahl-Skov P, Andersen KE, Bindslev-Jensen C (2003) Diagnostic value of scratch-chamber test, skin prick test, histamine release ans specific IgE in birch-allergic patients with oral allergy syndrome to apple. Allergy 58:950–953
11. Turjanmaa K, Palosuot T, Alenius H, et al. (1997) Latex allergy diagnosis: in vivo and in vitro standardization of a natural rubber latex extract. Allergy 52:41–50
12. Turjanmaa K, Alenius H, Mäkinen-Kiljunen S, et al. (1995) Commercial skin prick test preparations in the diagnosis of rubber latex allergy (Abstract) J Allergy Clin Immunol 93:S299
13. Rancé F, Juchet A, Brémont F, Dutau G (1997) Correlations between skin prick tests using commercial extracts and fresh foods, specific IgE, and food challenges. Allergy 52:1031–1035
14. Ale SI, Maibach HI (2000) Occupational contact urticaria. In: Kanerva L, Elsner P, Wahlberg JE, Maibach HI (eds) Handbook of Occupational Dermatology. Springer, Berlin, pp 200–216

Part III

Testing in Cutaneous Systemic Adverse Drug Reactions: Interest and Limitations

Testing Procedures in Cutaneous Systemic Adverse Drug Reactions

12

J.-M. Lachapelle

12.1 General Considerations

Cutaneous adverse drug reactions (CADRs) to systemically administered drugs have increased in number during the last few years. This is due to the expanding number of new active molecules used in the treatment of a variety of diseases. CADR are varied and described in full detail in oriented manuals of dermatology [1–4].

Diagnosis of CADR may be straightforward in some cases (Fig. 12.1), but less obvious in some others. The link between the occurrence of a CADR and the systemic administration of a drug (considered to be the culprit agent) is sometimes difficult to assess. The problem is even more complex when several drugs are administrated concomitantly. Several criteria can be taken into account to find the relationship between drug administration and the occurrence of CADRs.

A careful analysis of such criteria has led French authors [5, 6] to describe a scale of imputation (or imputability). This scale includes intrinsic and extrinsic factors. Intrinsic factors are chronological and semeiological, whereas extrinsic ones are based on literature survey. The procedure of evaluation is rather complicated and needs experience. Its detailed description does not fit within the scope of this book. When correctly applied, it provides useful information; its use is highly recommended when CADRs to new drugs are reported. Thus far, its routine adoption has not been reached worldwide.

12.2 Tools of Investigation in CADR

The link between the occurrence of a CADR and the implication of one (or more) suspected drug(s) is a difficult task for the clinician. It implies the use of several tools of investigation, listed in Table 12.1. It is important to put together the various sources of information to reach a high level of imputability, the spirit of which is similar to the determination of a relevance score in patch testing (and other testing) procedures, as explained in Chap. 8.

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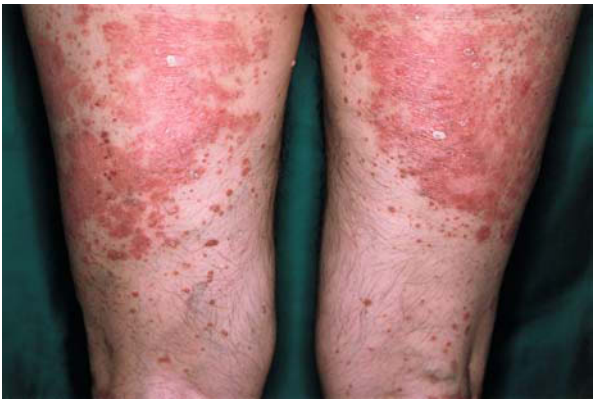

Fig. 12.1 Systemic drug eruption to a sulphonamide: eczematous symmetrical rash on the thighs

12.3 Histopathological Limitations in Diagnosis of a CADR

The histopathological signs of CADRs listed in Table 12.1 are very typical in some cases, such as fixed drug eruption, Stevens-Johnson syndrome, or Lyell's syndrome. Nevertheless, they are not pathognomonic in many others, but may provide useful information [7]. In maculopapular eruptions (the most frequent reaction pattern), differential diagnosis between a CADR and a viral infection can be proposed cautiously to the clinician, taking into account that some CADRs are triggered (or exacerbated) by a virus reactivation, for example, cytomegalovirus, Epstein-Barr virus, etc. The limitations of the histopathological signs are listed in Table 12.2.

By any means it is wise that the dermatopathologist concludes his/her report by the term "compatible (or non-compatible) with CADR."

12.4 Patch Testing in CADR

The use of patch testing in CADR has led to many publications. A general review of the subject has been made by Bruynzeel and Gonçalo [8]. Generally speaking, insufficient standardization in patch testing procedures is evident. Most publications refer to individual cases; extended series of positive and/or negative patch tests results referring to various drugs are lacking. It is noteworthy that more publications are devoted to positive results rather than to negative ones; this is the reason why a Working Party of the European Society of Contact Dermatitis (ESCD) for the study of skin testing in investigating CADR was created. The members of the Working Party have defined some guidelines for performing skin patch tests in CADR [9].

Table 12.1 CADR: tools of investigation for assessment of drug imputation

Clinical examination	Clinical symptoms are characteristic (or not) of a well-defined variety of CADR.
Chronological criteria	Anamnestic data are of crucial importance. Theoretically, there is a chronological link between the administration of a drug and the occurrence of CADR, and, in the same way, between the withdrawal of the drug and the resolution of CADR. Such a time schedule suffers some exceptions. Fading of clinical symptoms may occur several weeks after withdrawal of the drug
Evaluation of additional events	Some occasional events may favor the clinical expression of CADR. These include viral infections (cytomegalovirus, Epstein-Barr virus, parvovirus B19, hepatitis B and C viruses; serological tests may be advised), immunological status, drug interference
Skin biopsy histopathological signs	Skin biopsy may be a contributory tool in some cases of CADR. Histopathological signs of CADR include: vacuolar alteration (Fig. 12.2) and clefts along the dermo-epidermal junction; accumulation of epidermal and/or dermal cytoid (Civatte's) bodies (apoptotic keratinocytes); melanin pigmentary incontinence; interface lymphocytic infiltrate; presence of eosinophils. Typical pictures mainly refer to fixed (bullous and non-bullous) drug eruptions (Fig. 12.3), lichenoid and psoriasiform drug eruptions, acute generalized exanthematic pustulosis. In eczematous CADR, histopathological signs are similar to those encountered in other types of eczema. Some CADRs (e.g., erythema multiforme, Stevens-Johnson syndrome, Lyell's syndrome, leucocytoclastic vasculitis) display characteristic histopathological features
Careful check of the literature	Checking the current literature referring to CADR is a tool of prime importance [4]. This approach includes modern routes of investigation, such as Medline, Internet etc.
Testing procedures	When evaluating the imputation of a drug in the occurrence of CADR, testing (patch and/or prick) procedures can play an undisputed role (see Sects. 12.4 and 12.5), but they are only one of the pieces of the jigsaw puzzle among the other available tools of investigation. Their limitations are linked to several factors, as detailed below.
Provocation test	When a CADR has faded, the systemic reintroduction of the suspected drug (at a lower dose) provokes a recurrent eruption when a positive relationship does exist between the rash and the drug. This procedure provides the more accurate etiological diagnosis; it is the best tool at our disposal nowadays, but it may be submitted to ethical approval in some countries (see Sect. 12.7).

CADR cutaneous adverse drug reactions

12.4.1 The Spectrum of CADRs for Which Patch Testing is Recommended

Positive patch test reactions can be expected to occur when the pathomechanisms of CADR involve delayed-type hypersensitivity (type IV according to the classification of Gell and Coombs) (Fig. 12.1).

Table 12.2 Histopathological limitations in diagnosis of a CADR

Histopathological signs	Limitations
Vacuolar alteration of epidermal basal cells	Can also be found in: lupus erythematosus, dermatomyositis, lichen planus, graft-vs.-host reaction, secondary syphilis etc.
Cytoid bodies (apoptotic keratinocytes)	Lupus erythematosus, lichen planus, graft-vs.-host reaction, secondary syphilis, etc.
Spongiosis and/or spongiotic vesicles in epidermis	Typical of many other eczematous eruptions
Eosinophils in dermal infiltrate	Non-contributory
Psoriasiform features	No distinction can be made between psoriasis and psoriasiform CADR

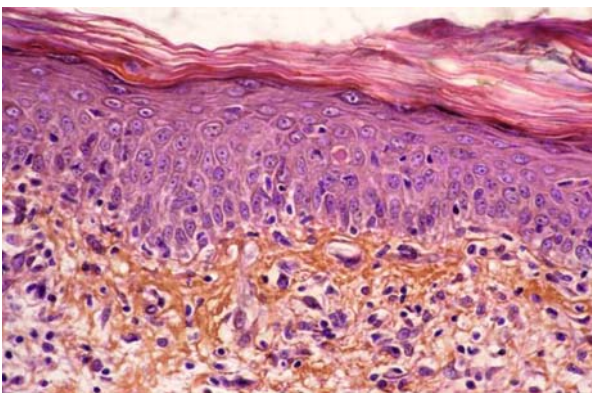

Fig. 12.2 Exanthematous drug eruption. Histopathological features are characteristic, but not pathognomonic: vacuolization of the dermo-epidermal junction implying necrosis of some keratinocytes of the basal layer, dermal lymphocytic infiltrate

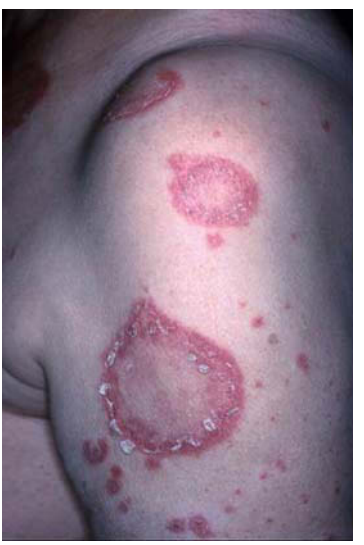

Fig. 12.3 Psoriasiform drug eruption to a beta-blocker. Clearly large marginated erythematosquamous plaques

As emphasized earlier (see Sect. 2.3.3.2) patch tests are usually positive when systemic reactivation of allergic contact dermatitis (SRCD) occurs, i.e., baboon syndrome or Fisher's systemic contact dermatitis.

