BJCP British Journal of Clinical Pharmacology

DOI:10.1111/bcp.12297

Immediate-type hypersensitivity drug reactions

Shelley F. Stone,[1,2] Elizabeth J. Phillips,[3,4] Michael D. Wiese,[5] Robert J. Heddle[6,7] & Simon G. A. Brown[1,2]

[1]Centre for Clinical Research in Emergency Medicine, Harry Perkins Institute of Medical Research and the University of Western Australia, Perth, Western Australia, [2]Department of Emergency Medicine, Royal Perth Hospital, Perth, Western Australia, [3]Institute for Immunology and Infectious Diseases, Murdoch University, Perth, Western Australia, Australia, [4]Vanderbilt University School of Medicine, Nashville, Tennessee, USA, [5]School of Pharmacy and Medical Sciences, University of South Australia, Adelaide, South Australia, [6]Clinical Immunology Unit, Royal Adelaide Hospital, Adelaide, South Australia and [7]Division of Human Immunology, SA Pathology, Adelaide, South Australia, Australia

Correspondence
Dr Shelley F. Stone, Centre for Clinical Research in Emergency Medicine, Harry Perkins Institute of Medical Research, Level 5, MRF Building, Rear 50 Murray St, Perth, WA 6000, Australia.
Tel.: +618 9224 0356
Fax: +618 9224 1494
E-mail: shelley.stone@uwa.edu.au

Keywords
adverse drug reaction, anaphylaxis, IgE-mediated, immediate-type

Received
3 September 2013
Accepted
18 November 2013
Accepted Article Published Online
29 November 2013

Hypersensitivity reactions including anaphylaxis have been reported for nearly all classes of therapeutic reagents and these reactions can occur within minutes to hours of exposure. These reactions are unpredictable, not directly related to dose or the pharmacological action of the drug and have a relatively high mortality risk. This review will focus on the clinical presentation, immune mechanisms, diagnosis and prevention of the most serious form of immediate onset drug hypersensitivity reaction, anaphylaxis. The incidence of drug-induced anaphylaxis deaths appears to be increasing and our understanding of the multiple and complex reasons for the unpredictable nature of anaphylaxis to drugs is also expanding. This review highlights the importance of enhancing our understanding of the biology of the patient (i.e. immune response, genetics) as well as the pharmacology and chemistry of the drug when investigating, diagnosing and treating drug hypersensitivity. Misdiagnosis of drug hypersensitivity leads to substantial patient risk and cost. Although oral provocation is often considered the gold standard of diagnosis, it can pose a potential risk to the patient. There is an urgent need to improve and standardize diagnostic testing and desensitization protocols as other diagnostic tests currently available for assessment of immediate drug allergy are not highly predictive.

Introduction

A variety of adverse reactions can occur within minutes to hours of exposure to a drug. Some can be related to the pharmacological action of the drug (WHO Adverse Reaction Terminology type 'A' for 'augmented') and usually have a low mortality [1]. Others are not readily predictable based on the structure and pharmacological action of the drug and have a relatively high mortality risk (Type 'B' for 'bizarre'). Type B reactions include immunologically-mediated reactions and a variety of idiosyncratic reactions such as acute porphyria and malignant hyperthermia.

Hypersensitivity is broadly defined as '*objectively reproducible symptoms or signs, initiated by exposure to a defined stimulus at a dose tolerated by normal subjects*' and may be caused by immunologic (allergic) and non-immunologic mechanisms [2]. Immunological mechanisms can be dependent on the presence of IgE, in which case reactions tend to start rapidly after exposure. Alternatively they may be independent of IgE, in which case they can occur either rapidly or after many hours, particularly if the mechanism is T-cell-mediated. Anaphylaxis is a severe, life-threatening, generalized (systemic) hypersensitivity reaction that has a rapid onset after exposure [3]. In the same way as hypersensitivity reactions, anaphylaxis may be attributed to immunologic mechanisms (either IgE-dependent or IgE-independent), or non-immunologic mechanisms [4]. The terms 'anaphylactoid' and 'pseudoallergic', referring to reactions that are not IgE-dependent, are discouraged in favour of the above aetiological classification, which is more descriptive and avoids the impression that non-IgE mediated reactions might be less serious (which they are not).

Late onset hypersensitivity reactions are often defined as those occurring >72 h after drug administration and these are usually T-cell-mediated and generally lack the

rapidly evolving life-threatening features that are typical of anaphylaxis [5, 6]. However, timing from exposure to reaction onset is variable and it is becoming apparent that T-cell and other cellular mechanisms may be important for accelerated reactions (within 1–72 h) and immediate onset (within 1 h) hypersensitivity.

Immediate onset hypersensitivity including anaphylaxis has been reported for nearly all classes of therapeutic reagents, including antibiotics, anticonvulsants, anaesthetics, neuromuscular blocking drugs (NMBD), chemotherapeutic drugs and non-steroidal anti-inflammatory drugs (NSAIDs). This review will focus on the clinical presentation, immune mechanisms, diagnosis and prevention of the most serious form of immediate onset drug hypersensitivity reaction, anaphylaxis.

Clinical presentation and management of the acute episode

Initial assessment

Immediate hypersensitivity drug reactions are dynamic in nature and can rapidly progress under observation, ranging in severity from trivial to lethal. Different reaction patterns are summarized in Table 1 [7, 8]. Observation for at least 1 h is required to ensure that a mild reaction is not progressing. Skin features may be absent in around 20% of cases [7, 9] and sudden cardiovascular collapse or even cardiac arrest can occur without any apparent skin features. Therefore, a provisional diagnosis and treatment as anaphylaxis is warranted if there is acute life-threatening bronchospasm or hypotension in the right context, i.e. no other diagnosis to explain the hypotension.

Diagnosis

In most cases the diagnosis of immediate-type hypersensitivity can be made clinically (Table 1), but where typical features are not present the collection of serial blood samples to measure mast cell tryptase (MCT) can help to confirm diagnosis. Changes in MCT levels over time are more predictive for anaphylaxis than single measurements and a persistently high MCT concentration will prompt investigation for an underlying mast cell disorder (i.e. mastocytosis) [10, 11]. Serial MCT testing is specific but not sensitive (50–80%), although sensitivity is higher for more severe reactions. Given this low negative predictive value, a negative MCT cannot be used to exclude anaphylaxis.

Management

Anaphylaxis is an uncommon emergency that often has to be treated by doctors with limited resuscitation skills and experience. All clinical practices that may be called on to treat anaphylaxis should have immediate access to a resuscitation trolley and written management prompts, for example the Australian Prescriber Anaphylaxis Wallchart (http://www.australianprescriber.com/magazine/34/4/artid/1210). The mainstays of immediate treatment of a severe reaction are to stop administration of the causative drug, call for assistance, place patient in a supine posture, administer intramuscular adrenaline and provide airway support. For reactions with hypotension, additional measures may be needed including aggressive fluid resuscitation with up to 5 l of normal saline in the first 30 min [12], intravenous infusion of adrenaline, and other potent vasoconstrictors such as metaraminol or vasopressin if the response to adrenaline is inadequate [13]. National guidelines for anaphylaxis treatment should be followed in these situations. Steroids have no proven role and are therefore not recommended for routine use [14]. However they may be considered for reactions with severe protracted wheeze or for skin reactions where a T-cell mechanism is thought likely (see below). There is likewise little evidence to support the use of antihistamines [15] and indeed parenteral antihistamines can themselves induce hypotension in previously stable patients [16]. Oral antihistamines are often used to provide symptomatic relief from itch.

Table 1

Clinical patterns of acute drug hypersensitivity reactions [7, 8]

Subjective reaction
Itch, dizziness/light-headedness, nausea, chest discomfort *but without any objective skin features, physical signs or physiological compromise*.
Skin only reaction
Generalized erythema, urticaria or angioedema without any sentinel features (see below) of other organ involvement
Anaphylaxis
Typical anaphylaxis: Any skin feature listed above *plus* at least one sentinel feature from either the respiratory *or* cardiovascular system: • *Respiratory*; dyspnoea, wheeze, stridor, hypoxaemia (cyanosis or $SpO_2 \leq 92\%$) • *Cardiovascular*; hypotension (SBP < 90 mmHg in an adult), collapse, altered conscious state, incontinence
Atypical anaphylaxis: Severe gastrointestinal features are commonly associated with, or may precede, anaphylactic shock. Also, skin features may be absent in up to 20% of anaphylaxis cases. Therefore, a sudden onset of symptoms affecting skin plus gastrointestinal tract (cramping abdominal pain, vomiting), or features affecting two or more organ systems (any two of gastrointestinal, respiratory, cardiovascular) or even severe bronchospasm or hypotension alone without any other features may justify a provisional diagnosis of anaphylaxis in the right context (see text).
Severe anaphylaxis: Anaphylaxis as defined above is classified as **'severe'** if there is hypoxaemia, hypotension, collapse, altered consciousness or incontinence.

