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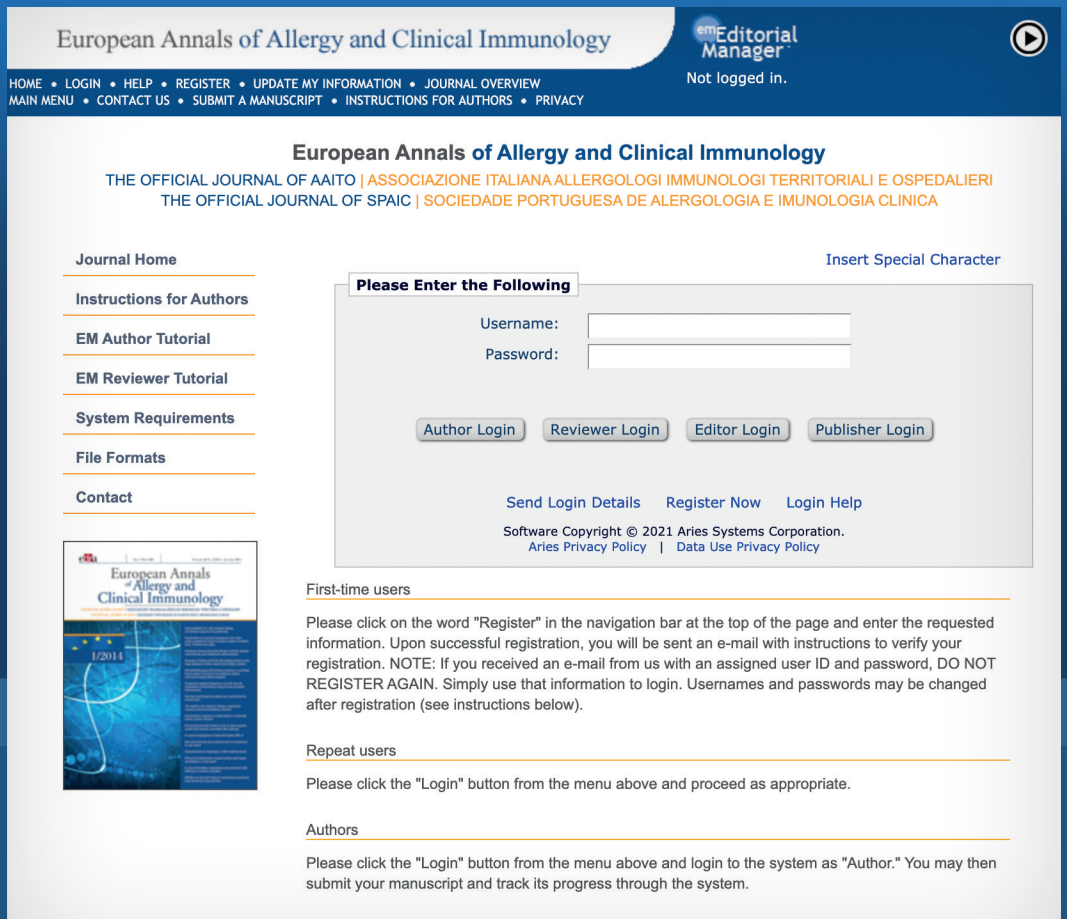
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EDRA SpA

Via G. Spadolini, 7
20141 Milano - Italy
Tel. 0039 (0)2-88184.1
Fax 0039 (0)2-88184.301
www.edizioniedra.it

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Table of Contents

Review

Current update on anaphylaxis: anaphylaxis management in recent guidelines 51
Darío Antolín-Amérigo, Carmen Vidal-Albareda, David González de Olano,
Belén de la Hoz-Caballer

Original articles

Sensitization to genuine markers of timothy grass pollen (*Phleum pratense*) in the North-Central region of Portugal 65
Inês Nunes, Graça Loureiro, Beatriz Tavares, Ana Todo-Bom, Rosário Cunha

The clinical and laboratory findings of infants with atopic dermatitis during diagnosis and follow-up 71
Anil Chousein, Handan Duman Senol, Emine Ece Özdoğru, Sanem Eren Akarcan,
Tuba Tuncel

Der p 23 sensitization in patients with house dust mite respiratory allergy 79
Laura Romero-Sánchez, Andrea Otero, María González-Rivas, Santiago Lojo,
Arturo González-Quintela, Carmen Vidal

Case report

Acetylsalicylic acid desensitization in an allergic pregnant woman post-vascular scaffolds implantation 86
Federica Rivolta, Alessandra Chiei Gallo, Andrea Sangalli, Valerio Pravettoni

Letters to the Editor

Concomitant evaluation of D-dimer and C-reactive protein in chronic spontaneous urticaria may show divergent values . 89
Maria Relvas, Joana Silva, Ana Luísa Matos, Francisca Alves, Margarida Gonçalo

Bronchospasm and iodinated contrast media (ICM): an accurate evaluation is mandatory. 93
Gennaro Liccardi, Matteo Martini, Maria Beatrice Bilò, Manlio Milanese,
Maria Vittoria Liccardi, Gloria Pane, Raffaele Zeccolini, Paola Rogliani

REVIEW

Eur Ann Allergy Clin Immunol Vol 56, N.2, 51-64, 2024

Darío Antolín-Amérigo, Carmen Vidal-Albareda, David González de Olano, Belén de la Hoz-Caballer

Current update on anaphylaxis: anaphylaxis management in recent guidelines

Allergy Service, Hospital Universitario Ramón y Cajal, Instituto Ramón y Cajal de Investigación Sanitaria (IRYCIS), Madrid, Spain

Key words

Anaphylaxis; hypersensitivity reaction; adrenaline; epinephrine; Immunoglobulin E.

Corresponding author
Darío Antolín-Amérigo
Allergy Service
Hospital Universitario Ramón y Cajal, Instituto Ramón y Cajal de Investigación Sanitaria (IRYCIS)
Carretera de Colmenar, Km 9,100
28034 Madrid, Spain
ORCID: 0000-0001-5699-4022
E-mail: dario.antolin2@gmail.com

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Summary

Anaphylaxis is a potentially fatal hypersensitivity reaction but frequently underrecognized. Although its incidence rates vary according to geographical location, it seems clear that there has been a general increase in recent years, either because of greater recognition of this entity or because it is progressing proportionally to the presence of allergic diseases in the world. The development of anaphylaxis management guidelines adapted to local or regional needs seems of utmost importance. Furthermore, it is necessary to assess their implementation and their positive effect regarding diagnosing and treating anaphylaxis. In this review we explore the currently existing definitions of anaphylaxis and its epidemiology, the potential triggers of anaphylaxis and guideline recommendations in terms of diagnosis and management, proposing a novel anaphylaxis calculator and reviewing the current scoring methods for anaphylactic episodes.

Impact statement

Currently existing definitions, scoring methods, potential triggers of anaphylaxis and its epidemiology, and guideline recommendations are reviewed, proposing a novel anaphylaxis calculator.

Introduction

The awareness of anaphylaxis as a life-threatening medical condition and its incidence have been increasing among different specialties, and so is the growth of evidence in the field. The reported increases probably reflect a true increase in the prevalence of allergic disease, but they are also confounded by the cumulative incidence of anaphylaxis, better awareness and recognition of anaphylaxis, and due to changes in anaphylaxis coding.

Anaphylaxis is recognized as a severe, life-threatening systemic hypersensitivity reaction, characterized by rapid onset and the potential to endanger life through respiratory or circulatory compromise.

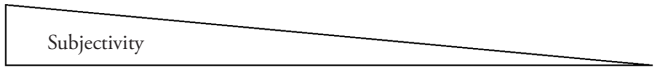
In our article we aimed to cover not only what it is included in the scientific Societies guidelines, but also other means of scoring the severity of the anaphylactic episodes which is in our view not sufficiently gathered in the literature.

Definitions

Anaphylaxis is an underrecognized acute syndrome comprising a life-threatening generalized or systemic hypersensitivity reaction characterized by a severe acute onset of symptoms, involv-

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Figure 1 *- Severity scoring.*



Small expert groups	Expert consensus (*e.g.* Delphi)	Data-informed	Data-driven
Most existing scoring systems were developed by a limited number of experts, and fit into this category	Dribin *et al.*	This approach uses data to inform otherwise subjective decisions by experts as to what symptoms constitute what level of severity	This approach uses raw symptom data and mathematical modelling to derive a score independent of expert input (*e.g.* nFASS score)

**Modified from Shaker et al. (72); subjectivity of studies regarding scoring anaphylaxis.*

ing different organ systems, generally involving airway, breathing or circulatory problems and is usually, although not always, associated with skin and mucosal changes, requiring immediate medical intervention (**figure 1**) (1-4).

The mechanism of anaphylaxis may be either: 1) immunologic, involving Immunoglubulin E (IgE), IgG or immune complexes; or 2) non-immunological (5).

The presenting symptoms can be very varied. However, in over 90% of cases, there are associated skin and mucosal changes. Furthermore, it usually (> 50% of cases) involves the airway (pharyngeal or laryngeal oedema) and/or breathing (bronchospasm with tachypnoea) and/or circulation (hypotension and/or tachycardia).

Epidemiology

None of the actual Academy guidelines, namely, EAACI, WAO, AAAAI, NICE, or the Australian Society of Allergology and Clinical Immunology, has a major focus in this area, but all include important information about epidemiology, supported by relevant references. Therefore, focusing on anaphylaxis remains as an unmet need, in terms of trustable data. The potential challenges could be the diverse and sometimes overlapping nomenclature and criteria for diagnosis of anaphylaxis. Hitherto, prevalence estimates of anaphylaxis vary widely, and many studies, particularly in developed countries, suggest that the prevalence is increasing.

There are differences in prevalence and causes throughout the world probably due to distinct environmental factors which might act differently to affect risk of different types of allergies according to genetic predisposition in diverse areas of the globe and that would account for the rise in the prevalence more rapidly than expected (**table I**) (1-13).

Data from the European anaphylaxis registry revealed that over one-quarter of cases (25%) occurred in patients under 18 years of age (14).

Although the fatality rate attributable to anaphylaxis remains low (15), the frequency of hospitalization for food and drug-induced anaphylaxis has been increasing in recent years (7, 16, 17).