Some CADR probably express a type IV reaction exclusively (e.g. maculopapular rash or eczematous reactions, whereas some others involve type I plus type IV reactions, or more complex immunological mechanisms (e.g. erythema multiforme, Stevens-Johnson syndrome).

A list of CADRs for which patch testing is recommended is presented in Table 12.3.

Table 12.3 A list of CADRs for which patch testing is recommended

Acute generalized exanthematic pustulosis (AGEP)
Eczematous eruptions (with no previous contact of the allergen with the skin)
Exanthematous maculopapular eruptions (Fig. 12.1)
Exfoliative dermatitis or erythroderma
Fixed drug eruption (bullous or non-bullous) (Fig. 12.5)
Granulomatous drug eruption
Hypersensitivity syndrome (DRESS)
Lichenoid drug eruptions (Fig. 12.4)
Photosensitivity (photoallergic drug eruptions); note that in this case photo patch testing is required (see Chap. 5)
Pityriasis rosea-like eruptions
Pseudolymphomatous drug eruptions
Psoriasiform drug eruptions (Fig. 12.3)
Systemic reactivation of allergic contact dermatitis (baboon syndrome, Fisher's systemic contact dermatitis)

Note: Urticarial drug reactions can be added to the list. It must be considered that patch testing is usually a first step of investigation to be implemented in a second step by prick testing (see Sect. 12.5) and/or intradermal testing (see Sect. 12.6).

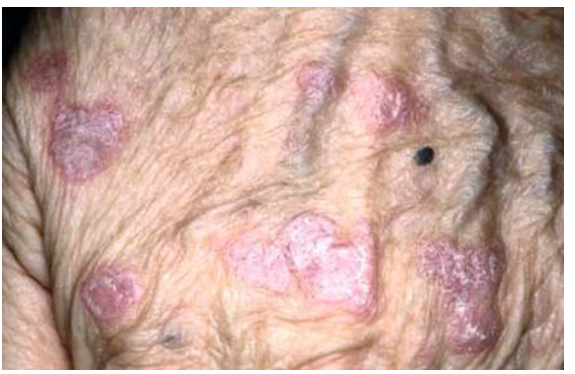

Fig. 12.4 Lichenoid drug eruption to methyldopa. Violaceous flat papules resembling (idiopathic) lichen planus

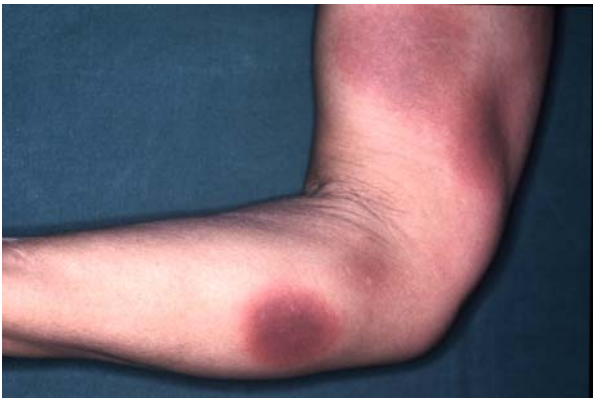

Fig. 12.5 Fixed drug eruption to piroxicam. Sharply marginated erythematous or erythemato-purplish lesions recurring at the same site a few hours for 1–2 days after exposure. The pigmentation deepens after several episodes

12.4.2 The Spectrum of CADRs for Which Patch Testing can be Performed (Being Still Controversial)

Some CADRs implying complex immunological pathomechanisms have been shown to provide positive patch test reactions [6, 7]. A list of CADRs for which patch testing can be performed is presented in Table 12.4.

Table 12.4 A list of CADRs for which patch testing can be performed (still controversial)

Erythema multiforme
Purpura
Stevens-Johnson syndrome
Toxic epidermal necrolysis (Lyell's syndrome)
Vasculitis (Fig. 12.7)

12.4.3 The Spectrum of CADRs for Which Patch Testing is of No Interest

In some CADRs, patch testing has no practical interest. These include acne-like (acneiform) eruptions, alopecia (and hypotrichosis), exacerbation of psoriasis, hypertrichosis, lupus erythematosus, nail changes due to drugs, pigmentary disorders, scleroderma-like reactions, and vesiculo-bullous eruptions (drug–induced pemphigoid, drug-induced pemphigus, linear IgA drug-induced bullous dermatosis).

12.4.4 Guidelines in Drug Patch Testing: General Rules

Some general principles should be borne in mind when patch testing in CADR [9]:

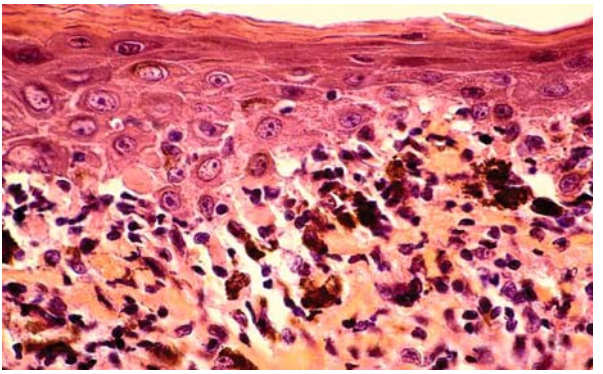

Fig. 12.6 Fixed drug eruption. Histopathological signs include vacuolar alteration along the dermo-epidermal junction, interface lymphocytic infiltrate, accumulation of apoptotic keratinocytes (cytoid or Civatte's bodies), and prominent melanin pigment incontinence

- An informed patient consent is needed
- Patch tests should be performed 6 weeks to 6 months after complete healing of CDAR and at least 1 month after discontinuation of systemic corticosteroids or other immunosuppressive drugs.
- Patch tests should be performed with the commercialized drug and, whenever possible, also with the pure active products and excipients (vehicles).
- Patch testing with drugs, sharing a similar chemical structure, or from the same pharmacological family, may also be important to detect cross-sensitization (see Sect. 3.13) [10].
- An immediate reading of patch tests (at 20 min) is advised to check the potential occurrence of an urticarial reaction. Readings are made at day 2, day 4, and day 7.
- In fixed drug eruptions (Fig. 12.5), patch tests should be performed both on normal skin and on the residual pigmented site of the fixed drug eruption (Fig. 12.7). It is classically observed that patch testing gives a positive response at the site of the lesion ("local memory") and not on intact skin (Fig. 12.6).

12.4.5 Technical Aspects of Drug Patch Testing

All information referring to patch test technology, as provided in Chap. 3, is applicable to patch testing in CADR. Nevertheless, additional information regarding particular aspects of the technology is required.

12.4.5.1 Patch Testing With the Marketed Drugs: Concentrations and Vehicles

The marketed drug used by the patient can be tested (in particular when the pure drug is not available). Pills should have their coating removed, then be ground to a very fine powder. As advised by Barbaud [11, 12], this powder is incorporated at 30% in white petrolatum and diluted at 10% in water.

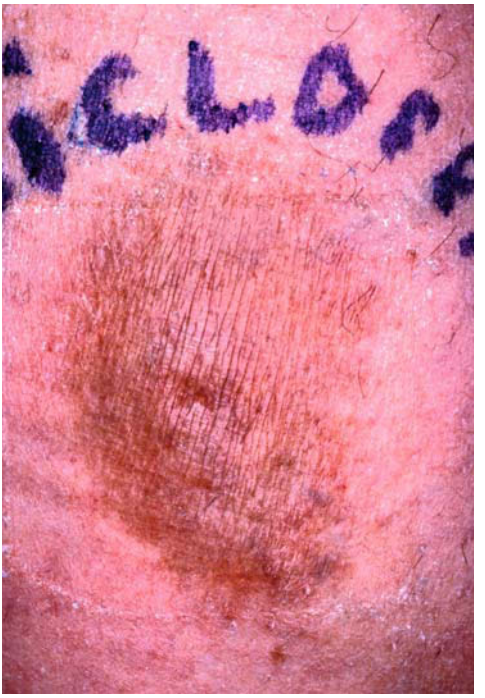

Fig. 12.7 Positive patch test to diclofenac (72 h) performed 2 months later at a previous site of fixed drug eruption

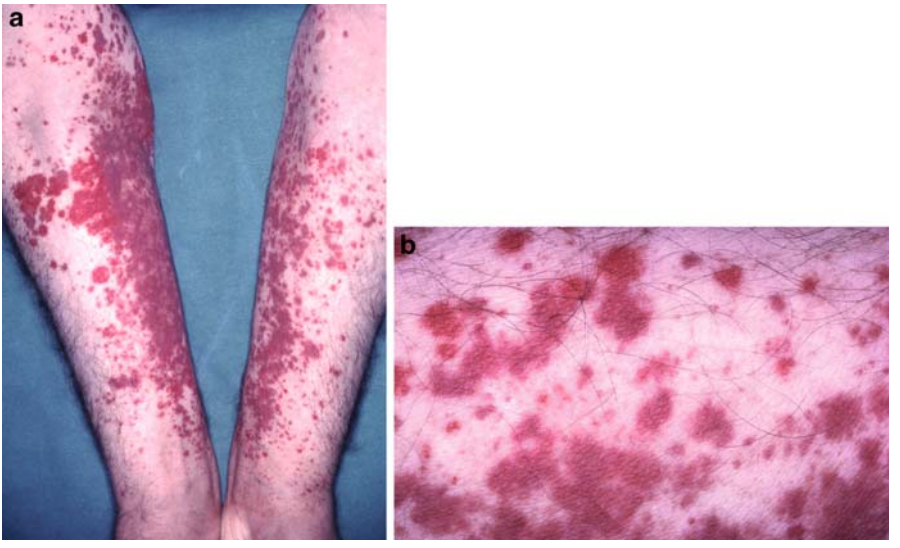

Fig. 12.8 **a**,**b** Drug-induced vasculitis (ofloxacine). (**a**) Symmetrical palpable purpura on the upper limbs. (**b**) Purpuric lesions at higher magnification. Patch tests to ofloxacine are negative

The powder contained in capsules is dispersed at 10% in petrolatum and/or diluted at 10% in water. The gel jacket portion of the capsules should be moistered and tested as is.