Epidemiology

Data on the incidence of drug-induced anaphylaxis are limited, with most studies referring to specific drugs, special patient conditions or reporting a series of case studies. Under-reporting is an issue for most countries. In an Australian study of anaphylaxis fatalities between 1997 and 2005, drug-induced anaphylaxis deaths appeared to be increasing, in contrast to anaphylaxis deaths due to other causes [17]. In a study of 315 anaphylactic reactions presenting to Australian Emergency Departments, 29.5% were due to oral medication, injected medication or injected diagnostic contrast [8]. Severe reactions characterized by hypotension, hypoxia or both, were associated with older age, pre-existing lung disease and drug causation [8]. The incidence of IgE-mediated hypersensitivity reactions during anaesthesia has been estimated at 100.6 (95% CI 76.2, 125.3) per million procedures [18]. Intra- and peri-operative anaphylaxis is associated with significant morbidity and a reported mortality of between 3.5% and 10%, with NMBDs most commonly implicated [19, 20]. In a study of 14.5 million Dutch individuals conducted over 2 years, the incidence of drug-induced anaphylaxis was 3.7 per million persons annually [21]. A review of 16 157 adverse drug reactions (ADRs) reported to the Portuguese Pharmacovigilence System between 2000–2010, identified 918 cases of anaphylaxis of which 24 (3%) were fatal [22]. Antibiotics were responsible for the majority of cases, followed by NSAIDs/paracetamol (acetaminophen), antineoplastic/cytotoxic drugs and immune-modulators [23]. Vaccines and radiographic contrast mediators were also important triggers of reactions classified as anaphylaxis.

Immune mechanisms

To understand the reasons for the unpredictable nature of anaphylaxis to drugs, it is important to have an understanding of the biology of the patient (i.e. immune response, genetics) as well as the pharmacology and chemistry of the drug. Most clinically relevant immune-mediated immediate-type reactions to drugs are thought to be either IgE-mediated or T-cell mediated [24, 25]. Generally, the more delayed the reaction is, the more likely it is to be T-cell mediated [26], with some notable exceptions being abacavir hypersensitivity in human immunodeficiency virus (HIV) patients and piperacillin reactions in cystic fibrosis patients. However, there may be considerable overlap in the timing of symptoms according to cause, and a significant proportion of IgE-mediated hypersensitivity reactions start >1 h after exposure [26, 27].

IgE-mediated adverse drug reactions

Upon first exposure, free or conjugated drug is taken up by dendritic cells, processed and presented to T and B-cells within the context of a Th2 response, resulting in production of allergen-specific IgE antibodies to the drug or a drug-protein complex. On subsequent exposure, the drug or drug–protein complex is recognized by IgE antibodies bound to their high affinity receptor on the surface of mast cells and basophils. Cross-linking of IgE leads to activation of a calcium-dependent protein kinase cascade and the subsequent release of inflammatory mediators such as histamine, prostaglandin D_2, sulfidoleukotrines, MCT and various cytokines [8, 28].

It is noteworthy that for a number of (presumed) immediate hypersensitivity reactions, drug-specific IgE is not detected. In penicillin allergy just 25–54% of patients with a positive skin test or oral challenge have detectable specific IgE [29, 30]. This is probably due to a lack of sensitivity of available assays for allergen-specific IgE and as such, many drug-mediated immediate hypersensitivity reactions are presumed to be IgE-mediated on the basis of clinical presentation and/or skin test findings without IgE being definitively identified. The debate about whether IgE has a significant role in radiocontrast reactions continues, with an emerging recognition that many radiocontrast media reactions may in fact be IgE mediated [31].

Direct cross-linking, haptenation and pro-haptens A few drugs with large molecular weights have multiple recurrences of a single epitope and thus are able to cross-link IgE molecules directly [32–34]. The best studied example of this is quaternary ammonium epitopes, which render some NMBDs multivalent.

Smaller molecular weight drugs (i.e. <1000 Da) are unable to directly cross-link IgE on their own. The hapten hypothesis is that chemically reactive small molecules called haptens are able to undergo a stable covalent binding to larger proteins. Haptenation thus results in an alteration of autologous proteins by drug epitopes and a drug-specific immune response can ensue. The best-described examples are penicillin antibiotics, which are chemically reactive and conjugate primarily to lysine residues on autologous proteins. Whilst there was evidence over 50 years ago for benzylpenicillin binding to human serum albumin (HSA) at pH 7.5 to 8.0 [35], it has only recently become apparent that various penicillin antibiotics (flucloxacillin, benzylpenicillin and piperacillin) preferentially react with different lysine residues on HSA. There are differences between individuals with respect to which lysine residues of HSA are modified and the binding is both concentration and time dependent [36–38]. Additional studies have identified that amoxicillin also binds to ferritin and other unidentified plasma proteins [39], and in mice, benzylpenicillin binds to a number of autologous spleen and plasma proteins [40]. It has also been hypothesized that penicillins can directly conjugate to the MHC molecules on antigen presenting cells [41]. The hapten that is thought to be predominantly

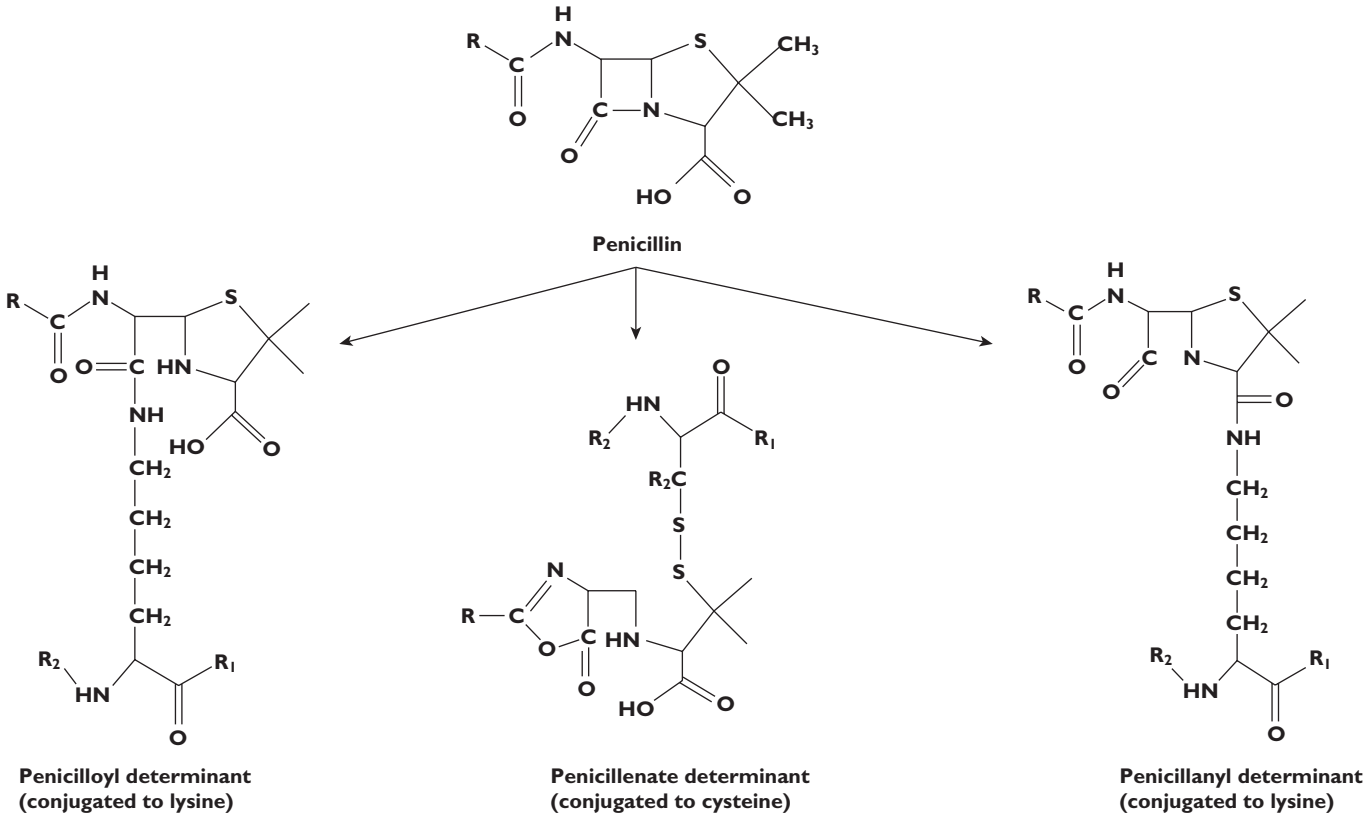

Figure 1

Chemical conjugation of penicillins to endogenous proteins. Penicillins undergo chemical rearrangement to form various covalent conjugates with endogenous proteins. The most common reaction is the formation of the penicilloyl determinant (also known as the major determinant), where the beta-lactam ring opens and attaches to the free amine group within lysine residues. Other less common products are collectively referred to as minor determinants and include the formation of the penicillanyl determinant, which typically involves attachment of the free carboxylic acid (attached to the thiazolidine ring) to the free amine group on lysine residues and the penicillenate determinant, which typically involves conjugation to the thiol group of cysteine residues within proteins. R penicillin side chain, R1 and R2 refer to the remainder of the autologous protein to which the penicillin has attached

responsible for penicillin allergy is the penicilloyl epitope (the major determinant, comprising about 95% of penicillin conjugated to autologous proteins, which is formed following opening of the β-lactam ring and acetylation of amine groups on target proteins), although other chemical modifications (referred to as minor determinants), such as penicillenate and penicillanyl have been described (Figure 1) [42].