The main causes of fatal anaphylaxis are drugs (29%-58.5%), insect stings (3.3%-54%) and food (2%-6.7%) (**table II**) (9, 18-25).

An important aspect are biphasic anaphylactic reactions which account for up to 20% but other studies report a prevalence from 1% to 7% among patients with anaphylaxis (26, 27). Existing anaphylaxis guidelines therefore recommend continuous follow-up for several hours after resolution of the initial reaction after administering intramuscular injection of adrenaline as first-line anaphylaxis treatment and glucocorticoids, histamine-1 receptor blockers, and beta-2-adrenergic receptor stimulants as second-line treatments (27). In a meta-analysis of retrospective-observational studies performed in Japan of the 31,570 eligible patients, 28,145 (89.2%) were treated with glucocorticoids on the day of admission. The overall percentage of biphasic reactions within 7 days of admission was 11.2%, and the authors stated that there were no significant differences in rates of biphasic reactions (10.7% in the glucocorticoids group *vs* 10.5% in the control group; odds ratio 1.03; 95% confidence interval 0.85-1.24; p = 0.77) between patients with anaphylaxis

Table I *- Worldwide incidence, prevalence and mortality of anaphylaxis.*

	European (globally)	**U.K.**	**U.S.A.**	**WAO**	**Children**	**AAAAI/ ACAAI**	**Spain**
Incidence	1.5-7.9/100,000 person-years	1-7 cases/100,000 person-years		50-112/100,000 persons-years	1-761/100,000 person-years	42/100,000 persons-years	
Prevalence	0.3%		1.6%	0.3-5.1%		1.6-5.1%	
Mortality		0.47-0.69 per 10^6 persons-years	63-99 deaths/ year (> 75% in hospitalized patients)	Drugs: 0.005-0.51 per 10^6 persons-years Food: 0.09-0.13			0.002-2.51 deaths/10^6 persons-years

Table II *- Principal triggers of anaphylaxis*.*

Trigger	Frequency in Spain	Frequency in Europe	Frequency in USA	Frequency Worldwide
Drugs/medication	46.7-62% NSAIDs, beta-lactams, iodinated contrast media	Beta-lactams and NSAIDNSAID	29%-58.5% Antibiotics, NSAIDs, immunomodulators, and biological agents	Geographical variations
Foods	22.6-24.1% Adults: fruits, nuts, shellfish and fish Children: egg, milk, nuts, fish and shellfish	Central: peanut, tree nuts, seeds like sesame, wheat, and shellfish Southern: LTP Children: peanut, hazelnut, milk, and egg Adults: wheat, celery, and shellfish; fruits such as peaches are also typical causes of food-induced anaphylaxis in adults in some European countries such as Spain and Italy	2%-6.7% Peanut and tree nuts are dominating elicitors of food-induced anaphylaxis in adults in North America and Australia	Vary locally. Food-induced anaphylaxis in children toto hen's egg (in infants and pre-school children), cow's milk, wheat, and peanut. Adults: nuts (U.S.A.), shellfish (Asia), buckwheat (Korea)
Insect stings	8.6-13.9%	Wasp: Central Europe	3.3%-54%	Bee: South Korea Wasp: Central Europe Snake Antivenom: Australia
Physical factors	3.4-4%			
Others (including latex)	7.26%			
Idiopathic	3.4-5%			6.5-35%

There are regional differences in terms of culprit agents related to anaphylaxis; modified from Tanno *et al.* (3) and Gold *et al.* (49).

treated with and without glucocorticoids on the day of admission and the authors concluded that they would not support the use of glucocorticoids to prevent biphasic reactions in hospitalized patients with severe anaphylaxis requiring adrenaline.

Therefore, the prevalence of the various causes of anaphylaxis is age-dependent and varies between different geographical areas (**tables I** and **II**). In general, food, drugs, and Hymenoptera venom are the most frequent elicitor groups worldwide (10, 24-41).

Drug-induced anaphylaxis is typically caused by antibiotics and nonsteroidal anti-inflammatory drugs (NSAIDs) with age and geographical variations (42-44). In terms of quality standards, NICE guidelines are a unique document as it proposes some criteria which are expected to contribute to improvements in the following outcomes (45):

- Incidence of anaphylactic episodes.
- Admission rate due to anaphylactic episodes.
- Mortality from anaphylactic episodes.

NICE quality standards are centered on aspects of health and social care that are commissioned locally and comprise a brief set of prioritized statements designed to drive measurable improvements in the 3 dimensions of quality – safety, experience and effectiveness of care, in terms of anaphylaxis (2, 45-49).

Key triggers include food, drugs, and stinging insects; in up to 20%, the elicitor is not identified. Their relative importance varies with age and geography studied. For ED (Emergency Department) presentations, drugs and foods are the most common elicitors of anaphylaxis, with age-related differences (6, 50).

Foods are the most frequent cause of anaphylaxis in children, with pollen allergy and asthma being important risk factors (4, 6).

Drug- and Hymenoptera venom-triggered anaphylaxis are more common in adults than in children (6) in general, and specifically to plant foods and NSAID. Drugs are the most frequent cause of anaphylaxis in hospitalized patients.

Compared to males, adult females have a higher frequency of anaphylaxis.

Cumulative incidence of anaphylaxis ranges from 26.5% to 54%, with a follow-up time of 1.5 years to 25 years (9). In general terms, long-term management, and several structured

actions could achieve the desired prevention of recurrence [(extracted from references 8 and 34):

- Avoidance and/or allergen immunotherapy and/or desensitization.
- Medical identification alert: *e.g.*, bracelet or wallet card.
- Register in electronic or paper medical record the suspected trigger(s).
- Anaphylaxis education and training.
- Public health measures, *e.g.*, improved food labelling.

Methods

As this is a narrative review, evidence has been retrieved from different sources. A literature search was performed using keywords agreed on by the authors. The search was performed using electronic databases (MEDLINE and PubMed), electronic libraries (Clinical Key, Science Direct, OVID), resources of clinical guidelines (UpToDate, Allergy Societies Society Guidelines, and Dynamed: https://www.dynamed.com/condition/anaphylaxis), and a database of systematic reviews (Cochrane Library). Publications were selected from between January 2015 and December 2022. The selection took into account the keywords: "anaphylaxis" and "guideline". Due to the revolution in terms of vaccines and adverse effects, we have also used the Brighton Collaboration Guideline for scoring anaphylaxis, which is currently used worldwide for drug/vaccination regulatory issues (available at: https://brightoncollaboration.us/anaphylaxis-case-definition-companion-guide/).

In addition, the filters used in PubMed were as follows: guideline, consensus development conference and consensus development conference (NIH), practice guidelines, review, systematic review, and meta-analysis. Guidelines, consensus reports, systematic reviews, and meta-analysis were included; nonsystematic reviews, comments, and other types of articles were excluded. We also included studies examining incidence, prevalence, natural history, clinical manifestations, pathogenesis, diagnosis, and treatment. All articles were reviewed by the authors. Following this review process, 89 publications were finally selected.

Diagnosis management − scoring systems: beyond the guidelines?

Anaphylaxis, as mentioned above, is known as a life-threatening generalized or systemic hypersensitivity reaction involving different organ systems (1, 3, 34, 51).

There is a list of criteria that detail the symptoms or combination of symptoms which meet the clinical characteristics of anaphylaxis. Traditionally, anaphylaxis was labeled when any of the 3 proposed criteria were met (**table IIIA**) (52), although recently the World Allergy Organization (WAO) has proposed a new set of criteria (34, 52–55).

There are 2 recently proposed criteria for diagnosing anaphylaxis:

1. EAACI guidelines 2021 (**table IIIA**).
2. WAO guidelines 2020 (**table IIIB**).

The first one, probably is more specific, including the concept of likely and/or known allergen in its criteria and the WAO guidelines specify the possibility of the absence of typical skin involvement in the symptoms - WAO guidelines 2020 (**table IIIB**).

Classification of severity of anaphylactic episodes

Classification of severity implies a challenge, given that there are not homogeneous definitions of subtypes of anaphylaxis (4). In fact, there are different classifications that attempt to classify anaphylaxis according to its severity (34, 52, 55-59). They are divided into grades, ranging from I to V, based on the degree of vital compromise. Not all of them cover the full range of symptoms that patients may present, and some of them were created specifically for a particular trigger, such as Hymenoptera venom (56) or food (58). This has led to the recent proposal of a classification that attempts to unify the previous ones (59-61) or to the creation of a new way of classifying anaphylaxis proposed by the WAO (34) but none of them have been widely used up to now. Given the trend on immunization, the Brighton Collaboration (BC) was established in 2000 with the aim of developing globally accepted standardized case definitions for adverse events following immunization (AEFI) as well as guidelines for the collection, analysis and presentation of surveillance data (61). It has served to improve the classification of the severity of an anaphylactic episode following immunization. The Brighton Collaboration Cased Definitions (BCCD), which are evidence based, were developed by a group of experts to describe, in simple terms, all signs and symptoms used to constitute the BCCD on anaphylaxis (62). The descriptive terms used in the case definitions could be used as a guide to develop educational and recording material (63). The checklist could be used when sudden or unexpected symptoms or signs occur post-vaccination, and where anaphylaxis is a possibility. In contrast, checklist 2 collects additional details such as demographic information as well as vaccine data. Such checklists are more comprehensive and are better suited to clinical trials or to the analysis of AEFI data at a regional or national level. For example, more extensive checklists could be used as part of an investigation of individual cases. If correctly completed, such checklists will aid in the assignment of a BCCD for anaphylaxis (63, 64).