Liquid preparations are tested both as is and diluted at 10% in water.

These concentrations are arbitrary, but are considered practical and useful by the members of the ESCD Working Party.

Some drugs are patch-tested at a lower concentration, for example, captopril, desloratadine, misoprostol (1% in petrolatum), to avoid false-positive reactions [12].

12.4.5.2
Patch Testing With Pure Substances: Concentrations and Vehicles

Whenever possible, the pure drug obtained from the manufacturer should be tested dispersed at 10% in petrolatum and also diluted at 10% in water and/or ethanol. This procedure can be adapted; concentrations and vehicles previously considered most adequate for certain drugs should also be chosen.

A complete investigation should include patch-testing with preservatives, coloring agents, and excipients, as is or dispersed at 10% in petrolatum or in the vehicles usually recommended for testing in allergic contact dermatitis.

Some improvements are still needed in this field of patch testing, in terms of concentrations and vehicles, in order to enhance the penetration into the skin of each individual drug. At the present time, we are at a craftsman's stage; improvements require scientific involvement based on multicentric studies and new technologies.

A series of patch tests referred to as the cutaneous adverse drug reaction series is manufactured by Chemotechnique and is now available on the market. This limited list, approved by the ESCD Working Party [13], is presented in Table 12.5. It is expected to be expanded in the future.

12.4.6
Readings of Drug Patch Tests

The results of drug patch testing are scored according to the ICDRG criteria for patch test reading (see Sect. 3.8). As drug patch tests can induce immediate positive reactions, especially with β-lactam antibiotics, these tests have to be read at 20 min in patients who have developed urticaria or anaphylactic shock. Immediate reactions on patch tests have been reported with β-lactam antibiotics, neomycin, gentamycin, bacitracin, and diclofenac [12]. Immediate positive results can be associated with generalized anaphylactic reactions [12].

12.4.7
False-Negative Patch Test Reactions

False-negative reactions can be related to two main reasons:

Table 12.5 Cutaneous adverse drug reaction series

	Concentration (%)
1. Penicillin G, potassium salt	10
2. Amoxicillin trihydrate	10
3. Dicloxacillin sodium salt hydrate	10
4. Cefotaxim sodium salt	10
5. Doxycyclin monohydrate	10
6. Minocyclin hydrochloride	10
7. Erythromycin base	10
8. Spiramycin base	10
9. Clarithromycin	10
10. Pristinamycin	10
11. Cotrimoxazole	10
12. Norfloxacine	10
13. Ciprofloxacine hydrochloride	10
14. Carbamazepine	1
15. Hydantoin	10
16. Diltiazem hydrochloride	10
17. Captopril	5
18. Acetylsalicylic acid	10
19. Diclofenac sodium salt	1
20. Ketoprofene	1
21. Piroxicam	1
22. Acetaminophen	10
23. Acyclovir	10
24. Hydroxyzine hydrochloride	1
25. Hydrochlorothiazide	10
26. Clindamycin phosphate	10
27. Cefradine	10
28. Cefalexin	10
29. Ibuprofen	10

Concentrations refer to petrolatum

- Insufficient penetration of the drug into the skin to elicit an allergic response.
- The allergen is not the drug itself, but one of its metabolites. The metabolites are delivered into the skin, when the drug is administered systemically, but not necessarily when the drug is applied onto the skin (depending on the enzymatic pathways involved).

12.4.8 False-Positive Patch Test Reactions

Application of the drug onto the skin can induce a false-positive reaction (due to an irritant effect). When a new drug is patch tested (therefore, without drug reference from

the literature) and gives a positive response, the interpretation of which being difficult, it is useful to patch test control subjects. Patch testing control subjects may require ethical approval.

12.5 Prick Testing in CADR

Prick testing has an undisputed interest in CADR when an immunological immediate-type reaction (type I) is suspected (mainly drug urticarial reactions), eventually associated with other complex immunological mechanisms.

The usefulness of prick testing is evident in urticaria provoked by penicillin. Prick tests are performed on the volar forearm with the commercialized form of the drug. Whenever possible, both the pure drug and the excipients have to be tested.

It is advised to use pure drugs at sequential dilutions (10^{-3}, 10^{-2}, 10^{-1} then pure) [9]. Technological aspects are similar to those described in Chap. 11.

12.5.1 Intradermal Testing in CADR

Intradermal tests (IDT) are performed only when prick tests show negative results 20 min after testing with the suspected drug [12]. They have to be done under hospital surveillance. It is necessary to obtain sterile forms of the drug. Some authors use non-injectable drugs [12]. The techniques involved require expertise, and IDT is performed almost exclusively in specialized university centers.

When read at 20 min, IDT would be considered as having positive results when the diameter of the reaction would be ≥6 mm.

12.5.2 Oral Provocation Test (Oral Challenge) in CADR

The oral provocation test (oral challenge) is conceptually the best tool of investigation in CADR, as it is intended to reproduce exactly the clinical conditions involved previously during the onset of the disease. To such extent, it can be compared with the ROAT test used for the investigation of ACD, closer to the reality than conventional patch testing (see Sect. 7.4).

Nevertheless, in current practice, the oral provocation test has obvious limitations and strict conditions of use. Indeed, some CADRs (a) are disseminated and therefore troublesome for the patient; (b) exhibit severe clinical symptoms (DRESS, vasculitis, Stevens-Johnson syndrome, etc); (c) or are even life-threatening (Lyell's syndrome). In all these circumstances, the oral provocation test is unethical and undeniably forbidden.

When CADRs are more discrete clinically (e.g., maculopapular eruptions of limited extent, fixed drug eruption, etc), the oral provocation test can be performed after

discussion. This is particularly true when other tests (see Sect. 12.4) are negative, and more precisely when the clinician is convinced that the drug is the culprit agent and that patch-testing or prick-testing negative results may be considered false-negatives (see Sect. 12.4.7).

When in doubt, the final decision is dependent on the evaluation of the risk/benefit ratio for the patient. It is often recommended to obtain the agreement of the local Ethical Committee.

The doses of the drug to be administered are not codified, and there are no generally accepted guidelines in the literature. The half- or the fourth part of the initial dose is reasonably acceptable.

References

1. Breathnach SM, Hinter H (1992) Adverse drug reactions and the skin. Blackwell Scientific Publications, Oxford
2. Zürcher K, Krebs A (1992) Cutaneous drug reactions. An integral synopsis of today's systemic drugs, 2nd edn. Karger, Basel
3. Pirchler WJ (2007) Drug Hypersensitivity. Karger, Basel
4. Litt JZ (2008) Litt's Drug Eruption Reference Manual, 14th edn. Informa Healthcare, London
5. Bégaud B, Evreux JC, Jouglard J, Lagier G (1985) Unexpected or toxic drug reaction assessment (imputation). Actualization of the method used in France. Thérapie 40:111–118
6. Moore N, Paux G, Begaud B, Biour M, Loupi E, Boismare F, Royer RJ (1985) Adverse drug reaction monitoring: doing it the French way. Lancet 2:1056–1058
7. Weedon D (2007) Skin Pathology, 2nd edn. Churchill Livingstone, Edinburgh
8. Bruynzeel DP, Gonçalo M (2006) Patch testing in adverse drug reactions. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 401–412
9. Barbaud A, Gonçalo M, Bruynzeel D, Bircher A (2001) Guidelines for performing skin tests with drugs in the investigation of cutaneous adverse drug reactions. Contact Dermatitis 45:321–328
10. Oliveira HS, Gonçalo M, Reis JP, FIgueiredo A (1999) Fixed drug eruption to piroxicam. Positive patch tests with cross-sensitivity to tenoxicam. J Dermatol Treatment 10:209–212
11. Barbaud A, Reichert-Penetrat S, Trechot P, Jacquin-Petit MA, Ehlinger A, Noirez V, Faure GC, Schmutz JL, Bene MC (1998) The use of skin testing in the investigation of cutaneous adverse reactions. Br J Dermatol 139:49–58
12. Barbaud A (2007) Place of drug skin tests. In: Pirchler WJ ed. Karger, Basel, pp 366–379
13. Barbaud A (2007) Drug skin tests and systemic cutaneous adverse drug reactions: an update. Expert Rev Dermatol 2:4 www.Future-drugs.com

Appendix

A Additional Series of Patch Tests

J.-M. Lachapelle

A.1 Introductory Remarks

As emphasized in Sect. 4.9, additional series of patch tests are very useful in daily practice. Each additional series of patch tests is a tool of investigation targeted to explore a specific field of our environment.

General principles and considerations have to be pointed out:

a. The list of allergens mentioned in each series is based on the literature and selected accordingly.
b. Each list is always incomplete, as new (potentially allergenic) chemicals are constantly introduced in the composition of end-products; this is particularly true for cosmetics, plastics, and/or rubber additives.
c. Each list is therefore indicative, and the alert clinician must be aware of the fact that it is needed to complete the investigation by other tests, such as open tests, semi-open tests, and ROAT's with patients' own products (see Chap. 7).
d. It is also flexible and must be cautiously adapted to environmental changes. Some allergens are either withdrawn from the market (for some uses) or used at lower concentrations. It can be anticipated that in such conditions the incidence of positive allergic patch test reactions to those allergens will decrease. This is called the "Dillarstone effect" [1]. Classical examples include, for example, Cl+ Me-isothiazolinone and, more recently, methyldibromoglutaronitrile [2]. But, surprisingly, this is not always the case. Isoeugenol, an important fragrance allergen in consumers, has been restricted to 200 ppm since 1998 according to the guidelines issued by the fragrance industry [3]. Despite of this, an epidemiological study, conducted in Great-Britain from 2001 to 2005, has revealed an increase in isoeugenol positive patch test reactions [4].