While the hapten hypothesis adequately explains conjugation of penicillins to autologous proteins, most drugs are chemically inert. The prohapten hypothesis states that some form of metabolism, typically via cytochrome P450 (CYP450) enzymes in the liver or skin is required in order to bind covalently to autologous proteins [43]. In some cases, the metabolite is so reactive that it spontaneously reacts with the CYP450 enzyme that catalyzed its formation [44]. The drug that has been most studied in this regard is sulfamethoxazole, although it has primarily been investigated in the context of T-cell mediated hypersensitivity reactions. Sulfamethoxazole is metabolized via CYP450 enzymes (CYP2C9) to sulfamethoxazole hydroxylamine [45], which spontaneously oxidizes to a nitroso intermediate (sulphamethoxazole-nitroso) [46], which is able to conjugate with cysteine residues on autologous proteins [47].

NSAID hypersensitivity Shared aspirin and NSAID hypersensitivity, including aspirin-exacerbated respiratory disease (AERD) and urticaria-angioedema without respiratory features, have long been thought of as a pharmacological reaction, mediated via cyclo-oxygenase I (COX-1) inhibition by aspirin and non-selective NSAIDs and reduction in production of prostaglandin E_2, thereby increasing synthesis of cysteinyl-leukotrienes and release of various mast cell mediators such as histamine and prostaglandin D_2 (so called leukotriene shunt) [48]. These reactions present shortly after drug exposure as rhinitis, angioedema, urticaria and bronchoconstriction, and

often occur after first exposure to an aspirin or NSAID (i.e prior sensitization is not required) in individuals with asthma, nasal polyposis or a history of chronic urticaria [49]. Approximately 10% of adult patients with asthma and 40% of patients with nasal polyps demonstrate this hypersensitivity to aspirin/NSAIDs. When given at high enough doses, all inhibitors of COX-1 (i.e. aspirin, non-selective NSAIDs) will precipitate these reactions [48], and the COX-2 selective inhibitors do not cross react, although there have been occasional reports to the contrary [50]. Paracetamol (acetominophen) inhibits COX-1 in high doses (i.e. >1000 mg/dose or >4 g day^{-1}), and have also been reported in association with this syndrome [51].

More recently, individuals who experience selective and immediate hypersensitivity reactions to a single NSAID, most commonly diclofenac, have been recognized [49]. These reactions present similarly to IgE mediated immediate hypersensitivity reactions, but the search for drug-specific IgE has proven to be largely elusive. Exceptions include the demonstration of specific IgE in 58% of individuals with a strong clinical history of anaphylaxis to propyphenazone [52], and the detection of IgE in 17 of 19 patients who were allergic to metamizole [53]. Diclofenac-specific IgE has been detected in a single patient [54], but another group could not detect specific IgE to diclofenac or five phase I metabolites that were bound to HSA in 59 patients (41 of whom were considered selective reactors to diclofenac) [55].

Given that drug specific IgE has rarely been identified, the possibility of non-IgE mediated mechanisms should be considered. However, NSAID-specific IgE may be present, yet undetectable by current assays. Unlike penicillins, NSAIDs are not inherently chemically reactive, and require some form of metabolism before they are able to conjugate with endogenous proteins. Zomepirac and tolmetin are NSAIDs that both require metabolism to protein reactive acyl glucuronides (phase II metabolites), and further oxidative metabolism prior to haptenization with autologous proteins [56]. Acyl glucuronide formation is also a metabolic pathway of diclofenac, and subsequent oxidative metabolism produces 4′-hydroxydiclofenac acyl glucuronide [57]. Conjugation of diclofenac acyl glucuronide to proteins in rat hepatocytes [58] and HSA [59] has also been demonstrated, indicating that diclofenac-specific IgE may be directed against additional metabolites and/or target proteins. The role of metabolism in mediating NSAID reactions is also supported by the observation of longer times from exposure to reaction onset in NSAID reactions compared with antibiotic reactions [8].

Is prior exposure required for the development of triggering IgE antibodies? Recent data suggest that previous exposure to the causative drug may not be an obligatory prerequisite for immune-mediated drug hypersensitivity. In patients treated for cetuximab-induced anaphylaxis, drug-specific IgE was found in pretreatment samples [60]. The antibodies were specific for the oligosaccharide galactose-α-1,3-galactose (alpha-gal), which is present on the Fab portion of the cetuximab heavy chain and is also very similar to substances in the ABO blood group. IgE mediated reactions and this type of cross-reactivity do not occur with infliximab or other large molecules where the alpha-gal moiety is in the Fc portion rather than the Fab portion. Allergic reactions to NMBAs are almost exclusively IgE-mediated. However, up to 75% of reactions have been reported upon first known contact with an NMBA [61, 62]. This suggests a possible cross-reaction with IgE antibodies generated by previous contact with apparently unrelated chemicals. Several studies support the hypothesis that pholcodine exposure could either lead to IgE-sensitization to this drug and other quaternary ammonium ions or increase the titre of specific IgE to quaternary ammonium ions, thereby increasing the risk of allergic reaction to NMBAs [63, 64].

T-cell mediated adverse drug reactions

All IgE-mediated ADRs require T-cell help, with T-cells contributing to immediate reactions through production of pro-inflammatory mediators and cytotoxicity. In specific patient populations, such as cystic fibrosis patients exposed to repeated courses of antibiotics, many immediate reactions (i.e. to piperacillin) are T-cell mediated [65, 66]. Currently, three different models have been proposed to account for the stimulation of T-cells by drugs, the hapten model, the pharmacological-interaction (p-i) model and the altered peptide repertoire model.

In the hapten model, compounds bind to certain amino acids via covalent bonds, with or without previous metabolism of the drug. These hapten-modified proteins are then processed into antigenic peptides and loaded onto MHC molecules on antigen presenting cells and activate T-cells [67]. The second model for drug interactions with T-cells, the p-i concept, has been proposed whereby a chemically inert drug in its native form without binding to a carrier molecule or being otherwise processed can non-covalently and processing-independently bind directly to HLA molecules or T-cell receptors (TCR) [68]. If the drug fits with a sufficient affinity into such molecules, the interaction via non-covalent bonds with proteins is strong enough to transmit a stimulatory signal via TCR leading to a stimulation of T-cells resulting in cytokine production, proliferation and/or cytotoxicity [69]. T-cell mediated reactions to drugs also do not require previous exposure to the drug [70].

More recently, strong evidence has supported an altered peptide repertoire model for abacavir hypersensitivity [71–73] and additional evidence suggests that this model may also apply for severe cutaneous ADRs such as carbamazepine-induced Stevens–Johnson syndrome/toxic epidermal necroylsis (SJS/TEN) [72, 74]. In this model,

as per the p-i model, rapid and non-covalent binding occurs between the drug and HLA. However, in the altered peptide repertoire model, the drug occupies key anchor sites within the antigen binding cleft and hence alters the repertoire of self-peptide ligands that are subsequently bound and presented to T-cells, creating an allograft reaction.

Non-immune-mediated drug hypersensitivity

Non-immune-mediated drug hypersensitivity may be caused by the non-specific and non-allergic release of histamine from mast cells. This is not well understood although there are many putative mechanisms for a direct effect on mast cells. Example of drugs causing this effect include vancomycin ('red-man syndrome') and narcotic analgesics. Radiocontrast media and NSAIDs may have direct effects on mast cells, but also have other clinical reaction phenotypes and immunopathogeneses as discussed above.

Snake antivenom reactions

Complement activation has been proposed as the principal mechanism underlying immediate onset reactions to snake antivenom on the basis of *in vitro* studies [75–77]. However, in a recent study of Sri Lankan snake bite victims, patients experiencing anaphylaxis to antivenom had rapid increases in plasma concentrations of MCT and histamine but not complement breakdown products C3a, C4a or C5a [78]. These data suggest anaphylaxis to antivenom is not triggered by complement activation, with possible alternative triggering mechanisms including immunogolublin or protein complexes binding to IgG receptors or non-specifically crosslinking IgE on the surface of mast cells. Shivering, sweating and fever may also occur at the onset of antivenom reactions, with or without other features of anaphylaxis. The mechanism behind these pyrogenic reactions is unknown.