Once the symptoms and signs that may indicate anaphylaxis have been reported on an AEFI report form and/or through the use of a checklist, a case definition for anaphylaxis can be assigned. It is intended that the task of assigning a case definition is performed at a regional or national level because of the complexity of the case definition. In addition, online tools are being developed by the BC to facilitate this process and these

Table III - (A) *Clinical criteria for diagnosing anaphylaxis (EAACI guidelines 2021) (4);* ***(B)*** *WAO anaphylaxis guidelines 2020 (36).*

A	B
1. Acute onset of an illness (minutes to several hours) with involvement of the skin, mucosal tissue or both (*e.g.*, generalized hives, pruritus or flushing, swollen lips-tongue- uvula and at least one of the following: **a**. respiratory compromise (*e.g.*, dyspnea, wheeze-bronchospasm, stridor, reduced PEF and hypoxemia); **b**. reduced BP or associated symptoms of end-organ dysfunction (*e.g.*, hypotonia [collapse], syncope, incontinence).	1. Acute onset of an illness (minutes to several hours) with simultaneous involvement of the skin, mucosal tissue, or both (*e.g.*, generalized hives, pruritus or flushing, swollen lips–tongue-uvula) and at least one of the following: **a**. respiratory compromise (*e.g.*, dyspnea, wheeze–bronchospasm, stridor, reduced PEF, hypoxemia); **b**. reduced BP or associated symptoms of end-organ dysfunction (*e.g.,* hypotonia (collapse), syncope, incontinence); **c**. severe gastrointestinal symptoms (*e.g.*, severe crampy abdominal pain, repetitive vomiting), especially after exposure to non-food allergens.
2. Two or more of the following that occur rapidly after exposure to a likely allergen for that patient (minutes to several hours): **a**. involvement of the skin–mucosal tissue (*e.g.,* generalized hives, itch-flush, swollen lips-tongue-uvula; **b**. respiratory compromise (*e.g.,* dyspnea, wheeze-bronchospasm, stridor, reduced PEF, hypoxemia); **c**. reduced BP or associated symptoms (*e.g.,* hypotonia [collapse], syncope, incontinence); **d**. persistent gastrointestinal symptoms (*e.g.,* crampy abdominal pain, vomiting).	2. Acute onset of hypotension or bronchospasm or laryngeal involvement after exposure to a known or highly probable allergen for that patient (minutes to several hours), even in the absence of typical skin involvement.
3. Reduced BP after exposure to known allergens for that patient (minutes to several hours): **a**. infants and children: low systolic BP (age specific) or > 30% decrease in systolic BP*; **b**. adult: systolic BP of < 90 mmHg or > 30% decrease from that person's baseline PEF, peak expiratory flow; BP, blood pressure. *Low systolic blood pressure for children is defined as < 70 mmHg: - from 1 month to 1 year, less than (70 mmHg + [2 × age]) - from 1 to 10 years and < 90 mmHg from 11 to 17 years.	

(A) Anaphylaxis is highly likely when any one of the three criteria is fulfilled; **(B)** Anaphylaxis is highly likely when any one of the two criteria is fulfilled.

will be freely accessed at the Brighton Collaboration website: http://www.brightoncollaboration.org/internet/en/index.html.
A potential limitation to collecting exhaustive adverse event reports is that health care providers are unlikely to spontaneously report enough symptoms and signs to allow application of BCCD. A checklist, based on the terms used in the case definition, could be developed and incorporated in an AEFI reporting form to remind vaccine providers to note whether specific features of anaphylaxis were present. Another limitation of the anaphylaxis case definition is the hypothetical inconsistencies in the reporter's use of the terms used to describe possible anaphylaxis. This would be addressed by the development of a glossary to describe those terms used in the anaphylaxis case definition. With these barriers in mind, Stafford *et al*. have created a clinical checklist for the application of the BCCD and a glossary of terms to harmonize the use of reporting terms (65).
As Stafford *et al*. stated recently, having a standardized, internationally agreed on quantitative measure of severity might be useful in facilitating risk communication, both with patients and with industry/regulators. Consistency and translatability in recording results are essential and currently constitute unmet needs in the field of anaphylaxis. However, any severity score must be fit for purpose, be informed by patient and clinician experience, and ideally be data driven to minimize the impact of subjectivity and provide objective validation (**figure 1** modified from Dribin *et al.*) (66).
In the meantime, the severity score proposed by Dribin *et al*. highlights the inconsistencies and limitations of the NIAID criteria for anaphylaxis (66).
Developing our acquaintance of the relationship between the definition of anaphylaxis (and indications for epinephrine treatment) and severity grading of symptoms is essential for further progress in this area. We need to achieve a global consensus on updated anaphylaxis criteria to improve anaphylaxis recognition and thus patient care − it is what patients deserve.

Calculator of anaphylaxis

For the diagnosis of anaphylaxis in an acute context, the 2020 EAACI Task Force on anaphylaxis, has suggested using clinical criteria, including rapid onset of multiple symptoms, measuring serum tryptase half to two hours after the start of the reaction, and baseline tryptase at least 24 hours after complete resolution of symptoms, to support diagnosing anaphylaxis retrospectively. A number of studies have assessed the diagnostic accuracy of

serum tryptase measurements for anaphylaxis, but the evidence is of very low certainty, derived from consecutive case series or case control studies (55, 67, 68).

In terms of practical management, taking the blood sample should not delay treating a patient with adrenaline where necessary. A sample taken later than 2 hours after the reaction may still demonstrate a raised tryptase level.

An event-related transient elevation of the serum tryptase level by at least 20% over the individual baseline plus 2 ng/mL absolute (20%+2) within a 2-4 h window after reaction supports a diagnosis of anaphylaxis (*tryptase peak rising level ≥ (20% × baseline tryptase) + 2 µg/L*) (69, 70).

It is interesting to note that a recent study quantified the actual benefit of measuring tryptase in Emergency Department patients with anaphylaxis, which found that around 2% of the patient cohort who did not meet diagnostic criteria for anaphylaxis would have elevated levels of serum-tryptase, higher than the normal ranges of the local laboratory (> 12 µg/L) (57).

A raised serum tryptase level can be associated with a mast cell disorder or hereditary alpha tryptasemia (71-73), so it is important to compare with a baseline level at least 24 hours after complete resolution of a reaction. Moreover, serum tryptase is not always elevated in anaphylaxis, especially in children and with food triggers in all ages (74).

Apart from tryptase, a number of other MC-derived compounds may serve as suitable parameters to document severe reactions following systemic MC activation. These substances include, among others, histamine and its metabolites, PGD2 and its metabolites, and heparin (75-77).

However, except for heparin, these mediators are less specific for MC compared to tryptase. Methods to determine these mediators are also much less available through widely distributed commercial assays and, moreover, none of these laboratory exams are validated for the diagnosis of anaphylaxis, so far (69).

Management

The evidence base for the acute management of anaphylaxis is weak and is established mainly by means of systematic reviews, due to the absence of randomized controlled trials. However, there seems to be a global consensus that intramuscular (IM) adrenaline is the treatment of choice as the first step in the management of acute anaphylaxis (19, 34, 78).

All in all, there is room for improvement in its management which shall be systematic (79).

Some of the unmet needs with regard to anaphylaxis are acute management and with regards prophylactic measures (3).

There are several factors influencing the successful treatment of an anaphylactic reaction such as specific training in anaphylaxis management along with rapid identification and treatment, which are critical (52, 79).

The basic principles for the treatment of anaphylaxis are common for all age groups, but it may be influenced by the context/setting, personnel, equipment and by the available medication (79, 80).

The necessary materials and medications to treat an anaphylactic episode are (5, 6):

1. Stethoscope, pulse-oxymeter and tensiometer (including equipment for blood pressure and cardiac continuous monitoring), watch or clock.
2. Tourniquets, syringes (with needles:1 mL, 10 mL, 20 mL) and needles IV (gauge 19, 21, 23 and 25) and IM, and catheter (gauges 14, 16, 18, 20 and 22).
3. Aqueous Adrenaline (1 mg/ml or 1/1,000) 0.01 mg/kg to a maximum of 0.5 mg (adult) and 0.3 mg (child).
4. Equipment to deliver oxygen (oxygen tank, valve with flow-meter, and extension tubing).
5. Equipment to deliver IV fluids.
6. Intubation material: ambu bag/valve/mask, self-inflating with reservoir (volume 700-1,000 mL (adult), 100-700 (child) and disposable face masks; oropharyngeal airways: 6 cm, 7 cm, 8 cm, 9 cm, 10 cm; pocket masks and nasal cannula, laryngeal mask airways.
7. Antihistamines IV (chlorpheniramine 10 mg (adult), 2.5-5 mg (child) or diphenhydramine 25-50 mg (adult) (1 mg/kg, maximum 50 mg (child)).
8. Corticosteroids IV (hydrocortisone 200 mg (adult), maximum 100 mg child or methylprednisolone 50-100 mg (adult) 1 mg/kg, maximum 50 mg (child).
9. Vasopressors IV (dopamine, noradrenaline…).
10. Glucagon.
11. Defibrillator.
12. Inhaled Beta-Adrenergics: salbutamol solution 2.5 mg/3 mL or 5 mg/3mL (adult) (2.5 mg/3 mL, child) given by nebulizer and face mask.
13. Other supplies: extension tubing, T connectors, 3-way stopcocks, armboards, written emergency protocol for anaphylaxis treatment, flow chart for recording times and events, gloves.

The emergency treatment of a patient who is suffering an anaphylactic reaction may differ if it occurs outside of a healthcare institution or inside a hospital/ambulatory center. Both the available resources and the accessibility to a hospital/ambulatory center may influence the first approach.

Management outside of a healthcare institution

The first step, regardless of the context and the severity of the anaphylactic reaction, would be the use of adrenaline autoinjector and subsequently an immediate phone calling for emergency support (**table IV**). Any healthcare professional shall be capable of initiating the treatment of a patient who is suffering from an anaphylactic reaction and calling for emergency support.