Therefore, it is often wise to maintain in the lists some allergens even if their use is decreasing. This remark is valid for the standard series (see Chap. 4) and for all additional series.

These are listed below:

- Bakery series
- Corticosteroid series
- Cosmetic series
- Epoxy resin series
- Hairdressing series
- Isocyanate series
- Metal series
- (Meth)acrylate series
- Plastics and glues series
- Rubber additives series
- Textile dyes and finish series

A.2 Bakery Series

Hand dermatitis is a common problem among bakers. Differential diagnosis includes irritant contact dermatitis (see Sect. 2.3), allergic contact dermatitis (see Sect. 2.1), and protein contact dermatitis (see Sect. 10.2).

Some patch tests of the standard series are of great interest, particularly balsams of Peru, Fragrance mix 1, Fragrance mix 2, and their individual constituents, such as eugenol and isoeugenol. Furthermore, additional patch tests are needed; they are listed in Table A.1.

Table A.1 Bakery series

	Concentration (%)
1. Sodium benzoate	5
2. 2-*tert*-Butyl-4 methoxyphenol	2
3. Anethole	5
4. Sorbic acid	2
5. Benzoic acid	5
6. Propionic acid	3
7. Octyl gallate	0.25
8. Dipentene (d-Limonene)	2
9. Ammonium persulfate (°)	2.5
10. Propyl gallate	1
11. Benzoyl peroxyde	1
12. Dodecyl gallate	0.25
13. Vanillin	10
14. Menthol	2
15. Butylhydroxytoluene	2

Concentrations refer to petrolatum
(°) Immediate reading (20 min) is mandatory, since this allergen may elicit a type I reaction (see Sect. 10.1)

The search for PCD is made by open (non-prick) and prick testing (see Chap. 11) with flour, yeast, alpha-amylase, etc.

A.3 Corticosteroid Series

Allergic contact dermatitis (ACD) to topical corticosteroids is not infrequent, but sometimes underestimated, due to its atypical clinical presentation, and usually very discrete. Indeed, the anti-inflammatory properties of corticosteroids modify the clinical aspects of the lesions. Nevertheless, in some cases, acute vesicular ACD to corticosteroids may occur (Fig. A.1)

Two corticosteroids, budesonide and tixocortol-21-pivalate (Fig. A.2), are considered the best markers for detecting corticosteroid ACD. They are included in the standard series (see Sect. 4.3).

A list of additional corticosteroids (Fig. A.3) is available (Table A.2). The list remains limited, because for most corticosteroids petrolatum is not convenient as vehicle. Ethanol is the first choice but, unfortunately, corticosteroids are unstable in ethanol and often degrade after 1 month of storage in refrigerator.

In practice, it is therefore important to test patients with their own corticosteroid preparations (including eventually ROATs).

Two remarks about readings of patch tests:

a. Three readings are advised: at days 2, 4, and 7. Reading at day 7 is of prime importance, taking into account the frequent occurrence of late reactions (see Sect. 3.7.4).
b. The edge effect (see Sect. 3.8.3) is commonly observed with corticosteroids.

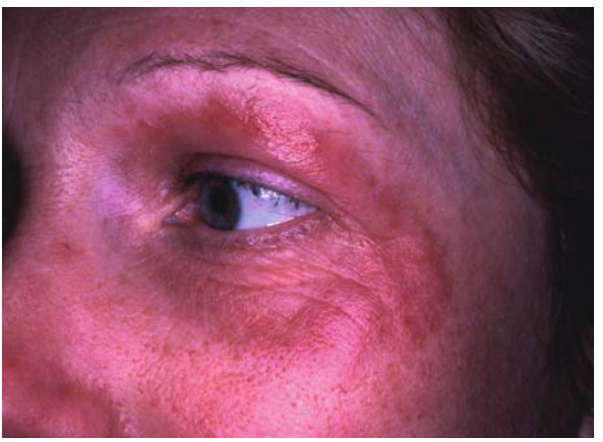

Fig. A.1 Allergic contact dermatitis of the eyelids to a cream containing alclomethasone-17,21-dipropionate

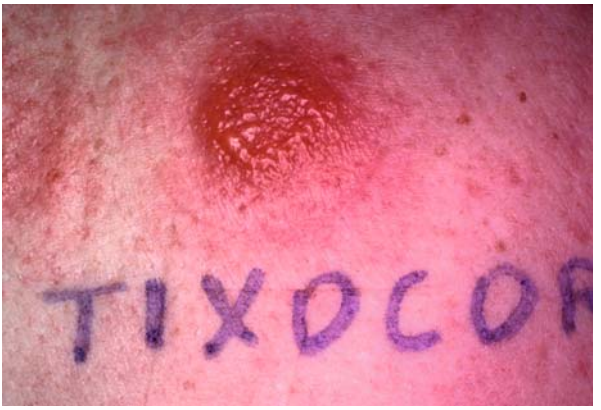

Fig. A.2 Patch test scored ++ to tixocortol pivalate. Reading at 48 h

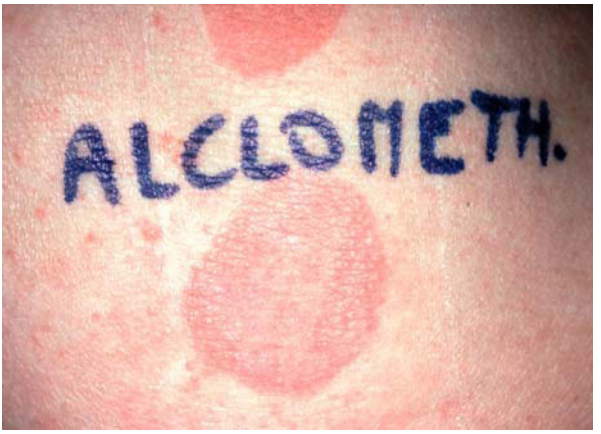

Fig. A.3 Patch test scored + to alclomethasone-17,21-dipropionate. Reading at 96 h

Table A.2 Corticosteroid series

	Concentration (%)
1. Betamethasone-17-valerate	1
2. Triamcinolone acetonide	1
3. Alclomethasone-17,21-dipropionate	1
4. Clobetasol-17-propionate	1
5. Dexamethasone-21-phosphate disodium salt	1
6. Hydrocortisone-17-butyrate	1 (eth.)

Concentrations refer to petrolatum unless otherwise stated

A.4
Cosmetic Series

Any proposal of a cosmetic series is ill-defined, arbitrary, provisional, and by any means incomplete due to the complexity of cosmetic products' formulations (Figs. A.4 and A.5). Nevertheless, bearing this in mind, it is worthwhile to build a list (Table A.3) of common allergens, a «core» of chemicals present in cosmetics throughout the world. Such a list is also appropriate for topical drugs (creams, ointments, lotions, gels, etc.), ACD being related either to the active drug itself or to one of the components of the vehicle. It may also be useful for detecting ACD to household products such as cleansers, laundry agents, and fabric softeners.

When cosmetic ACD is suspected, it is recommended to test the patient with his (her) own products, including patch tests, open tests (see Sect. 7.2), semi-open tests (see Sect. 7.3), and ROATs (see Sect. 7.4).

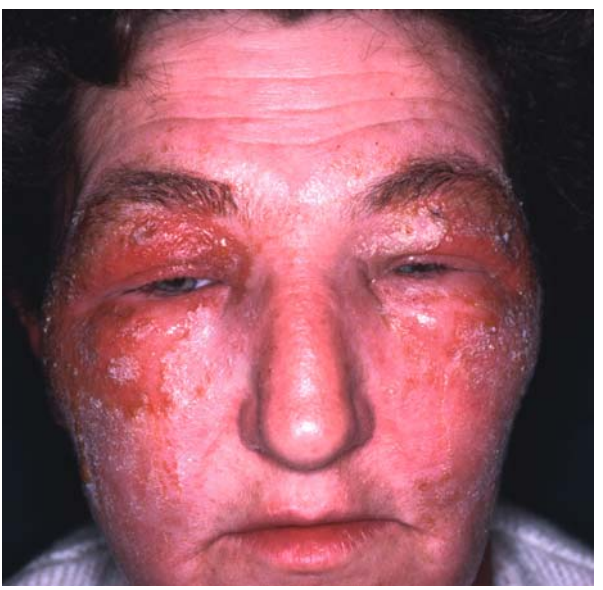

Fig. A.4 Allergic contact dermatitis to a face cream. Positive allergic patch tests to fragrance mix 1 and to fragrance mix 2

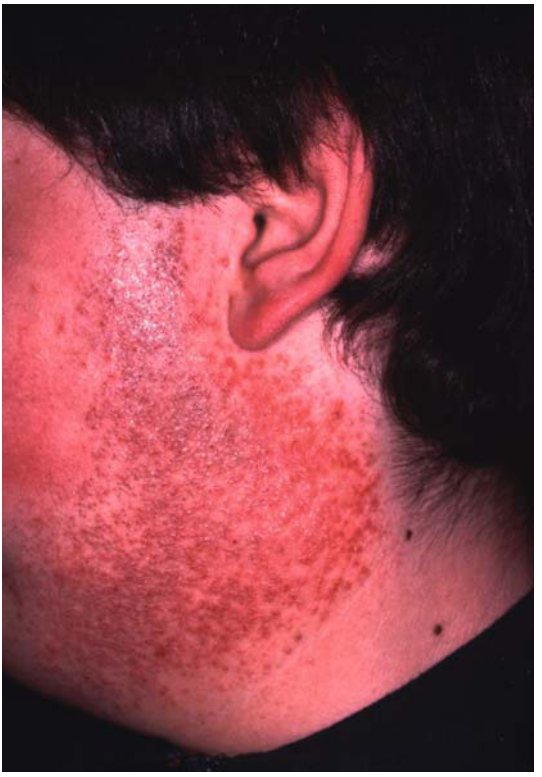

Fig. A.5 Allergic contact dermatitis to an aftershave lotion. Positive allergic patch test reaction to imidazolidinylurea. The ROAT test with the aftershave lotion was positive after eight applications