Pharmacogenomics of anaphylaxis and drug allergy

Although it is assumed that there will be genetic determinants driving predisposition to drug-induced anaphylaxis and other IgE mediated immediate drug reactions, supporting data are scarce. Cytokine variants such as TNF-α 308 A→G, IL-13 and IL-4RA, have been entertained, as have variants in expression of IgE receptors on target cells [79]. A single Chinese study postulated that alleles in the HLA-DRB region may be involved in penicillin allergy through modulation of development of penicillin-specific serum IgE [80]. HLA associations using high resolution HLA typing in well phenotyped populations of patients with IgE type, immediate beta-lactam and other immediate and possible IgE mediated drug allergies have not been adequately studied. Other drug-induced reactions may be important to rule out in patients presenting with a potential IgE mediated reaction. This would include angiotensin-converting enzyme inhibitor (ACEI) associated angioedema, which can be sporadic and become apparent several years after these drugs are started. ACEI associated angioedema is likely mediated, at least in part, by decreased breakdown of vasoactive peptides such as bradykinin which is mainly inactivated by aminopeptidase P (APP). A recent meta-analysis of pharmacogenetic studies suggested the gene region encoding XPNPEPE2, an X-linked gene encoding membraneous APP, may be important in ACEI associated angioedema [81, 82]. AERD has been associated with the class 2 HLA allele HLA-DPB1*0301 in European and Asian populations, which was supported by a recent genome wide association study [83, 84]. Part of the challenge in studying the pharmacogenomics of true IgE-mediated reactions is that many of these reactions are not static over the lifetime of an individual. For instance approximately 10% of individuals who have positive skin tests and have experienced immediate reactions to penicillin and other beta-lactams will lose their skin test reactivity each year. In addition, of the 10–15% of hospitalized patients labelled as penicillin allergic, more than 90% will tolerate penicillins [25]. Although the vast majority of these may never have been truly beta-lactam or penicillin allergy, it has been clearly documented that IgE mediated reactions dissipate in some patients over time, making the study of genetic determinants difficult.

In contrast to IgE mediated drug reactions, several recent studies have implicated the major histocompatibility complex (MHC) as a major genetic determinant of T-cell mediated drug hypersensitivity syndromes. This has particularly reference to T-cell mediated and delayed drug hypersensitivities such as drug induced hypersensitivity syndrome (DIHS) or drug reaction with eosinophilia and systemic symptoms (DRESS) and the severe skin syndromes such as SJS/TEN [25]. As described above, the immunopathogenesis of HLA class I restricted drug hypersensitivity reactions such as abacavir hypersensitivity (HLA-B*57:01) or carbamazepine (HLA-B*15:02) associated SJS/TEN has been elegantly elucidated [85]. Unlike drug-related IgE mediated reactions where the risk of a repeat reaction to the same drug may decrease with time, the risk for these T-cell mediated and specifically HLA class I restricted reactions appears to be primarily genetically determined and associated with memory T-cell responses that lead to not only a sustained risk but the potential for a more severe reaction on re-exposure. Therefore lifelong avoidance of the drug in question and all structurally related drugs is recommended. Understanding the basis for HLA–drug interactions shows promise for being able to predict the potential for severe T-cell mediated drug reactions either before drugs are used in man, or in the early pre-marketing phase of drug development.

Clinical history and diagnosis of immediate drug reactions

Several guidelines for the investigation of suspected anaphylaxis have been recently published by anesthesiology and/or allergy societies [86–89]. Accurate diagnosis is essential to prevent future reactions and assess cross-reactivity to predict which drugs from the same class should also be avoided, or which may be used as an alternative. Diagnostic tests for drug allergy are imperfect, so clinical assessment always takes precedent in decision making. This requires a combination of careful history, obtaining all medical records, outlining drug history and ancillary *in vivo* and *ex vivo/in vitro* testing as indicated. The timing of drug administration in relation to development of symptoms can provide invaluable information. In addition, de-challenge and rechallenge information is helpful. Probability scores such as the Naranjo score have been developed in an attempt to ascertain the likelihood that a specific drug is related to a specific ADR and other Bayesian approaches have also been advocated [90]. Several studies have demonstrated that the actual incidence of drug allergy is lower than patients' histories suggest [91–94], and that many patients are labelled with drug allergy, but few actually have a drug allergy on more careful history and testing [25]. Misdiagnosis of drug hypersensitivity leads to substantial unnecessary costs and put patients at risk. Oral provocation is often considered the gold standard diagnostic test, but poses a potential risk to the patient and is contra-indicated in patients with severe T-cell mediated syndromes such as DRESS and TEN [95]. Of the other diagnostic tests currently available for assessment of immediate drug allergy, none is highly predictive.

Skin tests

Standards for prick and intradermal skin testing have not been universally accepted, and a significant variability in specificity and sensitivity has been reported. However, skin tests remain the mainstay of diagnosis of IgE-mediated reactions. Skin prick and intradermal testing (SPT/IDT) may be useful in cases where patients have received multiple drugs and the causative agent of the allergic reaction is unclear [96]. SPT/IDT is the preferred diagnostic test for penicillin allergy. The major (benzylpenicilloyl polylysine (PLL)) and minor (benzylpenicillin, sodium benzylpenilloate and benzylpenicilloic acid) determinants of benzyl penicillin have been validated for use in skin testing, and with appropriate use, there is a negative predictive value of >95% and a positive predictive value of 40–60% [97, 98]. With increasing exposure to other types of penicillins in the community such as amoxicillin and ampicillin, the incidence of side chain specific penicillin reactions to these drugs has increased and utilizing multiple reagents in penicillin skin testing could be recommended [89]. Many patients lose their penicillin allergy over time with less than 30% maintaining their positive skin test for 10 years. Skin testing with antibiotics other than penicillin is not standardized and both the negative and positive predictive value is unknown. Also, drug metabolites or haptens that have been defined for benzyl penicillin have not been defined for most other drugs, and hence skin testing may result in false negative results through lack of sensitivity and appropriate reagents or false positives through irritant reactions (e.g. macrolides and quinolones) [99, 100].

In vitro *testing*

In vitro tests have a high potential utility in the diagnosis of ADRs. However the sensitivity of currently available tests is low. Allergen-specific IgE assays (i.e. ImmunoCAP) are commercially available for the major determinants of penicillin G, penicillin V, ampicillin, amoxicillin and cefaclor and many other drugs including morphine and NMBDs. These have a very low sensitivity compared with SPT/IDT and since SPT/IDT for drugs such as penicillin has proven to be a safe procedure with a high negative predictive value, it is not clear the role these tests would have in routine clinical practice. In view of their very low sensitivity, allergen-specific IgE assays should never be used as a means to rule out drug allergy but may help avoid unnecessary or dangerous oral challenges due to their high specificity [30, 101].

Upon stimulation with specific-allergens that cross-link IgE bound to its high affinity receptor, basophils rapidly express surface activation markers such as CD63 and CD203c. Basophil Activation Tests (BAT) using *in vitro* cell stimulation and flow cytometry assays have been used as a tool to predict *in vivo* responses to allergen [102, 103]. The sensitivity of the BAT for the diagnosis of drug allergy has been reported: beta-lactams 33–67% [104–106], quinolones 71.1% [107] and rocuonium 80% [108]. Assessment of noraminophenazone-induced CD63 expression on basophils has been shown sensitive to detect hypersensitivity to pyrazolones, but testing early after the reactions seems to be critical for the test positivity [109, 110]. Again, these tests lack the sensitivity and negative predictive value needed to be useful as routine diagnostic tests for immediate drug reactions.

Drug provocation testing

As the sensitivity of other tests to diagnose drug allergy are quite low, drug provocation testing (DPT) can be considered for those patients who have tested negative via skin and *in vitro* tests, who have no risk factors and for whom diagnosis is mandatory [111]. The general guidelines for performing a DPT are under strict hospital surveillance with emergency room facilities [89]. It should also be remembered that single drug challenges may be effective in ruling out immediate life-threatening IgE mediated type reactions but will not rule out the presence of a delayed or T-cell mediated reaction which may take more than one

dose of the drug to become clinically apparent, even on re-exposure. DPT should not be used for more dangerous T-cell mediated reactions (DRESS, SJS/TEN).

Secondary prevention/ desensitization

Desensitization can be used when there is a strong clinical indication for choice of a drug over available alternatives in the context of suspected or confirmed allergy to the drug, especially if the previous reaction was an immediate onset reaction. Desensitization should never be attempted in patients who have experienced severe drug related symptoms such as fever, internal organ involvement, mucosal involvement or severe cutaneous involvement such as DRESS or SJS/TEN [112]. Figure 2 provides an outline of the management and treatment of a potential immediate onset drug allergy from diagnosis to desensitization using penicillin as an example. Unfortunately, many *in vitro* and *in vivo* tests available for diagnosing penicillin allergy are not available or feasible as point of care testing or not standardized for other drugs. Although penicillins and other beta-lactams are the most commonly used drugs for which rapid desensitization protocols are currently used, rapid desensitization protocols have been published for other classes of drugs such as chemotherapeutic and small and large molecule biological agents [113–117]. Whenever possible, rapid oral desensitization is preferable to intravenous desensitization since the cost is much lower and the procedure has equivalent efficacy [118]. Desensitization to penicillin and other drugs may be useful in those documented to be skin test positive, although up to 30% of patients may experience adverse effects and this may prolong the time and number of doses over which the desensitisation occurs [119–121]. Desensitization is typically a low risk procedure and may be carried out in the hospital setting with close monitoring, however occasionally in patients with positive immediate prick or intradermal skin tests and a history of severe reactions (e.g. to platinum based chemotherapeutic agents) ICU admission, pre-medication and significant nursing and clinician time is needed because of the high risk of anaphylaxis. However, once desensitization is complete, patients are usually able to tolerate full doses of the drug in question for full treatment length with minimal side effects [119].