Table IV - *Indications for prescribing adrenaline autoinjectors, regardless of the context and the severity of the anaphylactic reaction.*

A	B	
Previous (severe or near fatal)* anaphylaxis triggered by foods, medication, aeroallergens, exercise- induced, latex or idiopathic	Arguments for 2 autoinjectors	Arguments for 1 autoinjector
Coexistent unstable or moderate-severe/severe asthma and food allergy*	European Medicines Agency recommends 2 autoinjectors	Only needing to carry one device may improve adherence
Venom allergy in untreated patients and risk of re-exposure	About 10% patients required a 2nd dose of adrenaline due to insufficient response to the 1st dose	Most autoinjectors are not used and have to be replaced after 12-18 months
Underlying systemic mastocytosis in adults with any previous systemic reaction. Children with very severe skin involvement (>50% body surface) and increased basal serum tryptase levels (>20 ng/ml) and with blistering in the first three years of life*	Rarely, injection in the wrong place	Most patients respond to 1 dose and 2nd doses are usually administered by emergency services
Previous mild-moderate reactions to food	Likelihood of delayed medical assistance (remote location or travel)	
Remote from medical help or repeated travel abroad with mild moderate reactions to food, medication, insect, latex or idiopathic*		
Cardiovascular disease		
Oral immunotherapy for food allergy		
Previous requirement of more than 1 adrenaline dose before arriving a healthcare institution*		
If available adrenaline dose is too low for body weight*		

(A) *At least 1 adrenaline autoinjector. Notwithstanding, consider prescribing 2 adrenaline autoinjectors; Modified from (4); **(B)** reasons for prescribing one or two adrenaline autoinjectors (modified from references 4, 6, 79, 87).

Rescue volunteers, paramedics, lifeguards, and nurses shall be trained, updated, and sufficiently skilled to cope with an anaphylactic reaction (81, 82).

Equipment and available medication (in a healthcare institution)

Every healthcare institution shall have a crash cart containing all the necessary equipment and medication to make a complete cardiorespiratory resuscitation and to treat a potential anaphylactic reaction (see paragraph Management). Every health care professional shall be familiar with the equipment and every medication shall be reviewed periodically. Whenever a patient begins to suffer an anaphylactic reaction, it has to be monitored as soon as possible (79).

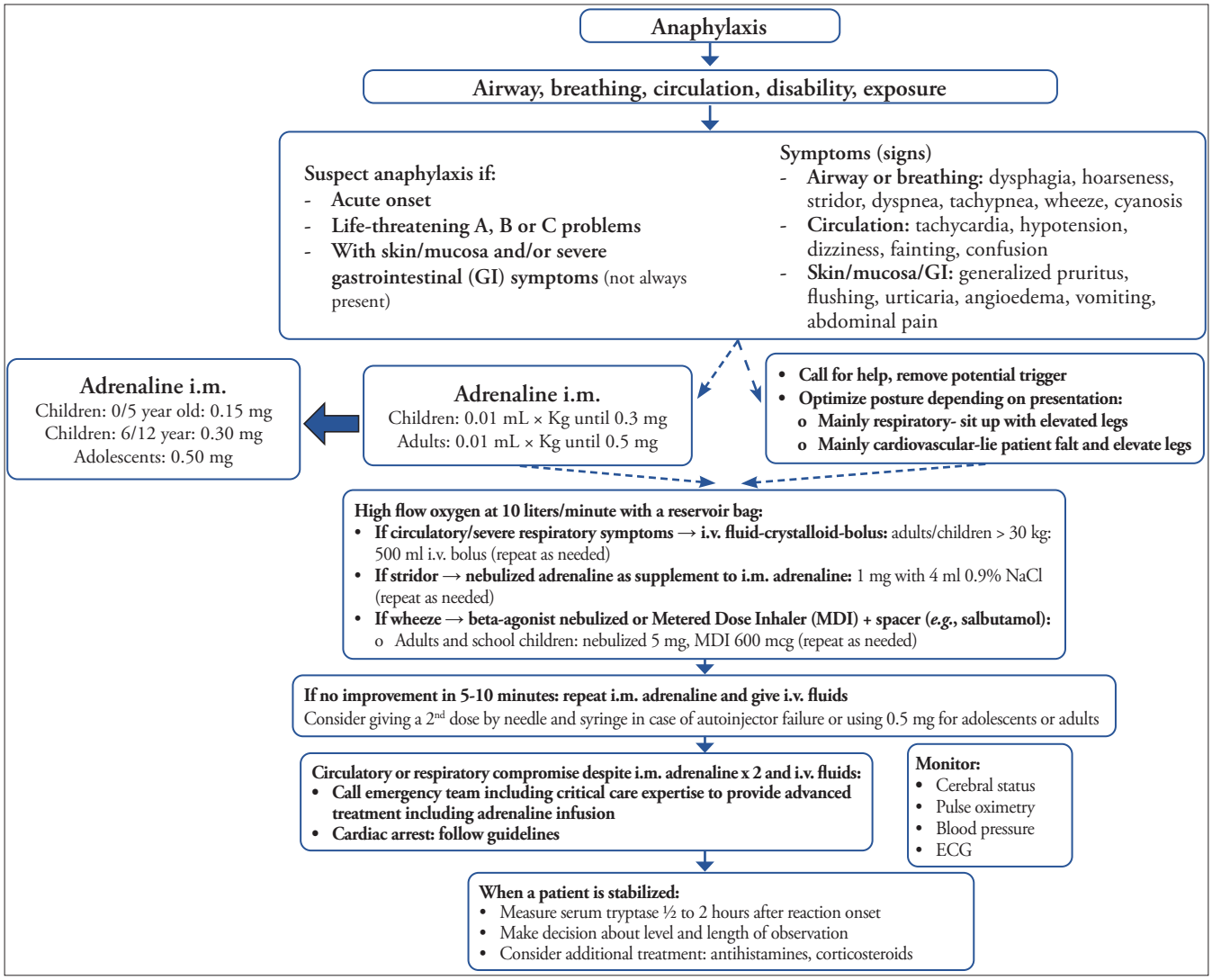
Minimal approach requirements for a potential anaphylactic episode

1. Stratifying patient's severity (**figure 2**).
2. Early emergency support request.
3. Initial treatment based on ABCDE approach.
4. Adrenaline (when indicated).
5. Preferential study and follow-up by an allergist: essential.

Patient position (extensible for inside and outside a healthcare institution)

Whenever a patient is being treated or has suffered from an anaphylactic episode, it should be placed in a comfortable position, laid down and with raised legs to gain venous return. This position shall be avoided when vomiting or any kind of respiratory failure. Postural changes shall be made with caution as they may cause the worsening of a hemodynamic compromise. Unconscious patients shall be placed in a lateral position. Pregnant women shall be placed in the left lateral position to avoid cava vein compression (83). Airways permeability shall be warranted at all times (79). In case of respiratory failure, semi-recumbent position can be considered. Anyhow, the UK Resuscitation Council 2021 Guidelines comprise further details on patient positioning in anaphylaxis (71).

Figure 2 *- Treatment scheme of an anaphylactic episode.*



*Modified from references 4, 79, 84. *Adrenaline preferably shall be adjusted by weight.*

In patients with a history of prior anaphylaxis, acute management consists of two steps:

1. Self-management of the patient using an emergency protocol, in which it is important to emphasize the key role of intramuscular adrenaline, encouraging the possibility of self-administration of adrenaline using auto-injector devices, no matter the severity of the anaphylactic reaction (**table IV**).
2. Additional interventions given by healthcare professionals once medical help has arrived, which must include further adrenaline if symptoms of anaphylaxis ongoing.

It is of note that the most effective and evidence-based treatment for anaphylaxis is adrenaline, based on observational studies, extrapolation from retrospective case reports, and scarce clinical trials (3, 32).

Airway protection

As patients with anaphylaxis can rapidly develop critical airway compromise, it is crucial to protect the airway while other treatments are administered (27).

Intubation should be considered early, as patients can decompensate rapidly, which can make endotracheal intubation more difficult or impossible and necessitate the placement of a surgical airway. If the patient shows intense laryngeal edema, non-invasive positive pressure ventilation or supraglottic airways

should be used with caution, as they may not be effective. Endotracheal intubation with direct or video laryngoscopy should be attempted first by an experienced clinician if the oral edema is judged to be navigable.
Supplemental oxygen should be administered to maintain oxygen saturation > 90% (27).

Medication
Delayed epinephrine administration is associated with biphasic reactions (78). It is a matter of debate whether glucocorticoids would prevent biphasic reactions in patients with anaphylaxis as they inhibit inflammatory responses by suppressing the function of mast cells (78). Recent retrospective cohort studies have failed to show any preventive effects of glucocorticoids on biphasic reactions.
Moreover, antihistamines have no role in the primary management of life-threatening signs and symptoms of anaphylaxis, including upper airway edema and shock. The rationale for their use arises from the effectiveness in other allergic diseases, but there are no specific data that suggest benefit in anaphylaxis (80).

Healthcare institution discharge after anaphylaxis
If the diagnosis of anaphylaxis (or suspicion) has been reached, patients shall remain for 6-8 hours after complete resolution of symptoms and up to 12-24 hours whenever a patient shows refractory symptoms, or in the event of very severe reactions or having a history of biphasic reactions. Other patients, who would also need to stay longer, would be the following: 1) patients with difficult access to a healthcare institution, 2) severe idiopathic anaphylaxis with slow progression; 3) severe asthma or severe respiratory symptoms; 4) reactions in which the allergen could be present if the patient is discharged; 5) severe condition of the patient (cardiocirculatory compromise) (79).
It is mandatory to have a complete medical record indicating all received medications and the evolution of the patient.
Recommendations after healthcare institution discharge because of anaphylaxis:
- Emergency department visit: if symptoms reappear.
- Antihistamines, corticosteroids, and prescription of adrenaline autoinjector(s) (independently of the severity of the anaphylactic reaction) (**table IV**).
- Avoidance of potential culprit agents (foods, medications, insects, *etc*.).
- Continued follow-up plan for general practitioners.
- Personalized plans, including referral to psychological support and patients' organizations (4).
- Allergen immunotherapy and desensitization: allergen immunotherapy (AIT) is potentially a curative therapy. AIT may increase the amount of food that the patient can tolerate, preventing allergic symptoms and reducing the risk of potentially life-threatening allergic reactions (77).

Venom immunotherapy (VIT) is the model of immunotherapy with regards the studies, its efficacy and the knowledge of the physiologic modifications of induction of immune tolerance produced (78). Moreover, it has been shown to improve the quality of life of venom-allergic patients when compared with patients who do not receive immunotherapy but carry adrenaline (79).
Food Allergy-OIT is recommended for persistent Cow's Milk, Hen Egg, or peanut allergy for children from around 4 to 5 years of age on the basis of its ability to increase the threshold for clinical reactions while on OIT (Grade A). At present, there are insufficient data to be able to recommend AIT for other foods and for adults outside clinical trials (77).