Table A.3 Cosmetic series

	Concentration (%)
Preservatives (anti-oxydants and/or disinfectants)	
1. Butylhydroxyanisole (BHA)	2
2. 2,6-Ditert-butyl-4-cresol (BHT)	2
3. Triclosan	2
4. Sorbic acid	2
5. Thimerosal (thiomersal)	0.1
6. Imidazolidinylurea	2
7. Diazolidinylurea	2
8. Hexamethylenetetramine	2
9. Chlorhexidine digluconate [a]	0.5 (aq.)
10. Chloracetamide	0.2
11. Ethylenediamine dihydrochloride	1

(continued)

Table A.3 (continued)

	Concentration (%)
12. 2-Bromo-2-nitropropane-1,3-diol (Bronopol)	0.5
13. Benzylalcohol	1
14. *tert*-Butylhydroquinone	1
15. Propylgallate	1
16. Dodecylgallate	0.25
17. DMDM Hydantoïne	2 (aq.)
18. Iodopropynyl butylcarbamate	0.1
Other (emollients, emulsifiers, etc.)	
19. Amerchol L 101	50
20. Isopropyl myristate	20
21. Triethanolamine	2
22. Sorbitan sesquioleate	20
23. Stearyl alcohol	30
24. Cetyl alcohol	5
25. Cocoamidopropylbetaïne	1 (aq.)
26. Dimethylaminopropylamine	1 (aq.)
27. Sodium metabisulfite	1
28. Tea Tree Oil	5
29. Laurylglycoside	3
30. Abitol[b]	10
31. Toluene sulfonamide formaldehyde resin [(c)]	10

Concentrations refer to petrolatum unless otherwise stated
[a]Can provoke immediate reactions (see Chap. 10)
[b]Adhesive: mascara
[c]Adhesive: nail lacquers

A.5 Epoxy Resin Series

The technologies implied in epoxy resin systems are very diversified and in continuous evolution. Epoxy resin itself is the most common allergen, but when ACD is suspected, it is advisable to test the patient with the epoxy resin used at the workplace (usually 1% pet.) and, additionally, to reactive diluents and hardeners listed in Table A.4. Indeed, many of these have well-documented allergenic properties (Fig. A.6). As for the other series, the list is indicative, and it is therefore possible that other reactive diluents and/or hardeners are involved to be tested also at a proper concentration.

Table A.4 Epoxy resin series

	Concentration (%)
Reactive diluents	
1. Cresylglycidylether	0.25
2. Phenylglycidylether	0.25
3. Butylglycidylether	0.25
4. 1,6-Hexanedioldiglycidylether (techn. grade)	0.25
5. 1,4-Butanedioldiglycidylether	0.25
6. *p-tert*-Butyphenylglycidylether	0.25
Hardeners	
7. Ethylenediamine dihydrochloride	1
8. Triethylenetetramine	0.5
9. 4,4′-Diaminodiphenylmethane	0.5
10. Isophoronediamine (IPD)	0.5
11. Hexamethylenetetramine	1
12. Trimethylhexane-1,6-diamine (isomere blend)	0.5

Concentrations refer to petrolatum

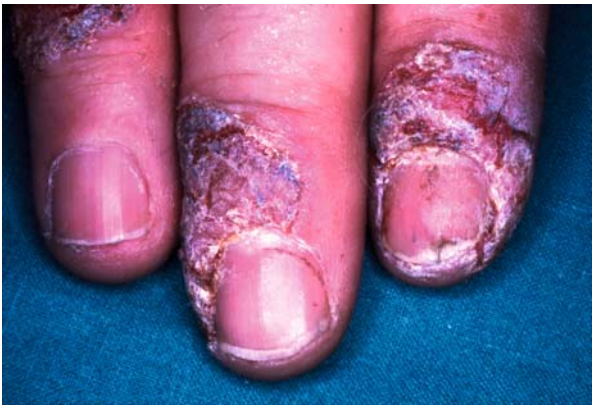

Fig. A.6 Allergic contact dermatitis to epoxy resins. The topography of lesions, confined strictly to the fingers, emphasizes the precision of the movements involved. Positive allergic patch test reactions to epoxy resin and to cresylglycidyl ether

A.6 Hairdressing Series

Hairdresser's hand dermatitis is frequent. Differential diagnosis includes irritant contact dermatitis (see Sect. 2.3), allergic contact dermatitis (see Sect. 2.1), and worsening by irritancy of atopic dermatitis (see Chap. 9).

ACD is a current problem in young hairdressers (Fig. A.7). In those cases, patch testing with the standard series may be very informative (*p*-phenylenediamine, nickel sulfate, formaldehyde), but insufficient for a targeted investigation. Additional patch testing with allergens listed in Table A.5 is highly recommended and very often of prime importance: the allergens are referred to as permanent waving formulations, permanent hair dyes, hair bleaches, and/or preservatives.

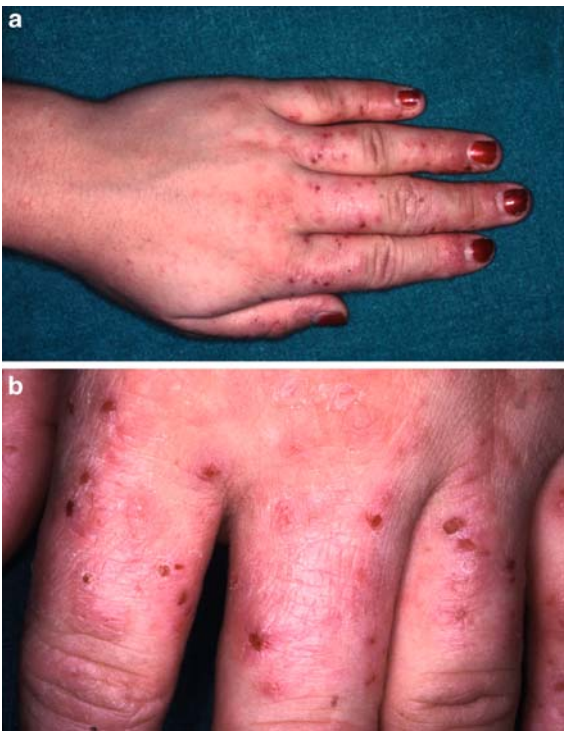

Fig. A.7 Allergic contact dermatitis to paraphenylenediamine in a female hairdresser. The lesions are slightly erythematous and highly pruritic. **(a)** The fact that the lesions are confined to the dorsal hands is explained by the precision of the occupational movement involved. **(b)** Multiple erosions due to pruritus are prominent

Table A.5 Hairdressing series

	Concentration (%)
1. Ammonium thioglycolate	2.5 (aq.)
2. Monoethanolamine	2
3. 2,5-Diaminotoluene sulfate	1
4. 4-Toluenediamine (dye complex)	1
5. 2-Nitro-4-phenylenediamine	1
6. 3-Aminophenol	1
7. 4-Aminophenol	1
8. Resorcinol	1
9. Pyrogallol	1
10. Glyceryl monothioglycolate (GMTG)	1
11. Chloracetamide	0.2
12. Cocamidopropylbetaïne	1 (aq.)
13. Ammonium persulfate (°)	2.5
14. Hydroquinone	1

Concentrations refer to petrolatum unless otherwise stated
(°) Immediate reading (20 min) is mandatory, since this allergen may elicit a type I reaction (see Sect. 10.1)

A.7 Isocyanate Series

Isocyanates are used in the manufacture of polyurethane foams, paints, plastics, lacquers, elastomers, adhesives, glues, printing plates, etc. A list of common allergens is presented in Table A.6.

Table A.6 Isocyanate series

	Concentration (%)
1. Toluene diisocyanate (TDI)	2
2. Diphenylmethane-4,4-diisocyanate (MDI)	2
3. Diaminodiphenylmethane	0.5
4. Isophoronediisocyanate (IPDI)	1
5. Isophoronediamine (IPD)	0.1
6. 1,6-Hexamethylene-diisocyanate (HDI)	0.1

Concentrations refer to petrolatum

A.8 Metals Series

A vast range of metals are available for patch testing, manufactured by different companies (e.g., 42 allergens marketed by Chemotechnique: updated list, March 2008). Many of them are of limited interest due to anecdotical occurrence of clinical observations related to those metals. Nevertheless, some deserve special attention and are listed in Table A.7. ACD to gold has been a controversial issue, but is now well-documented by several studies. Goldsodium thiosulfate is the most suitable allergen for detecting allergy to gold.

Table A.7 Metals series

	Concentration (%)
1. Goldsodium thiosulfate	0.5
2. Ammoniated mercury	1
3. Palladium chloride	2
4. Copper sulfate	2
5. Ammonium tetrachloroplatinate	0.25 (aq.)
6. Aluminium	as is

Concentrations refer to petrolatum unless otherwise stated

A.9 (Meth)Acrylate Series

Acrylic and methacrylic resins are thermoplastics formed by the derivates of acrylic and methacrylic acids. Numerous acrylic and methacrylic monomers exist, and as a result, a multitude of different polymers and resins are produced. Uses of acrylates and methacrylates are varied. The most often quoted are in dentistry, leather finishes, adhesives, glues, paints (Fig. A.8), printing inks and coatings, artificial nails, etc., and many others are described in the literature.