The immune mechanisms of successful desensitization remain unclear and unlike airborne allergens and

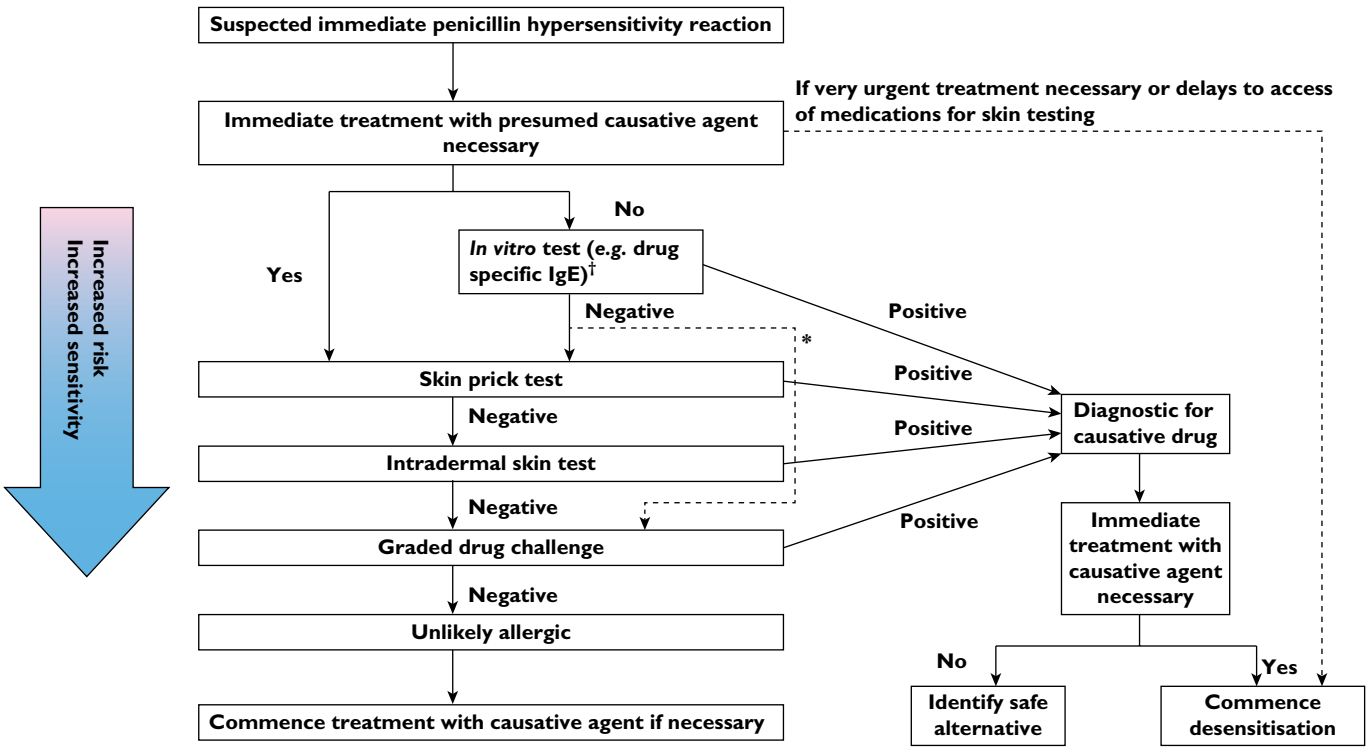

Figure 2

Pathway for diagnosis and treatment of suspected immediate penicillin hypersensitivity reactions. *Potential pathway in children where skin testing is poorly tolerated, in older adults with particularly thin skin or in others if interpretation of skin testing is likely to be limited by skin rash or inherent irritability of suspect medication. †Basophil activation test is available at some centres and has similar risk to other *in vitro* tests and increased sensitivity, but the test is not standardized between laboratories

Hymenoptera venom, desensitization to drugs is a temporary process and must be repeated each time there is treatment interruption [122]. Patients desensitized based on history should generally be referred for follow-up skin testing and oral challenge following completion of their treatment as many will be negative on testing and able to tolerate the drug in the future. More protracted desensitization protocols performed over a longer period of time from several hours to days or weeks also exist for patients who have experienced more delayed and likely T-cell mediated reactions (e.g. trimethoprim-sulfamethoxazole, allopurinol) and the mechanism by which these protocols are successful are similarly unknown.

Competing Interests

All authors have completed the Unified Competing Interest form at http://www.icmje.org/coi_disclosure.pdf (available on request from the corresponding author) and declare no support from any organization for the submitted work.

The authors would like to thank Miss Claire Cotterell for assistance with literature searches.

REFERENCES

1 Edwards IR, Aronson JK. Adverse drug reactions: definitions, diagnosis, and management. Lancet 2000; 356: 1255–9.

2 Johansson SGO, Hourihane JO, Bousquet J, Bruijnzeel-Koomen C, Dreborg S, Haahtela T, Kowalski ML, Mygind N, Ring J, van Cauwenberge P, van Hage-Hamsten M, Wuthrich B. A revised nomenclature for allergy – an EAACI position statement from the EAACI nomenclature task force. Allergy 2001; 56: 813–24.

3 Sampson HA, Munoz-Furlong A, Campbell RL, Adkinson NF, Jr, Bock SA, Branum A, Brown SG, Camargo CA Jr, Cydulka R, Galli SJ, Gidudu J, Gruchalla RS, Harlor AD Jr, Hepner DL, Lewis LM, Lieberman PL, Metcalfe DD, O'Connor R, Muraro A, Rudman A, Schmitt C, Scherrer D, Simons FE, Thomas S, Wood JP, Decker WW. Second symposium on the definition and management of anaphylaxis: summary report – Second National Institute of Allergy and Infectious Disease/Food Allergy and Anaphylaxis Network symposium. J Allergy Clin Immunol 2006; 117: 391–7.

4 Simons FE, Ardusso LR, Bilo MB, El-Gamal YM, Ledford DK, Ring J, Sanchez-Borges M, Senna GE, Sheikh A, Thong BY, World Allergy Organization. World Allergy Organization guidelines for the assessment and management of anaphylaxis. World Allergy Organ J 2011; 4: 13–37.

5 Boguniewicz M, Leung DY. Hypersensitivity reactions to antibiotics commonly used in children. Pediatr Infect Dis J 1995; 14: 221–31.

6 Levine BB. Immunologic mechanisms of penicillin allergy. A haptenic model system for the study of allergic diseases of man. N Engl J Med 1966; 275: 1115–25.

7 Brown SG. Clinical features and severity grading of anaphylaxis. J Allergy Clin Immunol 2004; 114: 371–6.

8 Brown SG, Stone SF, Fatovich DM, Burrows SA, Holdgate A, Celenza A, Coulson A, Hartnett L, Nagree Y, Cotterell C, Isbister GK. Anaphylaxis: clinical patterns, mediator release, and severity. J Allergy Clin Immunol 2013; 132: 1141–9 e5.

9 Brown AF, McKinnon D, Chu K. Emergency department anaphylaxis: a review of 142 patients in a single year. J Allergy Clin Immunol 2001; 108: 861–6.

10 Brown SG, Mullins RJ, Gold MS. Anaphylaxis: diagnosis and management. Med J Aust 2006; 185: 283–9.

11 Brown SG, Stone SF. Laboratory diagnosis of acute anaphylaxis. Clin Exp Allergy 2011; 41: 1660–2.

12 Fisher MM. Clinical observations on the pathophysiology and treatment of anaphylactic cardiovascular collapse. Anaesth Intensive Care 1986; 14: 17–21.

13 Brown SG. Cardiovascular aspects of anaphylaxis: implications for treatment and diagnosis. Curr Opin Allergy Clin Immunol 2005; 5: 359–64.

14 Sheikh A, Shehata YA, Brown SG, Simons FE. Adrenaline for the treatment of anaphylaxis: Cochrane systematic review. Allergy 2009; 64: 204–12.

15 Sheikh A, Ten Broek V, Brown SG, Simons FE. H1-antihistamines for the treatment of anaphylaxis: Cochrane systematic review. Allergy 2007; 62: 830–7.

16 Ellis BC, Brown SG. Parenteral antihistamines cause hypotension in anaphylaxis. Emerg Med Australas 2013; 25: 92–3.

17 Liew WK, Williamson E, Tang ML. Anaphylaxis fatalities and admissions in Australia. J Allergy Clin Immunol 2009; 123: 434–42.

18 Mertes PM, Alla F, Trechot P, Auroy Y, Jougla E, Anaphylactoides GER. Anaphylaxis during anesthesia in France: an 8-year national survey. J Allergy Clin Immun 2011; 128: 366–73.

19 Mertes PM, Aimone-Gastin I, Gueant-Rodriguez RM, Mouton-Faivre C, Audibert G, O'Brien J, Frendt D, Brezeanu M, Bouaziz H, Guéant JL. Hypersensitivity reactions to neuromuscular blocking agents. Curr Pharm Des 2008; 14: 2809–25.