Personalized plans
Every patient should receive a written record with instructions such as: avoidance measures and when and how to use the adrenaline autoinjector (**table IV**). All patients shall be referred to an Allergy Department to investigate the potential culprit agent and to minimize present and future risks. New guidelines point out the importance of prescribing personalized plans to achieve the best approach in terms of continued follow-up of patients who have suffered an anaphylactic reaction (4, 6, 52, 79, 85-87).

Education and training for health care professionals (HCP) and patients at risk of anaphylaxis
The EAACI Task Force on anaphylaxis, along with other guidelines, recommends providing structured, comprehensive training to improve the knowledge and use of adrenaline autoinjectors in people at risk of anaphylaxis. This is in addition to basic instructions about autoinjector use (4).
Although it is unclear what types of training and support are most effective, education is essential if patients at risk of anaphylaxis are to successfully recognize and manage future episodes. Diverse approaches are available, including the use of adrenaline, autoinjector training devices, and online approaches. Training HCP using anaphylaxis simulators will improve its management (4).

Other potential educational interventions
Some studies have also found that supporting patients to practice using an adrenaline autoinjector or needle and syringe containing 0.9% saline can reduce anxiety or improve quality of life.
In the case of anaphylaxis during an in-hospital-based food/drug challenge, patients and caregivers may be encouraged to administer their own adrenaline autoinjector to improve their confidence in this procedure. It is also important for allergists to follow a patient's anaphylaxis management plan during a provocation challenge (*e.g.*, giving in adrenaline at the first sign of anaphylaxis) to reinforce this self-management approach (88).

Approaches to prevent anaphylaxis in schools

School policies should reflect anaphylaxis guidelines, but more research is needed to understand how guidelines and legislation in schools are best implemented. Anaphylaxis due to food allergy, occurs in schools more than in any other community location (6).

Therefore, it may be helpful to target secondary schools and community settings with educational support to help raise general awareness, empower adolescents to confident self-manage food allergy, and enable schools to develop protocols to minimize any adverse events if they occur.

Other approaches investigated to improve the management of anaphylaxis included nurses checking whether students were carrying autoinjectors and availability of a 24-hour helpline (89).

Knowledge gaps

In our opinion, there are three main areas of work that we consider should be a priority to improve the management of anaphylaxis as described in the guidelines, which are those related to the diagnosis of the disease, its classification and its management (66, 88-91).

The difficulty in diagnosing anaphylaxis is that there is no pathognomonic set of signs or symptoms, especially when no cutaneous manifestations are present and there is not an immediate history of drug administration or allergen exposure. What is typical is the rapid progression in severity or intensity of symptoms. For this reason, it would be advisable to unify the diagnostic criteria and disseminate them, especially among specialties that handle clinical emergencies.

Although the most important aspect is to recognize the entity and initiate appropriate treatment (88, 89), it is also necessary to use the same tools to classify the severity of a reaction. It would be advisable to find a consensus and try to find a single and suitable classification of anaphylaxis.

It would also be important to improve knowledge and tools for the treatment of anaphylaxis (87). Turner *et al*. undertook a thorough literature search about the pharmacokinetics (PK) of adrenaline autoinjectors (66, 92, 93).

In community settings, adrenaline can be provided for emergency use as an adrenaline autoinjector (AAI) device (**table IV**), although these are not available in many countries (88, 89). Besides, there is a barrier in the use of AAI and is the price. Allergy societies and Patients' Organizations in Spain are joining forces to achieve a symbolic price, which is for many patients high, given the expiration date of the AAI, so as to assure that every patient at risk would carry AAI.

Given the currently available data, it takes at least 5-10 min to achieve early peak plasma concentration for most devices and the authors state that the specific autoinjector device seems to be the most important determinant of pharmacokinetics, with different devices giving rise to different plasma adrenaline profiles. Needle length does not seem to be the most important factor; rather, the force and speed of injection (which varies from one device to another) is likely to be of greater importance. In general, peak plasma adrenaline concentration is lower and time-to-peak concentration is longer with increased skin-to-muscle depth. However, it is difficult to draw conclusions with the current available data, due to a lack of head-to-head comparisons, small numbers of study participants, and the failure to acknowledge the biphasic nature of intramuscular adrenaline absorption for analysis purposes.

Several studies tried to assess the impact of body mass index, using the parameter of skin-to- muscle depth (STMD), assessed by ultrasound. Notwithstanding, the numbers in each subgroup would not allow to detect any small differences between groups. Other studies found significant differences in terms of lower SMTD and faster and greater increase in plasma adrenaline in the normal weight men and *vice versa* (66, 78, 88, 89).

Although PK data are available, the most helpful study comparing three different devices is unpublished. According to all international guidelines, the recommended dose of IM adrenaline in adults is 0.5 mg; thus, a dose of 0.3 mg given by AAI may be inadequate and should be repeated after 5-10 min in the absence of resolution of symptoms (66).

Conclusions

Anaphylaxis is a potentially fatal hypersensitivity reaction. Although its incidence rates vary according to geographical location, it seems clear that there has been a general increase in recent years, either because of greater recognition of this entity or because it is progressing proportionally to the presence of allergic diseases in the world. On many occasions, episodes of anaphylaxis are treated by physicians working in Emergency departments and are not directly evaluated by an allergist. It is necessary to establish common and consensual guidelines to help professionals and patients to know how to recognize anaphylaxis, as well as to be able to classify it, treat it correctly and make future recommendations after the acute episode.

Studies comprising the epidemiology of anaphylaxis, as well as the importance of biphasic reactions seem crucial to exactly define priorities in terms of research and resources.

Preventive measures, including personalized action plans may circumvent the burden of anaphylaxis for patients, relatives, and caregivers.

The development of anaphylaxis management guidelines adapted to local or regional needs seems of utmost importance, but it is necessary to assess their implementation and their positive effect regarding diagnosing and treating anaphylaxis.

There is a clear need for establishing multinational, large databases/registries to collect high-quality epidemiologic risk factor

data and diagnosis or treatment outcomes for improving the management of patients with anaphylaxis.

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Contributions

DA-A: conceptualization, formal analysis, methodology, project administration, writing - original draft, writing - review & editing. CV-A, DGdO, BDlH-C: conceptualization, writing - original draft, writing - review & editing.

Conflict of interests

The authors declare that they have no conflict of interests.

References

1. Simons FER, Ardusso LR, Bilò MB, Cardona V, Ebisawa M, El-Gamal YM, et al. International consensus on (ICON) anaphylaxis. World Allergy Organ J. 2014;7(1):9. doi: 10.1186/1939-4551-7-9.
2. Anaphylaxis: assessment and referral after emergency treatment Clinical guideline. 2011. Available at: www.nice.org.uk/guidance/cg134. Last access date: 06/30/2023.
3. Tanno LK, Touati N, Allichon S, Martin B, Ebisawa M, Ansotegui I, et al. A Proposal from the Montpellier World Health Organization Collaborating Centre for Better Management and Prevention of Anaphylaxis. J Allergy Clin Immunol Pract. 2021;9(2):676-83.e1. doi: 10.1016/j.jaip.2020.09.062.
4. Muraro A, Worm M, Alviani C, Cardona V, DunnGalvin A, Garvey LH, et al. EAACI guideline: Anaphylaxis (2021 update). Allergy. 2022;77(2):357-77. doi: 10.1111/all.15032.
5. Johansson SGO, Bieber T, Dahl R, Friedmann PS, Lanier BQ, Lockey RF, et al. Revised nomenclature for allergy for global use: Report of the Nomenclature Review Committee of the World Allergy Organization, October 2003. J Allergy Clin Immunol. 2004;113(5):832-6. doi: 10.1016/j.jaci.2003.12.591.
6. Worm M, Moneret-Vautrin A, Scherer K, Lang R, Fernandez-Rivas M, Cardona V, et al. First European data from the network of severe allergic reactions (NORA). Allergy. 2014;69(10):1397-404. doi: 10.1111/all.12475.
7. Bilò MB, Corsi A, Martini M, Penza E, Grippo F, Bignardi D. Fatal anaphylaxis in Italy: Analysis of cause-of-death national data, 2004-2016. Allergy. 2020;75(10):2644-52. doi: 10.1111/all.14352.
8. Turner PJ, Gowland MH, Sharma V, Ierodiakonou D, Harper N, Garcez T, et al. Increase in anaphylaxis-related hospitalizations but no increase in fatalities: an analysis of United Kingdom national anaphylaxis data, 1992-2012. J Allergy Clin Immunol. 2015;135(4):956-963.e1. doi: 10.1016/j.jaci.2014.10.021.
9. Garvey LH, Dewachter P, Hepner DL, Mertes PM, Voltolini S, Clarke R, et al. Management of suspected immediate perioperative allergic reactions: an international overview and consensus recommendations. Br J Anaesth. 2019;123(1):e50-64. doi: 10.1016/j.bja.2019.04.044.
10. Hopkins PM, Cooke PJ, Clarke RC, Guttormsen AB, Platt PR, Dewachter P, et al. Consensus clinical scoring for suspected perioperative immediate hypersensitivity reactions. Br J Anaesth. 2019;123(1):e29-37. doi: 10.1016/j.bja.2019.02.029.
11. Liew WK, Williamson E, Tang MLK. Anaphylaxis fatalities and admissions in Australia. J Allergy Clin Immunol. 2009;123(2):434-42. doi: 10.1016/j.jaci.2008.10.049.
12. Helbling A, Hurni T, Mueller UR, Pichler WJ. Incidence of anaphylaxis with circulatory symptoms: a study over a 3-year period comprising 940,000 inhabitants of the Swiss Canton Bern. Clin Exp Allergy. 2004;34(2):285-90. doi: 10.1111/j.1365-2222.2004.01882.x.
13. Tanno LK, Bierrenbach AL, Simons FER, Cardona V, Thong BY-H, Molinari N, et al. Critical view of anaphylaxis epidemiology: open questions and new perspectives. llergy Asthma Clin Immunol. 2018:14:12. doi: 10.1186/s13223-018-0234-0.
14. Tanno LK, Caminati M, Pouessel G, Senna G, Demoly P. Epidemiology of anaphylaxis: is the trend still going up? Curr Opin Allergy Clin Immunol. 2023;23(5):349-56. doi: 10.1097/ACI.0000000000000933.
15. Lee S, Bellolio MF, Hess EP, Erwin P, Murad MH, Campbell RL. Time of Onset and Predictors of Biphasic Anaphylactic Reactions: A Systematic Review and Meta-analysis. J allergy Clin Immunol Pract. 2015;3(3):402-8. doi: 10.1016/j.jaip.2014.12.010.
16. Zeke A, Sudhir A. Management of allergic reactions and anaphylaxis in the emergency department. Emerg Med Pract. 2022;24(7):1-24.
17. de la Hoz Caballer B, Diéguez Muñoz MC, Gonzalez de Olano D, Habernau Mena A, Muñoz Cano R. Guía de actuación en anafilaxia: galaxia. 2022. Available at: https://www.guiagalaxia.com/. doi: 10.18176/19264-19-0.
18. Francuzik W, Ruëff F, Bauer A, Bilò MB, Cardona V, Christoff G, et al. Phenotype and risk factors of venom-induced anaphylaxis: A case-control study of the European Anaphylaxis Registry. J Allergy Clin Immunol. 2021;147(2):653-62.e9. doi: 10.1016/j.jaci.2020.06.008.
19. Aurich S, Dölle-Bierke S, Francuzik W, Bilo MB, Christoff G, Fernandez-Rivas M, et al. Anaphylaxis in Elderly Patients-Data From the European Anaphylaxis Registry. Front Immunol. 2019;10:750. doi: 10.3389/fimmu.2019.00750.
20. Cho H, Kwon J-W. Prevalence of anaphylaxis and prescription rates of epinephrine auto-injectors in urban and rural areas of Korea. Korean J Intern Med. 2019;34(3):643-50. doi: 10.3904/kjim.2018.094.
21. Tham EH, Leung ASY, Pacharn P, Lee S, Ebisawa M, Lee BW, et al. Anaphylaxis - Lessons learnt when East meets West. Pediatr allergy Immunol Off Publ Eur Soc Pediatr Allergy Immunol. 2019;30(7):681-8. doi: 10.1111/pai.13098.
22. Jeon YH, Lee S, Ahn K, Lee SY, Kim KW, Kim HH, et al. Infantile Anaphylaxis in Korea: a Multicenter Retrospective Case Study. J Korean Med Sci. 2019;34(13):e106. doi: 10.3346/jkms.2019.34.e106.
23. Wood RA, Camargo CAJ, Lieberman P, Sampson HA, Schwartz LB, Zitt M, et al. Anaphylaxis in America: the prevalence and characteristics of anaphylaxis in the United States. J Allergy Clin Immunol. 2014;133(2):461-7. doi: 10.1016/j.jaci.2013.08.016.
24. Liew WK, Chiang WC, Goh AE, Lim HH, Chay OM, Chang S, et al. Paediatric anaphylaxis in a Singaporean children cohort: changing food allergy triggers over time. Asia Pac Allergy. 2013;3(1):29-34. doi: 10.5415/apallergy.2013.3.1.29.
25. Nabavi M, Lavavpour M, Arshi S, Bemanian MH, Esmaeilzadeh H, Molatefi R, et al. Characteristics, Etiology and Treatment of