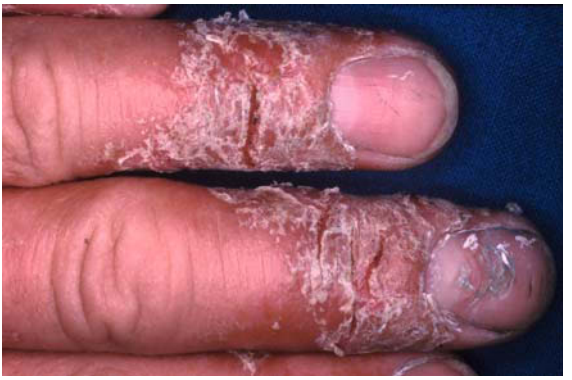

Fig. A.8 Allergic contact dermatitis to acrylates in a painter

Table A.8 Meth(acrylate) series

	Concentration (%)
1. Methyl methacrylate (MMA)	2
2. *n*-Butyl methacrylate (EMA)	2
3. 2-Hydroxyethyl methacrylate (2-HEMA)	2
4. 2-Hydroxypropyl methacrylate (2-HPMA)	2
5. Ethyleneglycol dimethacrylate (EGDMA)	2
6. Triethyleneglycol dimethacrylate (TREGDMA)	2
7. 1,4-Butanediol dimethacrylate (BUDMA)	2
8. Urethane dimethacrylate (UEDMA)	2
9. 2,2-Bis {4-(methacryloxy)-phenyle} propane (BIS-MA)	2
10. 2,2-Bis {4-(2hydroxy-3-methacryloxypropoxy)-phenyle} propane (BIS-GMA)	2
11. 1,6-Hexanediol diacrylate (HDDA)	0.1
12. Tetrahydrofurfuryl methacrylate	2
13. Tetraethyleneglycol dimethacrylate (TEGDMA)	2
14. *N*,*N*-Dimethylaminoethyl methacrylate	0.2
15. Ethyl cyanoacrylate	10

Concentrations refer to petrolatum

Some companies (e.g., Chemotechnique) provide several (meth)acrylate series in relationship with specific uses. They are labeled (a) (Meth)acrylate series (adhesives, dental, and others); (b) (Meth)acrylate series (nails-artificial); (c) (Meth)acrylate series (printing).

The series presented here is not related to specific uses; it is therefore certainly imperfect; nevertheless, it is considered very useful in most cases (Table A.8).

A.10 Plastics and Glues Series

Note that this series is in some way misleading, as many new allergens are regularly introduced in the technological procedures involved in the plastic and glues industry. Caution is therefore needed in its interpretation (Table A.9).

1,2-Benzisothiazolin-3-one (BIT) is an allergen of current increasing interest. It is used in many industries as a preservative in water-based solutions. It has been reported recently in disposable polyvinyl chloride gloves.

Patch testing with patient's own resin(s) is mandatory. It is also important to refer to the (meth)acrylate, epoxy resin, and isocyanate series.

Table A.9 Plastics and glues series

	Concentration (%)
1. Phenolformaldehyde resin	5
2. Toluenesulfonamide formaldehyde resin	10
3. Abitol	10
4. Turpentine oil	10
5. 4-*tert*-Butylphenol	1
6. 4-*tert*-Butylcatechol	0.25
7. Di-*n*-butylphtalate	5
8. Tricresyl phosphate	5
9. Triphenyl phosphate	5
10. Dimethyl phtalate	5
11. Di-2-ethylhexyl phtalate	5
12. Bisphenol A	1
13. Abietic acid	10
14. Hydroquinone	1
15. Phenylsalicylate	1
16. 2,6-Ditert-butyl-4-cresol (BHT)	2
17. 2(2-Hydroxy-5-methylphenyl)benzotriazol	1
18. Benzoyl peroxide	1
19. Azodiisobutyrodinitrile	1
20. Resorcinol monobenzoate	1
21. 2-Phenylindole	2
22. 2-*tert*-Butyl-4-methoxyphenol (BHA)	2
23. 2-Monomethylol phenol	1
24. Diphenylthiourea	1
25. 2-*n*-Octyl-4-isothiazolin-3-one	0.1
26. Cyclohexanone resin	1
27. Triglycidyl isocyanurate	0.5
28. 1,2-Benzisothiazolin-3-one (BIT)	0.1

Concentrations refer to petrolatum

A.11 Rubber Additives Series

Rubber items are of common use in daily life. The technology of rubber vulcanization is complex and involves the occurrence of various chemicals, some of which have a high allergenic potential. It is the reason why the more frequent are included in the standard series. When rubber allergy is suspected, an additional series of allergens is available (Table A.10). It must be mentioned that the list is only indicative and provisional, as new technologies are regularly introduced in the rubber industry, leading to the emergence of

Table A.10 Rubber additives series

	Concentration (%)
1. Tetramethylthiuram disulfide	1
2. Tetramethylthiuram monosulfide	1
3. Tetraethylthiuram disulfide	1
4. Dipentamethylenethiuram disulfide	1
5. *N*-Cyclohexyl-*N*-phenyl-4-phenylenediamine	1
6. *N*,*N*-Diphenyl-4-phenylenediamine	1
7. *N*-Cyclohexylbenzothiazyl sulfenamide	1
8. Dibenzothiazyl disulfide	1
9. Morpholinylmercaptobenzothiazole	1
10. 1,3-Diphenylguanidine	1
11. Zinc diethyldithiocarbamate	1
12. Zinc dibutyldithiocarbamate	1
13. Zinc dibenzylthiocarbamate	1
14. *N*,*N* Di-beta naphtyl-4-phenylenediamine	1
15. *N*-Phenyl-2-naphtylamine	1
16. Hexamethylenetetramine	2
17. Diphenylthiourea	1
18. Zinc dimethyldithiocarbamate	1
19. 2,2,4-Trimethyl-1,2-dihydroquinoline	1
20. Diethylthiourea	1
21. Dibutylthiourea	1
22. Dodecylmercaptan	0.1
23. *N*-Cyclohexylthiophthalimide	1
24. Diaminodiphenylmethane	0.5
25. 1,3-Diphenylguanidine	1
26. 4,4′-Dihydroxybiphenyl	0.1
27. 4-*tert*-Butylcatechol	0.25

Concentrations refer to petrolatum

new allergens. Therefore, it is advised to test with the suspected rubber items, for example, gloves, boots, etc. (see Sect. 7.5.3), and to obtain from the manufacturer detailed information about the additives used in the vulcanization process.

Moreover, prick testing with natural rubber latex (see Chap. 11) is recommended.

A.12 Textile Dyes and Finish Series

Textile dyes and finish series has gained importance in the last years. The series (Table A.11) can be divided into three groups of allergens:

Table A.11 Textile dyes and finish series

	Concentration (%)
Disperse dyes	
1. Disperse orange 1	1
2. Disperse orange 3	1
3. Disperse brown 1	1
4. Disperse red 1	1
5. Disperse red 17	1
6. Disperse yellow 3	1
7. Disperse yellow 9	1
8. Disperse blue 3	1
9. Disperse blue 35	1
10. Disperse blue 85	1
11. Disperse blue 106	1
12. Disperse blue 153	1
13. Disperse blue 124	1
14. Disperse blue mix 106/124	1
Other dyes	
15. Basic red 46	1
16. Reactive black 5	1
17. Reactive blue 21	1
18. Reactive blue 238	1
19. Reactive orange 107	1
20. Reactive red 123	1
21. Reactive red 238	1
22. Reactive red 228	1
23. Reactive violet 5	1
24. Acid red 118	5
25. Direct orange 34	5
26. Acid red 359	5
Textile finish resins	
27. Dimethylol dihydroxyethyleneurea	4.5 (aq)
28. Dimethyl dihydroxyethyleneurea	4.5 (aq)
29. Dimethylol dihydroxyethyleneurea modified	5 (aq)
30. Ethyleneurea,melamineformaldehyde (*)	5
31. Urea formaldehyde	10
32. Melamine formaldehyde	7

Concentrations refer to petrolatum unless otherwise stated
(*) emulsified with sorbitan sesquioleate 5%

A.12.1 Disperse Dyes

Disperse dyes are so-called because they are partially soluble in water. These synthetic dyes have either an anthraquinone (disperse anthraquinone dyes) or an azoic structure (disperse azo dyes). They are the most commonly employed dyes in the textile industry to color synthetic fibers (Fig. A.9) such as polyester, acrylic and acetate, and sometimes nylon, particularly, in stockings. They are not used for natural fibers. These molecules are the main textile sensitizers. Disperse Orange 3 is positive in a great majority of PPD-positive people, because hydrolysis occurs in the skin into PPD. Disperse Orange 3 can also be found in some semi-permanent hair dyes.

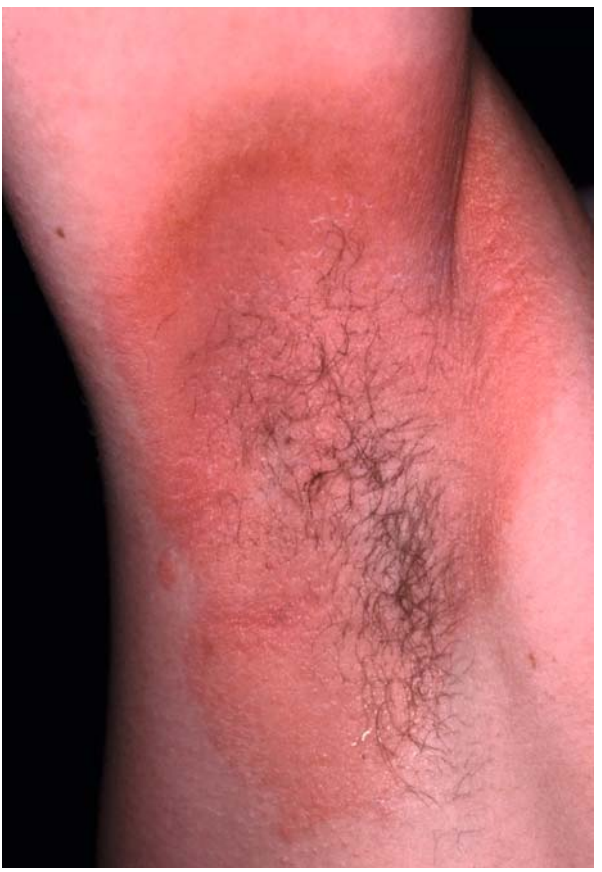

Fig. A.9 Allergic contact dermatitis to the dye in a blue dress. Allergen dissolution by sweat accounts for the axillary location. The Disperse Blue 106 patch test was positive

A.12.2
Other Dyes

Other dyes are acid, basic, direct, and fiber-reactive dyes. All of these are less common allergens.