20 Sadleir PH, Clarke RC, Bunning DL, Platt PR. Anaphylaxis to neuromuscular blocking drugs: incidence and cross-reactivity in Western Australia from 2002 to 2011. Br J Anaesth 2013; 110: 981–7.

21 Vanderklauw MM, Stricker BHC, Herings RMC, Cost WS, Valkenburg HA, Wilson JHP. A population-based case-cohort study of drug-induced anaphylaxis. Br J Clin Pharmacol 1993; 35: 400–8.

22 Ribeiro-Vaz I, Marques J, Demoly P, Polonia J, Gomes ER. Drug-induced anaphylaxis: a decade review of reporting to the Portuguese Pharmacovigilance Authority. Eur J Clin Pharmacol 2013; 69: 673–81.

23 Renaudin JM, Beaudouin E, Ponvert C, Demoly P, Moneret-Vautrin DA. Severe drug-induced anaphylaxis: analysis of 333 cases recorded by the Allergy Vigilance Network from 2002 to 2010. Allergy 2013; 68: 929–37.

24 Pichler WJ, Adam J, Daubner B, Gentinetta T, Keller M, Yerly D. Drug hypersensitivity reactions: pathomechanism and clinical symptoms. Med Clin North Am 2010; 94: 645–64, xv.

25 Rive CM, Bourke J, Phillips EJ. Testing for drug hypersensitivity syndromes. Clin Biochem Rev 2013; 34: 15–38.

26 Bircher AJ, Hofmeier KS. Drug hypersensitivity reactions: inconsistency in the use of the classification of immediate and nonimmediate reactions. J Allergy Clin Immun 2012; 129: 263–4.

27 Romano A, Torres MJ, Castells M, Sanz ML, Blanca M. Diagnosis and management of drug hypersensitivity reactions. J Allergy Clin Immun 2011; 127: S67–S73.

28 Stone SF, Cotterell C, Isbister GK, Holdgate A, Brown SG. Elevated serum cytokines during human anaphylaxis: identification of potential mediators of acute allergic reactions. J Allergy Clin Immunol 2009; 124: 786–92.

29 Blanca M, Mayorga C, Torres MJ, Reche M, Moya MC, Rodriguez JL, Romano A, Juarez C. Clinical evaluation of Pharmacia CAP System RAST FEIA amoxicilloyl and benzylpenicilloyl in patients with penicillin allergy. Allergy 2001; 56: 862–70.

30 Fontaine C, Mayorga C, Bousquet PJ, Arnoux B, Torres MJ, Blanca M, Demoly P. Relevance of the determination of serum-specific IgE antibodies in the diagnosis of immediate beta-lactam allergy. Allergy 2007; 62: 47–52.

31 Brockow K, Ring J. Classification and pathophysiology of radiocontrast media hypersensitivity. Chem Immunol Allergy 2010; 95: 157–69.

32 Vervloet D, Arnaud A, Senft M, Didier A, Bongrand P, Charpin J. Anaphylactic reactions to suxamethonium prevention of mediator release by choline. J Allergy Clin Immun 1985; 76: 222–5.

33 Cador D, Senft M, Bongrand P, Fourneron JD, Furstos R, Vervloet D. Role of the quaternary ammonium-ions in the mechanisms of anaphylactic reactions to Succinylcholine (Sch). J Allergy Clin Immun 1985; 75: 150.

34 Didier A, Cador D, Bongrand P, Furstoss R, Fourneron P, Senft M, Philip-Joet F, Charpin D, Charpin J, Vervloet D. Role of the quaternary ammonium ion determinants in allergy to muscle-relaxants. J Allergy Clin Immun 1987; 79: 578–84.

35 Levine BB, Ovary Z. Studies on the mechanism of the formation of the penicillin antigen. III. The N-(d-alpha-benzylpenicilloyl) group as an antigenic determinant responsible for hypersensitivity to penicillin G. J Exp Med 1961; 114: 875–904.

36 Jenkins RE, Meng X, Elliott VL, Kitteringham NR, Pirmohamed M, Park BK. Characterisation of flucloxacillin and 5-hydroxymethyl flucloxacillin haptenated HSA in vitro and in vivo. Proteomics Clin Appl 2009; 3: 720–9.

37 Whitaker P, Meng X, Lavergne SN, El-Ghaiesh S, Monshi M, Earnshaw C, Peckham D, Gooi J, Conway S, Pirmohamed M, Jenkins RE, Naisbitt DJ, Park BK. Mass spectrometric characterization of circulating and functional antigens derived from piperacillin in patients with cystic fibrosis. J Immunol 2011; 187: 200–11.

38 Meng X, Jenkins RE, Berry NG, Maggs JL, Farrell J, Lane CS, Stachulski AV, French NS, Naisbitt DJ, Pirmohamed M, Park BK. Direct evidence for the formation of diastereoisomeric benzylpenicilloyl haptens from benzylpenicillin and benzylpenicillenic acid in patients. J Pharmacol Exp Ther 2011; 338: 841–9.

39 Magi B, Marzocchi B, Bini L, Cellesi C, Rossolini A, Pallini V. Two-dimensional electrophoresis of human serum proteins modified by ampicillin during therapeutic treatment. Electrophoresis 1995; 16: 1190–2.

40 Warbrick EV, Thomas AL, Stejskal V, Coleman JW. An analysis of beta-lactam-derived antigens on spleen cell and serum proteins by ELISA and Western blotting. Allergy 1995; 50: 910–7.

41 Posadas SJ, Pichler WJ. Delayed drug hypersensitivity reactions – new concepts. Clin Exp Allergy. 2007; 37: 989–99.

42 Baldo BA, Pham NH. Immunoglobulin E binding determinants on beta-lactam drugs. Curr Opin Allergy Clin Immunol 2002; 2: 297–300.

43 Pichler WJ, Naisbitt DJ, Park BK. Immune pathomechanism of drug hypersensitivity reactions. J Allergy Clin Immunol 2011; 127 (3 Suppl.): S74–81.

44 Friedmann PS, Lee MS, Friedmann AC, Barnetson RS. Mechanisms in cutaneous drug hypersensitivity reactions. Clin Exp Allergy 2003; 33: 861–72.

45 Cribb AE, Spielberg SP. Sulfamethoxazole is metabolized to the hydroxylamine in humans. Clin Pharmacol Ther 1992; 51: 522–6.

46 Cribb AE, Miller M, Leeder JS, Hill J, Spielberg SP. Reactions of the nitroso and hydroxylamine metabolites of sulfamethoxazole with reduced glutathione. Implications for idiosyncratic toxicity. Drug Metab Dispos 1991; 19: 900–6.

47 Callan HE, Jenkins RE, Maggs JL, Lavergne SN, Clarke SE, Naisbitt DJ, Park BK. Multiple adduction reactions of nitroso sulfamethoxazole with cysteinyl residues of peptides and proteins: implications for hapten formation. Chem Res Toxicol 2009; 22: 937–48.

48 Stevenson DD, Szczeklik A. Clinical and pathologic perspectives on aspirin sensitivity and asthma. J Allergy Clin Immunol 2006; 118: 773–86.

49 Chaudhry T, Hissaria P, Wiese M, Heddle R, Kette F, Smith WB. Oral drug challenges in non-steroidal anti-inflammatory drug-induced urticaria, angioedema and anaphylaxis. Intern Med J 2012; 42: 665–71.

50 Baldassarre S, Schandene L, Choufani G, Michils A. Asthma attacks induced by low doses of celecoxib, aspirin, and acetaminophen. J Allergy Clin Immunol 2006; 117: 215–7.

51 Simon RA. Adverse respiratory reactions to aspirin and nonsteroidal anti-inflammatory drugs. Curr Allergy Asthma Rep 2004; 4: 17–24.

52 Himly M, Jahn-Schmid B, Pittertschatscher K, Bohle B, Grubmayr K, Ferreira F, Ebner H, Ebner C. IgE-mediated immediate-type hypersensitivity to the pyrazolone drug propyphenazone. J Allergy Clin Immunol 2003; 111: 882–8.

53 Zhu D, Becker WM, Schulz KH, Schubeler K, Schlaak M. The presence of specific IgE to salicyloyl and O-methylsalicyloyl in aspirin-sensitive patients. Asian Pac J Allergy Immunol 1992; 10: 25–32.

54 Riemer AB, Gruber S, Pali-Scholl I, Kinaciyan T, Untersmayr E, Jensen-Jarolim E. Suppression of gastric acid increases the risk of developing immunoglobulin E-mediated drug hypersensitivity: human diclofenac sensitization and a murine sensitization model. Clin Exp Allergy 2010; 40: 486–93.

55 Harrer A, Lang R, Grims R, Braitsch M, Hawranek T, Aberer W, Vogel L, Schmid W, Ferreira F, Himly M. Diclofenac hypersensitivity: antibody responses to the parent drug and relevant metabolites. PLoS ONE 2010; 5: e13707.