Pediatric and Adult Anaphylaxis in Iran. Iran J Allergy Asthma Immunol. 2017;16(6):480-7.
26. Abunada T, Al-Nesf MA, Thalib L, Kurdi R, Khalil S, ElKassem W, et al. Anaphylaxis triggers in a large tertiary care hospital in Qatar: a retrospective study. World Allergy Organ J. 2018;11(1):20. doi: 10.1186/s40413-018-0200-9.
27. Gomes ER, Demoly P. Epidemiology of hypersensitivity drug reactions. Curr Opin Allergy Clin Immunol. 2005;5(4):309-16. doi: 10.1097/01.all.0000173785.81024.33.
28. Drug allergy: an updated practice parameter. Ann allergy, asthma Immunol Off Publ Am Coll Allergy, Asthma, Immunol. 2010;105(4):259-73. doi: 10.1016/j.anai.2010.08.002.
29. Centre for Clinical Practice at NICE (UK). Anaphylaxis: Assessment to Confirm an Anaphylactic Episode and the Decision to Refer After Emergency Treatment for a Suspected Anaphylactic Episode. Manchester (UK): National Institute for Health and Clinical Excellence (UK); 2011.
30. Dzingina M, Stegenga H, Heath M, Jones D, Rogers G, Kleijnen J, et al. Assessment and referral after emergency treatment of a suspected anaphylactic episode: summary of NICE guidance. BMJ. 2011;343:d7595. doi: 10.1136/bmj.d7595.
31. Vidal C, Antolín-Amérigo D, Reaño M, Valero A, Sastre J; Collaborators; SEAIC Board of Directors. Safety and Quality Recommendations in Allergy Medicine (Spanish acronym, RESCAL). J Investig Allergol Clin Immunol. 2018;28(Suppl. 1):1-39. doi: 10.18176/jiaci.0267.
32. National Institute for Health and Care Excellence. Anaphylaxis Assessment to confirm an anaphylactic episode and the decision to refer after emergency treatment for a suspected anaphylactic episode: NICE; 2016. (Clinical guideline [CG119]). Available at: https://www.nice.org.uk/guidance/qs119/resources. Last access date: 06/30/2023.
33. Muraro A, Werfel T, Hoffmann-Sommergruber K, Roberts G, Beyer K, Bindslev-Jensen C, et al. EAACI Food Allergy and Anaphylaxis Guidelines: Diagnosis and management of food allergy. Allergy Eur J Allergy Clin Immunol. 2014;69(8):1008-25. doi: 10.1111/all.12429.
34. Nwaru BI, Hickstein L, Panesar SS, Roberts G, Muraro A, Sheikh A. Prevalence of common food allergies in Europe: a systematic review and meta-analysis. Allergy. 2014;69(8):992-1007. doi: 10.1111/all.12423.
35. Tejedor Alonso MA, Moro Moro M, Múgica García M V. Epidemiology of anaphylaxis. Clin Exp allergy J Br Soc Allergy Clin Immunol. 2015;45(6):1027-39. doi: 10.1111/cea.12418.
36. Cardona V, Ansotegui IJ, Ebisawa M, El-Gamal Y, Fernandez Rivas M, Fineman S, et al. World allergy organization anaphylaxis guidance 2020. World Allergy Organ J. 2020;13(10):100472. doi: 10.1016/j.waojou.2020.100472.
37. Anaphylaxis: assessment and referral after emergency treatment. London: National Institute for Health and Care Excellence (NICE); 2020.
38. Sampson HA, Muñoz-Furlong A, Campbell RL, Adkinson NFJ, Bock SA, Branum A, et al. 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(2):391-7. doi: 10.1016/j.jaci.2005.12.1303.
39. Loprinzi Brauer CE, Motosue MS, Li JT, Hagan JB, Bellolio MF, Lee S, et al. Prospective Validation of the NIAID/FAAN Criteria for Emergency Department Diagnosis of Anaphylaxis. J allergy Clin Immunol Pract. 2016;4(6):1220-6. doi: 10.1016/j.jaip.2016.06.003.
40. Simons FER, Ardusso LRF, Dimov V, Ebisawa M, El-Gamal YM, Lockey RF, et al. World Allergy Organization Anaphylaxis Guidelines: 2013 update of the evidence base. Int Arch Allergy Immunol. 2013;162(3):193-204. doi: 10.1159/000354543.
41. Muraro A, Roberts G, Clark A, Eigenmann PA, Halken S, Lack G, et al. The management of anaphylaxis in childhood: position paper of the European academy of allergology and clinical immunology. Allergy. 2007;62(8):857-71. doi: 10.1111/j.1398-9995.2007.01421.x.
42. Mueller HL. Diagnosis and treatment of insect sensitivity. J Asthma Res. 1966;3(4):331-3. doi: 10.3109/02770906609106941.
43. Cox LS, Sanchez-Borges M, Lockey RF. World Allergy Organization Systemic Allergic Reaction Grading System: Is a Modification Needed? J allergy Clin Immunol Pract. 2017;5(1):58-62.e5. doi: 10.1016/j.jaip.2016.11.009.
44. Sampson HA. Anaphylaxis and emergency treatment. Pediatrics. 2003;111(6 Pt 3):1601-8.
45. Ring J, Behrendt H. Anaphylaxis and anaphylactoid reactions. Classification and pathophysiology. Clin Rev Allergy Immunol. 1999;17(4):387-99. doi: 10.1007/BF02737644.
46. Blazowski L, Majak P, Kurzawa R, Kuna P, Jerzynska J. A severity grading system of food-induced acute allergic reactions to avoid the delay of epinephrine administration. Ann allergy, asthma Immunol Off Publ Am Coll Allergy, Asthma, Immunol. 2021;127(4):462-470.e2. doi: 10.1016/j.anai.2021.04.015.
47. Kohl KS, Gidudu J, Bonhoeffer J, Braun MM, Buettcher M, Chen RT, et al. The development of standardized case definitions and guidelines for adverse events following immunization. Vaccine. 2007;25(31):5671-4. doi: 10.1016/j.vaccine.2007.02.063.
48. Bonhoeffer J, Bentsi-Enchill A, Chen RT, Fisher MC, Gold MS, Hartman K, et al. Guidelines for collection, analysis and presentation of vaccine safety data in surveillance systems. Vaccine. 2009;27(16):2289-97. doi: 10.1016/j.vaccine.2008.11.035.
49. Gold MS, Gidudu J, Erlewyn-Lajeunesse M, Law B. Can the Brighton Collaboration case definitions be used to improve the quality of Adverse Event Following Immunization (AEFI) reporting? Anaphylaxis as a case study. Vaccine. 2010;28(28):4487-98. doi: 10.1016/j.vaccine.2010.04.041.
50. Erlewyn-Lajeunesse M, Dymond S, Slade I, Mansfield HL, Fish R, Jones O, et al. Diagnostic utility of two case definitions for anaphylaxis: a comparison using a retrospective case notes analysis in the UK. Drug Saf. 2010;33(1):57-64. doi: 10.2165/11318970-000000000-00000.
51. Stafford A, Patel N, Turner PJ. Anaphylaxis-moving beyond severity…. J Allergy Clin Immunol. 2021;148(1):83-5. doi: 10.1016/j.jaci.2021.04.017.
52. Dribin TE, Sampson HA, Camargo CA, Brousseau DC, Spergel JM, Neuman MI, et al. Persistent, refractory, and biphasic anaphylaxis: A multidisciplinary Delphi study. J Allergy Clin Immunol. 2020;146(5):1089-96. doi: 10.1016/j.jaci.2020.08.015.
53. Sala-Cunill A, Cardona V, Labrador-Horrillo M, Luengo O, Esteso O, Garriga T, et al. Usefulness and limitations of sequential serum tryptase for the diagnosis of anaphylaxis in 102 patients. Int Arch Allergy Immunol. 2013;160(2):192-9. doi: 10.1159/000339749.
54. Brown SGA, Blackman KE, Heddle RJ. Can serum mast cell tryptase help diagnose anaphylaxis? Emerg Med Australas. 2004;16(2):120-4. doi: 10.1111/j.1742-6723.2004.00562.x.
55. Valent P, Bonadonna P, Hartmann K, Broesby-Olsen S, Brockow K, Butterfield JH, et al. Why the 20% + 2 Tryptase Formula Is a