A.12.3
Textile Finish Resin Allergens

Textile finish resins are used to enhance the touch and quality of clothing. Some of them (urea formaldehyde and melamine formaldehyde) significantly release formaldehyde.

It is recommended in all cases to patch test with patient's own clothing. Patch tests are sometimes irritant, inducing slight erythema and edema fading at the second reading.

A.13
Other Series

Other additional series of patch tests are proposed by companies. They are not included in the Appendix, as they are in some way misleading. Two examples of such series are shoe series and plant series. Instead of presenting series of allergens, it is more appropriate to suggest strategies of patch testing, when confronted with those problems.

A.13.1
Shoe Dermatitis

ACD of the feet caused by shoe allergens is fairly common [5] and should be considered in all patients with foot eczema (Figs. A.10 and A.11).

Three steps of investigation are recommended:

Step 1: Is shoe dermatitis ACD? Differential diagnosis embraces irritant contact dermatitis (often linked with maceration), atopic dermatitis, juvenile plantar dermatosis, and eventually other dermatoses such as tinea pedum, psoriasis, palmoplantar pustulosis, lichen planus, pityriasis rubra pilaris, etc. It has to be kept in mind that ACD can be superimposed on the primary skin disease and, taking this into account, patch testing is advised in most cases.

Step 2: The components of shoes are extremely varied. Therefore, the first approach is to test patients with different pieces of the shoe, cut with a scalpel (see Sect. 7.5.3). Positive patch tests to solid materials are usually relevant, but they give no information about the potential allergens. The simultaneous application of the standard series (see Sect. 4.3) can afford a first indication but further investigation is most often needed.

Step 3: The third step is to patch test patients with different allergens present in the additional series (rubber additives, plastics and glues, textile dyes, etc.) selected according to

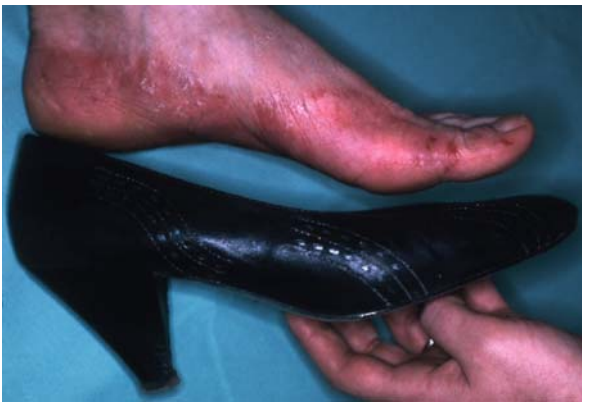

Fig. A.10 Allergic contact dermatitis to a glue used in shoe manufacture. The topography of the mildly edematous, erythematosquamous eczema is highly typical. The formaldehyde paratertiary butylphenol resin patch test was positive

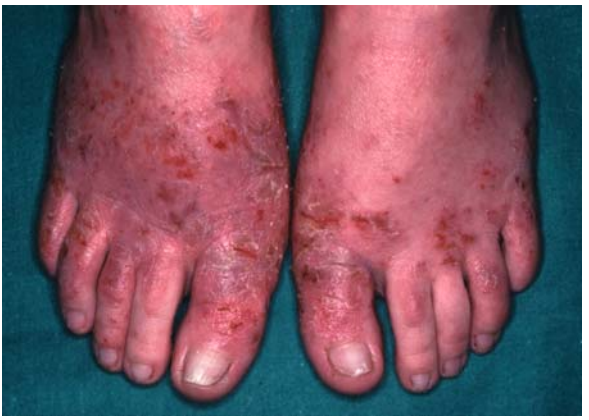

Fig. A.11 Allergic contact dermatitis to rubber used in shoe manufacture. The topography of oedematous, erythematosquamous eczema is highly typical. The mercaptobenzothiazole patch test was positive

the recent literature [5]. Concomitantly, having detailed information on shoe construction and all component chemicals is a helpful and ideal approach in diagnosing shoe allergy. However, this information is often hard to obtain from the manufacturer. In spite of this, the step is crucial for further advice in the choice of alternative shoes.

A.13.2 Plant Dermatitis

Plant dermatitis (phytodermatitis) includes a large variety of skin reactions. The most frequent are mechanical and/or chemical irritation, allergic (sometimes photoworsened)

contact dermatitis, phototoxic and/or photoallergic contact dermatitis (photophytodermatitis), contact (immunological or non-immunological) urticaria, and protein contact dermatitis. A classical example of ACD is primula dermatitis (Fig. A.12a, b).

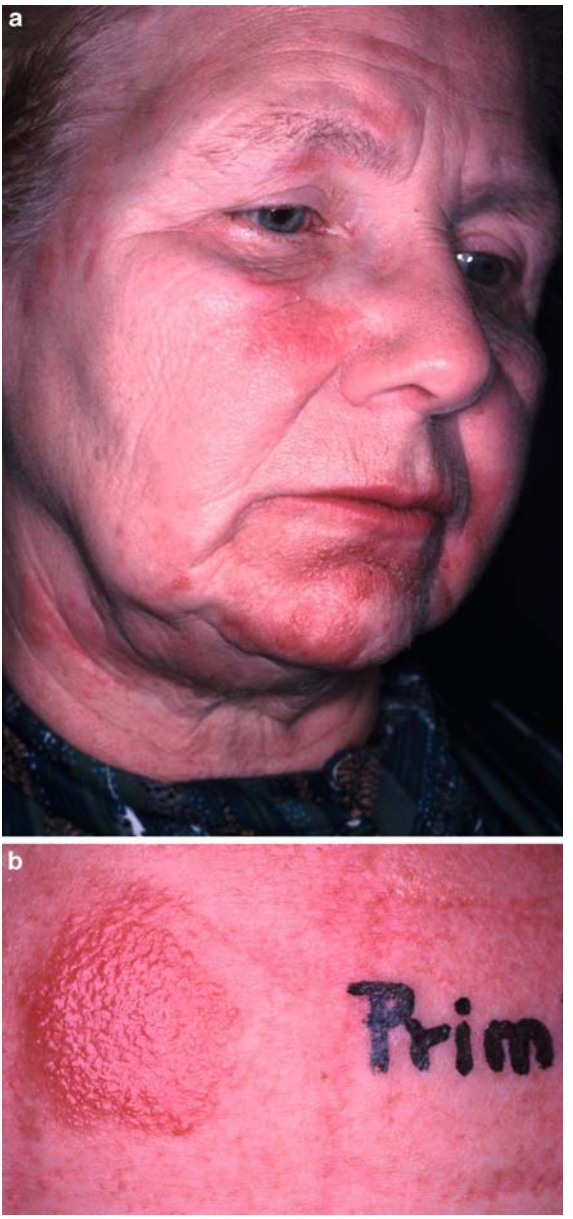

Fig. A.12 Allergic contact dermatitis to *Primula obconica*. **(a)** The lesions are handborne and in the present case affect the temples, cheeks, chin, and neck. **(b)** The patch test to primin was positive, scored ++

Facing such a diversity of reactions is a difficult diagnostic task for the dermatologist. When ACD to a presumably well-identified plant is suspected, different steps of investigation can be undertaken.

Step 1: Patch tests with a few grams of fresh plant material are easy to be carried out. It is important that patch tests are performed to several plant pieces (Fig. A.13a, b) such as roots, stems, leaves, and reproductive organs (flowers and/or fruits). In addition, it is wise to test crushed leaves or slices of stem [6]. Woods (either indigenous or tropical) should not be tested as is, because of the risk of irritation or active sensitization. Wood dust can be tested in petrolatum, 10–20% (weight/weight). Irritant reactions are frequent with plant materials, and have to be considered when doubtful or weakly positive reactions are observed.

Step 2: Patch testing with plant extracts is a useful tool of investigation. Most plant allergens are likely to be soluble in acetone, ethanol, or ether. Thus, a filtered acetone or ethanol extract of dried plant material or a short ether extract of fresh material usually produces a

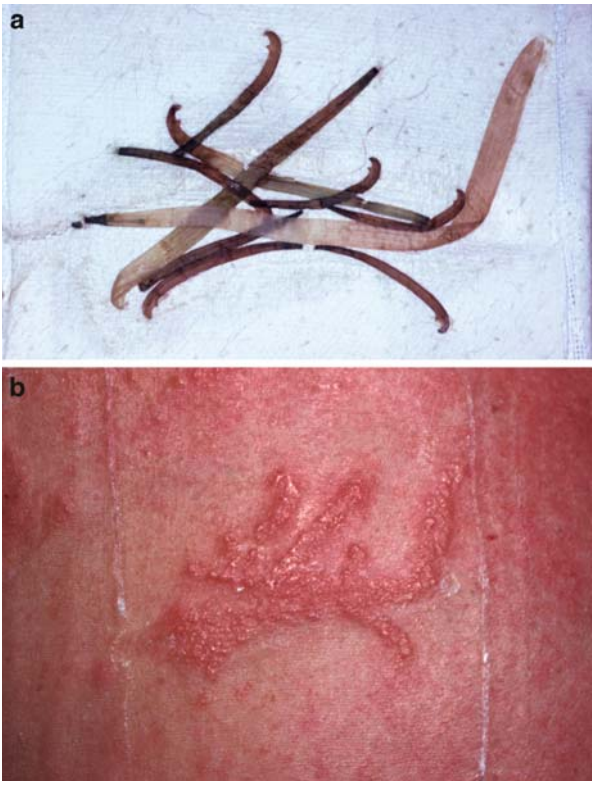

Fig. A.13 Positive allergic patch test reactions to chrysanthemum. **(a)** Various parts of the plant applied to the skin. **(b)** Positive allergic patch test reaction at 48 h

solution suitable for patch testing. Water extracts are not recommended due to chemical degradation [6]. A similar approach is also suitable for indigenous or tropical woods. Photopatch testing (see Chap. 5) is obviously the tool of investigation for photoallergic contact dermatitis.