56 Chen Q, Doss GA, Tung EC, Liu W, Tang YS, Braun MP, Didolkar V, Strauss JR, Wang RW, Stearns RA, Evans DC, Baillie TA, Tang W. Evidence for the bioactivation of zomepirac and tolmetin by an oxidative pathway: identification of glutathione adducts *in vitro* in human liver microsomes and *in vivo* in rats. Drug Metab Dispos 2006; 34: 145–51.

57 King C, Tang W, Ngui J, Tephly T, Braun M. Characterization of rat and human UDP-glucuronosyltransferases responsible for the *in vitro* glucuronidation of diclofenac. Toxicol Sci 2001; 61: 49–53.

58 Kretz-Rommel A, Boelsterli UA. Diclofenac covalent protein binding is dependent on acyl glucuronide formation and is inversely related to P450-mediated acute cell injury in cultured rat hepatocytes. Toxicol Appl Pharmacol 1993; 120: 155–61.

59 Bolze S, Bromet N, Gay-Feutry C, Massiere F, Boulieu R, Hulot T. Development of an in vitro screening model for the biosynthesis of acyl glucuronide metabolites and the assessment of their reactivity toward human serum albumin. Drug Metab Dispos 2002; 30: 404–13.

60 Chung CH, Mirakhur B, Chan E, Le Q, Berlin J, Morse M, Murphy BA, Satinover SM, Hosen J, Mauro D, Slebos RJ, Zhou Q, Gold D, Hatley T, Hicklin DJ, Platts-Mills TA. Cetuximab-induced anaphylaxis and IgE specific for galactose-alpha-1,3-galactose. N Engl J Med 2008; 358: 1109–17.

61 Nel L, Eren E. Peri-operative anaphylaxis. Br J Clin Pharmacol 2011; 71: 647–58.

62 Mertes PM, Tajima K, Regnier-Kimmoun MA, Lambert M, Iohom G, Gueant-Rodriguez RM. Perioperative anaphylaxis. Med Clin North Am 2010; 94: 761–89.

63 Florvaag E, Johansson SGO, Irgens A, de Pater GH. IgE-sensitization to the cough suppressant pholcodine and the effects of its withdrawal from the Norwegian market. Allergy 2011; 66: 955–60.

64 Florvaag E, Johansson SGO. The pholcodine story. Immunol Allergy Clin 2009; 29: 419–27.

65 Jenkins RE, Yaseen FS, Monshi MM, Whitaker P, Meng X, Farrell J, Hamlett J, Sanderson JP, El-Ghaiesh S, Peckham D, Pirmohamed M, Park BK, Naisbitt DJ. Beta-lactam antibiotics form distinct haptenic structures on albumin and activate drug-specific T-lymphocyte responses in multiallergic patients with cystic fibrosis. Chem Res Toxicol 2013; 26: 963–75.

66 Parmar JS, Nasser S. Antibiotic allergy in cystic fibrosis. Thorax 2005; 60: 517–20.

67 Coulter EM, Farrell J, Mathews KL, Maggs JL, Pease CK, Lockley DJ, Basketter DA, Park BK, Naisbitt DJ. Activation of human dendritic cells by p-phenylenediamine. J Pharmacol Exp Ther 2007; 320: 885–92.

68 Adam J, Pichler WJ, Yerly D. Delayed drug hypersensitivity: models of T-cell stimulation. Br J Clin Pharmacol 2011; 71: 701–7.

69 Adam J, Eriksson KK, Schnyder B, Fontana S, Pichler WJ, Yerly D. Avidity determines T-cell reactivity in abacavir hypersensitivity. Eur J Immunol 2012; 42: 1706–16.

70 Chessman D, Kostenko L, Lethborg T, Purcell AW, Williamson NA, Chen Z, Kjer-Nielsen L, Mifsud NA, Tait BD, Holdsworth R, Almeida CA, Nolan D, Macdonald WA, Archbold JK, Kellerher AD, Marriott D, Mallal S, Bharadwaj M, Rossjohn J, McCluskey J. Human leukocyte antigen class I-restricted activation of CD8(+) T cells provides the immunogenetic basis of a systemic drug hypersensitivity. Immunity 2008; 28: 822–32.

71 Ostrov DA, Grant BJ, Pompeu YA, Sidney J, Harndahl M, Southwood S, Oseroff C, Lu S, Jakoncic J, de Oliveira CA, Yang L, Mei H, Shi L, Shabanowitz J, English AM, Wriston A, Lucas A, Phillips E, Mallal S, Grey HM, Sette A, Hunt DF, Buus S, Peters B. Drug hypersensitivity caused by alteration of the MHC-presented self-peptide repertoire. Proc Natl Acad Sci U S A 2012; 109: 9959–64.

72 Illing PT, Vivian JP, Dudek NL, Kostenko L, Chen Z, Bharadwaj M, Miles JJ, Kjer-Nielsen L, Gras S, Williamson NA, Burrows SR, Purcell AW, Rossjohn J, McCluskey J. Immune self-reactivity triggered by drug-modified HLA-peptide repertoire. Nature 2012; 486: 554–8.

73 Norcross MA, Luo S, Lu L, Boyne MT, Gomarteli M, Rennels AD, Woodcock J, Margulies DH, McMurtrey C, Vernon S, Hildebrand WH, Buchli R. Abacavir induces loading of novel self-peptides into HLA-B*57: 01: an autoimmune model for HLA-associated drug hypersensitivity. AIDS 2012; 26: F21–9.

74 Wei CY, Chung WH, Huang HW, Chen YT, Hung SI. Direct interaction between HLA-B and carbamazepine activates T cells in patients with Stevens-Johnson syndrome. J Allergy Clin Immunol 2012; 129: 1562–9.

75 Sutherland SK. Serum reactions. An analysis of commercial antivenoms and the possible role of anticomplementary activity in de-novo reactions to antivenoms and antitoxins. Med J Aust 1977; 1: 613–5.

76 Warrell DA. Snake bite. Lancet 2010; 375: 77–88.

77 Gawarammana I, Keyler D. Dealing with adverse reactions to snake antivenom. Ceylon Med J 2011; 56: 87–90.

78 Stone SF, Isbister GK, Shahmy S, Mohamed F, Abeysinghe C, Karunathilake H, Ariaratnam A, Jacoby-Alner TE, Cotterell CL, Brown SG. Immune response to snake envenoming and treatment with antivenom; complement activation, cytokine production and mast cell degranulation. PLoS Negl Trop Dis 2013; 7: e2326.

79 Gueant JL, Gueant-Rodriguez RM, Gastin IA, Cornejo-Garcia JA, Viola M, Barbaud A, Mertes PM, Blanca M, Romano A. Pharmacogenetic determinants of immediate and delayed reactions of drug hypersensitivity. Curr Pharm Des 2008; 14: 2770–7.

80 Yang J, Qiao HL, Zhang YW, Jia LJ, Tian X, Gao N. HLA-DRB genotype and specific IgE responses in patients with allergies to penicillins. Chin Med J 2006; 119: 458–66.

81 Woodard-Grice AV, Lucisano AC, Byrd JB, Stone ER, Simmons WH, Brown NJ. Sex-dependent and race-dependent association of XPNPEP2 C-2399A polymorphism with angiotensin-converting enzyme inhibitor-associated angioedema. Pharmacogenet Genomics 2010; 20: 532–6.

82 Mahmoudpour SH, Leusink M, van der Putten L, Terreehorst I, Asselbergs FW, de Boer A, Maitland-van der Zee AH. Pharmacogenetics of ACE inhibitor-induced angioedema and cough: a systematic review and meta-analysis. Pharmacogenomics 2013; 14: 249–60.

83 Kim SH, Hur GY, Choi JH, Park HS. Pharmacogenetics of aspirin-intolerant asthma. Pharmacogenomics 2008; 9: 85–91.

84 Park BL, Kim TH, Kim JH, Bae JS, Pasaje CF, Cheong HS, Kim LH, Park JS, Lee HS, Kim MS, Choi IS, Choi BW, Kim MK, Shin S, Shin HD, Park CS. Genome-wide association study of aspirin-exacerbated respiratory disease in a Korean population. Hum Genet 2013; 132: 313–21.

85 Wei CY, Chung WH, Huang HW, Chen YT, Hung SI. Direct interaction between HLA-B and carbamazepine activates T cells in patients with Stevens-Johnson syndrome. J Allergy Clin Immunol 2012; 129: 1562–9.

86 Harper NJN, Dixon T, Dugue P, Edgar DM, Fay A, Gooi HC, Herriot R, Hopkins P, Hunter JM, Mirakian R, Pumphrey RS, Seneviratne SL, Walls AF, Williams P, Wildsmith JA, Wood P, Nasser AS, Powell RK, Mirakhur R, Soar J, Working Party of the Association of Anaesthetists of Great Britain and Ireland. Suspected anaphylactic reactions associated with anaesthesia. Anaesthesia 2009; 64: 199–211.

87 Ewan PW, Dugue P, Mirakian R, Dixon TA, Harper JN, Nasser SM. BSACI guidelines for the investigation of suspected anaphylaxis during general anaesthesia. Clin Exp Allergy 2010; 40: 15–31.

88 Mertes PM, Malinovsky JM, Jouffroy L, Aberer W, Terreehorst I, Brockow K, Demoly P. Reducing the risk of anaphylaxis during anesthesia: 2011 updated guidelines for clinical practice. J Invest Allerg Clin 2011; 21: 442–53.