Diagnostic Gold Standard for Severe Systemic Mast Cell Activation and Mast Cell Activation Syndrome. Int Arch Allergy Immunol. 2019;180(1):44-51. doi: 10.1159/000501079.
56. Passia E, Jandus P. Using Baseline and Peak Serum Tryptase Levels to Diagnose Anaphylaxis: a Review. Clin Rev Allergy Immunol. 2020;58(3):366-76. doi: 10.1007/s12016-020-08777-7.
57. Hansen K, Bjornsson HM, Gunnbjornsdottir MI. [Utility of serum tryptase in Emergency Department patients with possible anaphylaxis]. Laeknabladid. 2021;107(10):470-5.
58. Lyons JJ, Yu X, Hughes JD, Le QT, Jamil A, Bai Y, et al. Elevated basal serum tryptase identifies a multisystem disorder associated with increased TPSAB1 copy number. Nat Genet. 2016;48(12):1564-9. doi: 10.1038/ng.3696.
59. Valent P, Horny HP, Escribano L, Longley BJ, Li CY, Schwartz LB, et al. Diagnostic criteria and classification of mastocytosis: a consensus proposal. Leuk Res. 2001;25(7):603-25. doi: 10.1016/s0145-2126(01)00038-8.
60. Schwartz LB. Diagnostic value of tryptase in anaphylaxis and mastocytosis. Immunol Allergy Clin North Am. 2006;26(3):451-63. doi: 10.1016/j.iac.2006.05.010.
61. Valent P, Akin C, Arock M, Brockow K, Butterfield JH, Carter MC, et al. Definitions, criteria and global classification of mast cell disorders with special reference to mast cell activation syndromes: a consensus proposal. Int Arch Allergy Immunol. 2012;157(3):215-25. doi: 10.1159/000328760.
62. Watkins J, Wild G. Improved diagnosis of anaphylactoid reactions by measurement of serum tryptase and urinary methylhistamine. Ann Fr Anesth Reanim. 1993;12(2):169-72. doi: 10.1016/S0750-7658(05)81026-3.
63. Awad JA, Morrow JD, Roberts LJ 2nd. Detection of the major urinary metabolite of prostaglandin D2 in the circulation: demonstration of elevated levels in patients with disorders of systemic mast cell activation. J Allergy Clin Immunol. 1994;93(5):817-24. doi: 10.1016/0091-6749(94)90371-9.
64. Keyzer JJ, de Monchy JG, van Doormaal JJ, van Voorst Vader PC. Improved diagnosis of mastocytosis by measurement of urinary histamine metabolites. N Engl J Med. 1983;309(26):1603-5. doi: 10.1056/NEJM198312293092603.
65. Turner PJ, Campbell DE, Motosue MS, Campbell RL. Global Trends in Anaphylaxis Epidemiology and Clinical Implications. J allergy Clin Immunol Pract. 2020;8(4):1169-76. doi: 10.1016/j.jaip.2019.11.027.
66. Turner PJ, Muraro A, Roberts G. Pharmacokinetics of adrenaline autoinjectors. Clin Exp Allergy. 2022;52(1):18-28. doi: 10.1111/cea.14055.
67. Soar J, Pumphrey R, Cant A, Clarke S, Corbett A, Dawson P, et al. Emergency treatment of anaphylactic reactions--guidelines for healthcare providers. Resuscitation. 2008;77(2):157-69. doi: 10.1016/j.resuscitation.2008.02.001.
68. Kemp AS, Hu W. New action plans for the management of anaphylaxis. Aust Fam Physician. 2009;38(1-2):31-5.
69. Nurmatov U, Worth A, Sheikh A. Anaphylaxis management plans for the acute and long-term management of anaphylaxis: a systematic review. J Allergy Clin Immunol. 2008;122(2):353-61, 361.e1-3. doi: 10.1016/j.jaci.2008.05.028.
70. Tan E, O'Sullivan M, Crozier T, Kolawole H, Mantas S, Said M, et al. Acute management of anaphylaxis in pregnancy. Aust J Gen Pract. 2022;51(6):405-8. doi: 10.31128/AJGP-03-21-5918.
71. Whyte AF, Soar J, Dodd A, Hughes A, Sargant N, Turner PJ. Emergency treatment of anaphylaxis: concise clinical guidance. Clin Med. 2022;22(4):332-9. doi: 10.7861/clinmed.2022-0073.
72. Shaker MS, Wallace D V, Golden DBK, Oppenheimer J, Bernstein JA, Campbell RL, et al. Anaphylaxis-a 2020 practice parameter update, systematic review, and Grading of Recommendations, Assessment, Development and Evaluation (GRADE) analysis. J Allergy Clin Immunol. 2020;145(4):1082-123. doi: 10.1016/j.jaci.2020.01.017.
73. ASCIA. Acute Management of Anaphylaxis. Allergy.org.au. 2023. Available at: https://www.allergy.org.au/images/ASCIA_HP_Guidelines_Acute_Management_Anaphylaxis_2023.pdf. Last access date: 06/30/2023.
74. Liu X, Lee S, Lohse CM, Hardy CT, Campbell RL. Biphasic Reactions in Emergency Department Anaphylaxis Patients: A Prospective Cohort Study. J Allergy Clin Immunol Pract. 2020;8(4):1230-8. doi: 10.1016/j.jaip.2019.10.027.
75. Liyanage CK, Galappatthy P, Seneviratne SL. Corticosteroids in management of anaphylaxis; a systematic review of evidence. Eur Ann Allergy Clin Immunol. 2017;49(5):196-207. doi: 10.23822/EurAnnACI.1764-1489.15.
76. Sheikh A, Ten Broek V, Brown SG, Simons FE. H1-antihistamines for the treatment of anaphylaxis: Cochrane systematic review. Allergy. 2007;62(8):830-7. doi: 10.1111/j.1398-9995.2007.01435.x.
77. Pajno GB, Fernandez-Rivas M, Arasi S, Roberts G, Akdis CA, Alvaro-Lozano M, et al. EAACI Guidelines on allergen immunotherapy: IgE-mediated food allergy. Allergy. 2018;73(4):799-815. doi: 10.1111/all.13319.
78. Golden DBK. Anaphylaxis to insect stings. Immunol Allergy Clin North Am. 2015;35(2):287-302. doi: 10.1016/j.iac.2015.01.007.
79. Oude Elberink JN, van der Heide S, Guyatt GH, Dubois AE. Immunotherapy improves health-related quality of life of adult patients with dermal reactions following yellow jacket stings. Clin Exp Allergy. 2009;39(6):883-9. doi: 10.1111/j.1365-2222.2009.03230.x.
80. Global implementation of the world health organization's International Classification of Diseases (ICD)-11: The allergic and hypersensitivity conditions model. Allergy. 2020;75(9):2206-18. doi: 10.1111/all.14468.
81. Muraro A, Agache I, Clark A, Sheikh A, Roberts G, Akdis CA, et al. EAACI Food Allergy and Anaphylaxis Guidelines: Managing patients with food allergy in the community. Allergy Eur J Allergy Clin Immunol. 2014;69(8):1046-57. doi: 10.1111/all.12441.
82. Epipen B. Adrenaline (Epinephrine) Injector Prescription. 2021:1-5.
83. Burrell S, Patel N, Vazquez-Ortiz M, Campbell DE, DunnGalvin A, Turner PJ. Self-administration of adrenaline for anaphylaxis during in-hospital food challenges improves health-related quality of life. Arch Dis Child. 2021;106(6):558-63. doi: 10.1136/archdischild-2020-319906.
84. Kelleher MM, Dunngalvin A, Sheikh A, Cullinane C, Fitzsimons J, Hourihane JO. Twenty four-hour helpline access to expert management advice for food-allergy-triggered anaphylaxis in infants, children and young people: a pragmatic, randomized controlled trial. Allergy. 2013;68(12):1598-604. doi: 10.1111/all.12310.