Step 3: Some commercial allergens are of great value when they are used for the identification of ACD to a well-defined category of plants. They are used individually, but never as a series. The most important allergens and their relationship with plant families are listed in Table A.12.

Contact urticaria and protein contact dermatitis to plants are investigated by prick tests (see Sect. 11.2).

This succinct presentation of plant dermatitis and its approach for a correct diagnosis is basic. Careful reading of chapters of books [6] and/or books entirely devoted to plant dermatitis (see Suggested Reading) is highly advisable.

Table A.12 List of plant allergens in relationship with plant families, to be adapted to each individual case

Allergens	Plants and/or sources of exposure	Concentration (%)
Achillea millefolium extract	Yarrow	1
Arnica montana extract	Mountain tobacco	0.5
Chamomilla romana (Anthemis nobilis) extract	Roman chamomile	1
Chrysanthemum cinerarii folium extract	Pyrethrum	1
Diallyldisulfide	Garlic (cooks)	1
Lichen acid mix (atranorin, usnic acid, evernic acid)	Lichens	0.3
α-methylene-γ-butyrolactone	Tulipa, Alstroemeria, Bonarea, Disocorea hispida, Erythronium, Gagea, Fritillaria	0.01
Παρthenolide	Tanacetum parthenium (feverfew), Parthenium hysterophorus L. (congress grass)	0.1
Primin	Primula obconica, Primulaceae	0.01
Propolis	Beekeepers, medications	10
Sesquiterpene lactone mix	Asteraceae/Compositae, Jubulaceae (Frullania)	0.1
Tanacetum parthenium extract	Feverfew	1
Tanacetum vulgare extract	Tansy	1
Taraxacum officinale extract	Dandelion	2.5

Concentrations refer to petrolatum

Note: Urushiol is a generic name that indicates a mixture of several close alkylcatechols contained in the sap of the Anacardiaceae. It is not commercially available, but is present in the standard series of the JCDS, at the concentration of 0.002% (see Sect. 4.3). It is a marker for poison ivy, poison oak, Lithrea, lacquer tree, and cashew nut tree dermatitis

References

1. Dillarstone A (1997) Cosmetic preservatives. Letter to the Editor. Contact Dermatitis 37:190
2. Johansen JD, Veien N, Laurberg G, Avnstorp C, Kaaber K, Andersen KE, Paulsen E, Sommerlund M, Thormann J, Nielsen NH, Vissing S, Kristensen O, Kristensen B, Agner T, Menné T (2008) Decreasing trends in methyldibromoglutaronitrile contact allergy – following regulatory intervention. Contact Dermatitis 59:48–51
3. Rastogi SC, Johansen JD (2008) Significant exposures to isoeugenol derivates in perfumes. Contact Dermatitis 48:278–281
4. White JML, White IR, Glendinning A, Fleming J, Jefferies D, Basketter DA, Mc Fadden JP, Buckley DA (2007) Frequency of allergic contact dermatitis to isoeugenol is increasing: a review of 3636 patients tested from 2001 to 2005. Br J Dermatol 157:580–582
5. Taylor JS, Erker E, Podmore P (2006) Shoes. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 703–716
6. Le Coz CJ, Ducombs G (2006) Plants and plant products. In: Frosch PJ, Menné T, Lepoittevin JP (eds) Contact Dermatitis, 4th edn. Springer, Berlin, pp 751–800

Suggested Reading

1. Adams RM (1990) Occupational Skin Disease, 2nd edn. WB Saunders, Philadelphia, PA, USA
2. Agache P, Humbert P (2004) Measuring the Skin. Non-Invasive Investigations, Physiology, Normal Constants. Springer, Berlin
3. Amin S, Lahti A, Maibach HI (1997) Contact Urticaria Syndrome, CRC Press, Boca Raton, FL
4. Angelini G, Bonamonte D (2001) Aquatic Dermatology. Springer, Berlin
5. Angelini G, Vena GA (1997) Dermatologia professionale e ambientale. ISED, Brescia, Italy
6. Avalos J, Maibach HI (1999) Dermatologic Botany. CRC Press, Boca Raton, FL
7. Bandmann H-J, Dohn W (1967) Die epicutantestung. Bergmann Verlag, München
8. Bandmann H-J, Fregert S (1982) Epicutantestung. Einführung in die praxis, 2nd edn. Springer, Berlin
9. Benezra C, Ducombs G, Sell Y, Foussereau J (1985) Plant Contact Dermatitis. B.C. Decker, Toronto
10. Boman A, Estlander T, Wahlberg JE, Maibach HI (2004) Protective Gloves for Occupational Use, 2nd edn. CRC Press, Boca Raton, FL
11. Breathnach SM, Hintner H (1992) Adverse Drug Reactions and the Skin. Blackwell, Oxford
12. Burns T, Breathnach S, Cox N, Griffiths Ch (2004) Rook's Textbook of Dermatology, 7th edn. Blackwell, Oxford
13. Carter RL (1992) A Dictionary of Dermatologic Terms, 4th edn. Williams and Wilkins, Baltimore, Maryland, USA
14. Chew A-L, Maibach HI (2006) Irritant Dermatitis. Springer, Berlin
15. Chowdhury MMU, Maibach HI (2005) Latex Intolerance: Basic Science, Epidemiology and Clinical Management. CRC Press, Boca Raton, FL
16. Cronin E (1980) Contact Dermatitis. Churchill Livingstone, Edinburgh
17. de Groot AC (2008) Patch testing. Test Concentrations and Vehicles for 4350 Chemicals, 3rd edn, acdegroot publishing, Schipsbootweg 5, NL 8351 HV Wapserveen
18. Foussereau J, Benezra C (1970) Les eczémas allergiques professionnels. Masson, Paris
19. Foussereau J, Benezra C, Maibach HI (1982) Occupational Contact Dermatitis: Clinical and Chemical Aspects. Munksgaard, Copenhagen
20. Freedberg IM, Eisen AZ, Wolff K, Austen KF, Goldsmith LA, Katz LI (2003) Fitzpatrick's Dermatology in General Medicine, 6th edn. McGraw-Hill, New York
21. Fregert S (1981) Manual of Contact Dermatitis, 2nd edn. Munksgaard, Copenhagen
22. Frosch PJ, Menné T, Lepoittevin J-P (2006) Contact Dermatitis, 4th edn. Springer, Berlin
23. Guy RH, Hostynek JJ, Hinz RS, Lorence CR (1999) Metals and the Skin. Topical Effects and Systemic Absorption. Marcel Dekker, New York
24. Kanerva L, Elsner P, Wahlberg JE, Maibach HI (2000) Handbook of Occupational Dermatology. Springer, Berlin

25. Kauppinen K, Alanko K, Hannuksela M, Maibach HI (1998) Skin Reactions to Drugs. CRC Press, Boca Raton, FL
26. Lachapelle JM, Frimat P, Tennstedt D, Ducombs G (1992) Précis de dermatologie professionnelle et de l'environnement. Masson, Paris
27. Lepoittevin JP, Basketter DA, Goossens A, Karlberg AT (1998) Allergic Contact Dermatitis. The Molecular Basis. Springer, Berlin
28. Litt JZ (2008) Litt's Drug Eruption Reference Manual Including Drug Interactions, 14th edn. Informa Healthcare, London
29. Lovell CR (1993) Plants and the Skin. Blackwell, Oxford
30. Maibach HI, Menné T (1989) Nickel and the Skin: Immunology and Toxicology. CRC Press, Boca Raton, FL
31. Malten KE, Nater J-P, Van Ketel WG (1976) Patch Testing Guidelines. Dekker and van de Vegt, Nijmegen
32. Marks JG, Elsner P, DeLeo V (2002) Contact and Occupational Dermatology, 3rd edn. Mosby, St. Louis
33. Menné T, Maibach HI (2000) Hand Eczema, 2nd edn. CRC Press, Boca Raton, FL
34. Mulder WMC, Meinardi MMHM, Bruynzeel DP (2009) Guide to Adverse Drug Reactions. Side Effects in Dermatology, 9th edn. IMP, Amsterdam
35. Pichler WJ (2007) Drug Hypersensitivity. Karger, Basel
36. Rietschel RL, Conde-Salazar L, Goossens A, Veien NK (1999) Atlas of Contact Dermatitis. Martin Dunitz, London
37. Rietschel RL, Fowler JF Jr (2008) Fisher's Contact Dermatitis, 6th edn, BC Decker, Hamilton, Ontario
38. Saurat J-H, Lachapelle J-M, Lipsker D, Thomas L (2009) Dermatologie et infections sexuellement transmissibles, 5th edn. Elsevier-Masson, Paris
39. Schliemann S, Elsner P (2007) Skin Protection. Practical Applications in the Occupational Setting. Karger, Basel
40. Sell Y, Benezra C, Guérin B (2002) Plantes et réactions cutanées. John Libbey Eurotext, Montrouge, France
41. van der Valk PGM, Maibach HI (1995) The Irritant Contact Dermatitis Syndrome. CRC Press, Boca Raton, FL
42. Wahlberg JE, Elsner P, Kanerva L, Maibach HI (2003) Management of Positive Patch Test Reactions. Springer, Berlin
43. Williams H, Bigby M, Diepgen T, Herxheimer A, Naldi L, Rzany B (2008) Evidence-Based Dermatology, 2nd edn. Wiley-Blackwell, London
44. Zhai H, Maibach HI, Wilhelm KP (2007) Marzulli and Maibach's Dermatotoxicology, 7th edn, CRC Press, Boca Raton, FL
45. Zürcher K, Krebs A (1992) Cutaneous Drug Reactions. An Integral Synopsis of Today's Systemic Drugs. Karger, Basel

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