89 Blanca M, Romano A, Torres MJ, Fernandez J, Mayorga C, Rodriguez J, Demoly P, Bousquet PJ, Merk HF, Sanz ML, Ott H, Atanasković-Marković M. Update on the evaluation of hypersensitivity reactions to betalactams. Allergy 2009; 64: 183–93.

90 Naranjo CA, Busto U, Sellers EM, Sandor P, Ruiz I, Roberts EA, Janecek E, Domecq C, Greenblatt DJ. A method for estimating the probability of adverse drug reactions. Clin Pharmacol Ther 1981; 30: 239–45.

91 Sastre J, Manso L, Sanchez-Garcia S, Fernandez-Nieto M. Medical and economic impact of misdiagnosis of drug hypersensitivity in hospitalized patients. J Allergy Clin Immunol 2012; 129: 566–7.

92 Wong BB, Keith PK, Waserman S. Clinical history as a predictor of penicillin skin test outcome. Ann Allergy Asthma Immunol 2006; 97: 169–74.

93 Stember RH. Prevalence of skin test reactivity in patients with convincing, vague, and unacceptable histories of penicillin allergy. Allergy Asthma Proc 2005; 26: 59–64.

94 Raja AS, Lindsell CJ, Bernstein JA, Codispoti CD, Moellman JJ. The use of penicillin skin testing to assess the prevalence of penicillin allergy in an emergency department setting. Ann Emerg Med 2009; 54: 72–7.

95 Messaad D, Sahla H, Benahmed S, Godard P, Bousquet J, Demoly P. Drug provocation tests in patients with a history suggesting an immediate drug hypersensitivity reaction. Ann Intern Med 2004; 140: 1001–6.

96 Fukushima K, Nakatsubo M, Noda M, Uenami T, Hayama Y, Tsuruta N, Oniki S, Saito Y, Niju T, Ikeda T. Anaphylaxis due to intravenous levofloxacin with tolerance to garenoxacin. Internal Med 2012; 51: 1769–72.

97 Bernstein IL, Li JT, Bernstein DI, Hamilton R, Spector SL, Tan R, Sicherer S, Golden DB, Khan DA, Nicklas RA, Portnoy JM, Blessing-Moore J, Cox L, Lang DM, Oppenheimer J, Randolph CC, Schuller DE, Tilles SA, Wallace DV, Levetin E, Weber R. Allergy diagnostic testing: an updated practice parameter. Ann Allergy Asthma Immunol 2008; 100 (3 Suppl. 3): S1–148.

98 Sogn DD, Evans R, 3rd, Shepherd GM, Casale TB, Condemi J, Greenberger PA, Kohler PF, Saxon A, Summers RJ, VanArsdel PP. Results of the National Institute of Allergy and Infectious Diseases Collaborative Clinical Trial to test the predictive value of skin testing with major and minor penicillin derivatives in hospitalized adults. Arch Intern Med 1992; 152: 1025–32.

99 Barbaud A, Trechot P, Reichert-Penetrat S, Commun N, Schmutz JL. Relevance of skin tests with drugs in investigating cutaneous adverse drug reactions. Contact Dermat 2001; 45: 265–8.

100 Empedrad R, Darter AL, Earl HS, Gruchalla RS. Nonirritating intradermal skin test concentrations for commonly prescribed antibiotics. J Allergy Clin Immunol 2003; 112: 629–30.

101 Schafer JA, Mateo N, Parlier GL, Rotschafer JC. Penicillin allergy skin testing: what do we do now? Pharmacotherapy 2007; 27: 542–5.

102 McGowan EC, Saini S. Update on the performance and application of basophil activation tests. Curr Allergy Asthma Rep 2013; 13: 101–9.

103 Leysen J, Sabato V, Verweij MM, De Knop KJ, Bridts CH, De Clerck LS, Ebo DG. The basophil activation test in the diagnosis of immediate drug hypersensitivity. Expert Rev Clin Immunol 2011; 7: 349–55.

104 Sanz ML, Gamboa PM, Mayorga C. Basophil activation tests in the evaluation of immediate drug hypersensitivity. Curr Opin Allergy Clin Immunol 2009; 9: 298–304.

105 De Weck AL, Sanz ML, Gamboa PM, Aberer W, Sturm G, Bilo MB, Montroni M, Blanca M, Torres MJ, Mayorga L, Campi P, Manfredi M, Drouet M, Sainte-Laudy J, Romano A, Merk H, Weber JM, Jermann TM. Diagnosis of immediate-type beta-lactam allergy *in vitro* by flow-cytometric basophil activation test and sulfidoleukotriene production: a multicenter study. J Invest Allerg Clin 2009; 19: 91–109.

106 Abuaf N, Rostane H, Rajoely B, Gaouar H, Autegarden JE, Leynadier F, Girot R. Comparison of two basophil activation markers CD63 and CD203c in the diagnosis of amoxicillin allergy. Clin Exp Allergy 2008; 38: 921–8.

107 Aranda A, Mayorga C, Ariza A, Dona I, Rosado A, Blanca-Lopez N, Andreu I, Torres MJ. *In vitro* evaluation of IgE-mediated hypersensitivity reactions to quinolones. Allergy 2011; 66: 247–54.

108 Leysen J, Bridts CH, De Clerck LS, Vercauteren M, Lambert J, Weyler JJ, Stevens WJ, Ebo DG. Allergy to rocuronium: from clinical suspicion to correct diagnosis. Allergy 2011; 66: 1014–9.

109 Gamboa PM, Sanz ML, Caballero MR, Antepara I, Urrutia I, Jauregui I, González G, Diéguez I, De Weck AL. Use of CD63 expression as a marker of *in vitro* basophil activation and leukotriene determination in metamizol allergic patients. Allergy 2003; 58: 312–7.

110 Gomez E, Blanca-Lopez N, Torres MJ, Requena G, Rondon C, Canto G, Blanca M, Mayorga C. Immunoglobulin E-mediated immediate allergic reactions to dipyrone: value of basophil activation test in the identification of patients. Clin Exp Allergy 2009; 39: 1217–24.

111 Bousquet PJ, Pipet A, Bousquet-Rouanet L, Demoly P. Oral challenges are needed in the diagnosis of beta-lactam hypersensitivity. Clin Exp Allergy 2008; 38: 185–90.

112 Scherer K, Brockow K, Aberer W, Gooi JH, Demoly P, Romano A, Schnyder B, Whitaker P, Cernadas JS, Bircher AJ. Desensitization in delayed drug hypersensitivity reactions – an EAACI position paper of the drug allergy interest group. Allergy 2013; 68: 844–52.

113 Brennan PJ, Rodriguez Bouza T, Hsu FI, Sloane DE, Castells MC. Hypersensitivity reactions to mAbs: 105 desensitizations in 23 patients, from evaluation to treatment. J Allergy Clin Immunol 2009; 124: 1259–66.

114 Castells MC, Tennant NM, Sloane DE, Hsu FI, Barrett NA, Hong DI, Laidlaw TM, Legere HJ, Nallamshetty SN, Palis RI, Rao JJ, Berlin ST, Campos SM, Matulonis UA. Hypersensitivity reactions to chemotherapy: outcomes and safety of rapid desensitization in 413 cases. J Allergy Clin Immunol 2008; 122: 574–80.

115 Cernadas JR, Brockow K, Romano A, Aberer W, Torres MJ, Bircher A, Campi P, Sanz ML, Castells M, Demoly P, Pichler WJ. General considerations on rapid desensitization for drug hypersensitivity – a consensus statement. Allergy 2010; 65: 1357–66.

116 Drug allergy: an updated practice parameter. Ann Allergy Asthma Immunol 2010; 105: 259–73.

117 Cox L, Esch RE, Corbett M, Hankin C, Nelson M, Plunkett G. Allergen immunotherapy practice in the United States: guidelines, measures, and outcomes. Ann Allergy Asthma Immunol 2011; 107: 289–99; quiz 300.

118 Wendel GD, Stark BJ, Jamison RB, Molina RD, Sullivan TJ. Penicillin allergy and desensitization in serious infections during pregnancy. N Engl J Med 1985; 312: 1229–32.

119 Yusin JS, Klaustermeyer W, Simmons CW, Baum M. Desensitization in patients with beta-lactam drug allergy. Allergol Immunopathol (Madr) 2013; 41: 298–303.

120 Stark BJ, Earl HS, Gross GN, Lumry WR, Goodman EL, Sullivan TJ. Acute and chronic desensitization of penicillin-allergic patients using oral penicillin. J Allergy Clin Immunol 1987; 79: 523–32.

121 Turvey SE, Cronin B, Arnold AD, Dioun AF. Antibiotic desensitization for the allergic patient: 5 years of experience and practice. Ann Allergy Asthma Immunol 2004; 92: 426–32.

122 Sancho-Serra Mdel C, Simarro M, Castells M. Rapid IgE desensitization is antigen specific and impairs early and late mast cell responses targeting FcepsilonRI internalization. Eur J Immunol 2011; 41: 1004–13.