85. Greenhawt M, Gupta RS, Meadows JA, Pistiner M, Spergel JM, Camargo CA et al. Guiding Principles for the Recognition, Diagnosis, and Management of Infants with Anaphylaxis: An Expert Panel Consensus. J Allergy Clin Immunol Pract. 2019;7(4):1148-56.e5. doi: 10.1016/j.jaip.2018.10.052.
86. Fernández-Rivas M, Gómez García I, Gonzalo-Fernández A, Fuentes Ferrer M, Dölle-Bierke S, Marco-Martín G, et al. Development and validation of the food allergy severity score. Allergy. 2022;77(5):1545-58. doi: 10.1111/all.15165.
87. Alvarez-Perea A, Cabrera-Freitag P, Fuentes-Aparicio V, Infante S. Advancements in Anaphylaxis Management. Curr Pharm Des. 2023;29(3):185-95. doi: 10.2174/1381612829666221021150946.
88. Waserman S, Avilla E, Harada L, Huang J, Kastner M. Decades of poor availability of epinephrine autoinjectors: Global problems in need of global solutions. Ann Allergy Asthma Immunol. 2020;124(2):205-7.e1. doi: 10.1016/j.anai.2019.11.009.
89. Cheng A. Emergency treatment of anaphylaxis in infants and children. Paediatr Child Health. 2011;16(1):35-40.
90. Committee on Infectious Diseases AA of P, Kimberlin DW, Barnett ED, Lynfield R, Sawyer MH, editors. Treatment of Anaphylactic Reactions. Red Book: 2021-2024 Report of the Committee on Infectious Diseases. American Academy of Pediatrics; 2021. doi: 10.1542/9781610025782-S1_005_005.
91. Duvauchelle T, Robert P, Donazzolo Y, Loyau S, Orlandini B, Lehert P, et al. Bioavailability and Cardiovascular Effects of Adrenaline Administered by Anapen Autoinjector in Healthy Volunteers. J Allergy Clin Immunol Pract. 2018;6(4):1257-63. doi: 10.1016/j.jaip.2017.09.021.apr.
92. Jext 300 micrograms Solution for Injection in pre-filled pen - Summary of Product Characteristics (SmPC) - (emc).
93. Jext 150 micrograms Solution for Injection in pre-filled pen - Summary of Product Characteristics (SmPC) - (emc).

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Inês Nunes[1], Graça Loureiro[1], Beatriz Tavares[1], Ana Todo-Bom[1], Rosário Cunha[2]

Sensitization to genuine markers of timothy grass pollen (*Phleum pratense*) in the North-Central region of Portugal

[1]Department of Allergy and Clinical Immunology, Hospital and University Center of Coimbra, Coimbra, Portugal
[2]Department of Clinical Pathology, Hospital and University Center of Coimbra, Coimbra, Portugal

Key words

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Corresponding author
Inês Nunes
Department of Allergy and Clinical Immunology
Hospital and University Center of Coimbra
Praceta Professor Mota Pinto
3004-561, Coimbra, Portugal
ORCID: 0000-0003-3889-2748
E-mail: inesnunes0929@gmail.com

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Impact statement

Phl p1 sensitization, marking genuine grass pollen sensitization, is the most frequently found in the North-Central region of Portugal.

Summary

Background. *Pollen is the main cause of respiratory allergy and its prevalence is increasing. Timothy grass (Phleum pratense) is one of the most common pollen sources, and one of the best characterized allergenic grasses. The major allergens Phl p1, Phl p2, Phl p5 and Phl p6 are considered markers of genuine grass pollen sensitization.* ***Methods.*** *It is a retrospective study. IgE levels of Phl p1 and Phl p5 in patients living in the North-Central region of Portugal were analyzed, considering age and area of residence (inland or coastal).* ***Results.*** *Among the 188 patients with IgE results for Phl p1 and Phl p5, sensitization to Phl p1 and Phl p5b was observed in 97.87% and 63.83%, respectively. The majority demonstrated co-sensitization to both Phl p1 and Phl p5b, while 68 patients were monosensitized to Phl p1 and 4 patients were monosensitized to Phl p5b. Either patients living in coastal or inland areas showed IgE levels of Phl p1 higher than Phl p5b.* ***Conclusions.*** *Regarding genuine grass pollen sensitization, Phl p1 sensitization is more prevalent than Phl p5b in the North-Central region of Portugal.*

Introduction

In the last decades, the prevalence of respiratory allergies induced by pollens has been increasing. Atmospheric pollen in the outdoor environment is the main cause of rhinitis, bronchial asthma, rhinoconjunctivitis and eczema in individuals with pollinosis (1). In Europe, especially in the Mediterranean countries, Poaceae family pollen (grasses) is one of the most important causes of pollinosis (2). In the Pooideae subfamily, timothy grass (*Phleum pratense*), orchard grass (*Dactylis glomerata*), perennial ryegrass (*Lolium perenne*) and bluegrass (*Poa pratensis*) are the most common pollen sources (3). Poaceae pollen is the most prevalent pollen and the main cause of pollinosis in Portugal (4). In pollen allergy diagnosis, it is important to distinguish co-sensitization to several allergen sources and sensitization to cross-reactive components. In pollen allergy treatment, the content of commercial pollen allergen immunotherapy (AIT) extracts is not always known, but the presence of major allergens is often stated (5, 6). Molecular diagnosis can improve allergy diagnosis (7), selecting patients eligible for AIT and allowing an accurate individual prescription, which will avoid inappropriate or ineffective therapies (8). Timothy grass pollen (*Phleum pratense*) is one of

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***Table I** - Timothy allergenic molecules listed by the IUIS Allergen Nomenclature Sub-Committee (http://www.allergen.org/index.php).*

Allergen name	Biochemical name	Allergenicity
Phl p 1	CCD-bearing protein/beta-expansin	95%
Phl p 2	Grass group II/III	65%
Phl p 3	Pollen allergen 1	60%
Phl p 4	CCD-bearing protein/ berberine bridge enzyme	75%
Phl p 5	Grass group V	95%
Phl p 6	Grass group VI	75%
Phl p 7	Polcalcin	10%
Phl p 11	Ole e 1-related protein	32%
Phl p 12	Profilin	15%
Phl p 13	Grass group XIII/ polygalacturonase	50%

the best characterized allergenic grasses (9), with ten allergenic molecules already described and officially listed by the IUIS Allergen Nomenclature Sub-Committee, at date (**table I**) (3, 10). ImmunoCAP singleplex IgE tests available for the Phl p allergenic molecules are Phl p1, Phl p2, Phl p4, Phl p5b, Phl p6, Phl p7, Phl p11 and Phl p12.

The major allergens Phl p1, Phl p2, Phl p5 and Phl p6 are considered markers of genuine sensitization (3). The present study aims at characterizing molecular allergens of timothy grass pollen genuine sensitization in patients living in the North-Central region of Portugal.

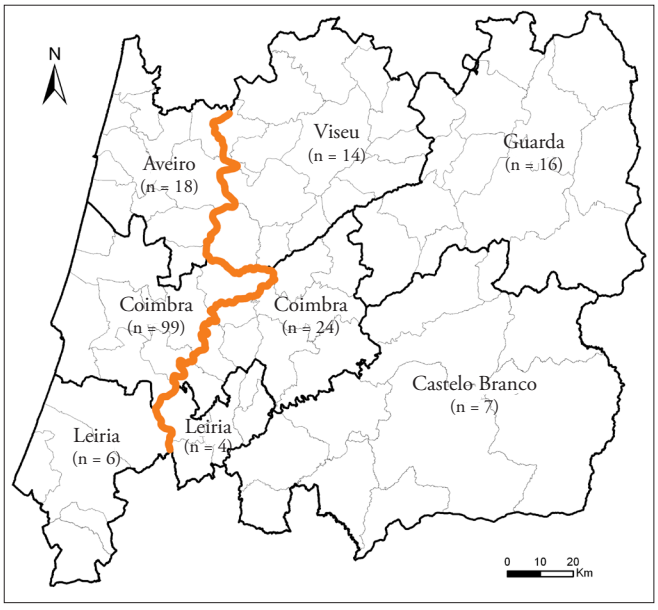
Materials and methods

Retrospective study performed in the Clinical Pathology Department of Coimbra University Hospital, a central hospital that receives patients from all over the North-Central region of Portugal. A parameterized search in Clinidata XXI 5.3.12 SP1 software was made. Patients who were asked for specific IgE antibodies to Phl p1, Phl p5 and Phl p6 were included, from 01/06/2014 until 31/07/2020. Since our laboratory does not have Phl p2 analysis available, it was not considered. For allergen 5, the isoform available is Phl p5b. IgE assay was made by fluorescence enzyme immunoassay (ImmunoCAP, Thermo Fisher Scientific) with a detection sensitivity between 0.01 and 100 kilounits per liter (kU_A/L). Results were classified as positive or negative according to the cutoff point of 0.35 kU_A/L. Exclusion criteria were previous AIT, repeated determinations from the same patient (the highest levels or those prior to AIT treatment were considered) and patients living outside North-Central region of Portugal (included districts of Coimbra, Guarda, Leiria, Castelo Branco, Viseu and Aveiro). As only one result of Phl p6 was found, genuine molecular sensitization pattern of *Phleum pratense* allergens focused only in the allergens Phl p1 and/or Phl p5b specific IgE assays. To characterize and compare the genuine molecular sensitization pattern of *Phleum pratense*, only patients who were asked for the two allergens and that were positive to at least one Phl p1 and/or Phl p5b specific IgE were considered. These patients were divided according to sensitization profile: monosensitization to Phl p1 (group I), monosensitization to Phl p5b (group II) and sensitization to both Phl p1 and Phl p5b allergens (group III). In each group the sensitization profile was analyzed according to age (children if age < 18 years and adult if age ≥ 18 years) and area of residence (coastal *versus* inland areas) (**figure 1**).

Statistical analysis

Data analysis was carried out using SPSS version 25 for Windows. Results were presented as median (interquartile range (IQR)) and mean (standard deviation). Statistical analysis was performed with nonparametric tests (Wilcoxon signed-rank and Mann Whitney U tests). A level of significance of 0.05 was considered (P-value ≤ 0.05).

***Figure 1** - Map of the North-Central region of mainland Portugal.*



The orange line limits the coast (on the left) and the inland areas (on the right side). The number of patients included in each district is shown on the map. Adapted from https://www.ccdrc.pt/.

Results

A total of 508 results were found, corresponding to 319 patients who were asked for at least one specific IgE to Phl p1 and/or Phl p5b (168 female, mean age 25.21 ± 1.67 years). The prevalence of Phl p1 and Phl p5b sensitization was 296/302 and 134/206, respectively, as shown in **table II**.

To characterize and compare the genuine molecular sensitization pattern of *Phleum pratense*, from the initial 319 patients, we considered the 188 patients with available results to both molecular allergens. None of the patients had both negative results. All the included patients had a positive result to Phl p1 and/or Phl p5b. This total group included 95 female and 93 male aged 4 to 65 years, of which 59 were children (mean age 11.44 ± 3.65 years, 66.1% male) and 159 were adults (mean age 31.43 ± 11.40 years, 41.86% male). In **table III** the characteristics of these patients divided in the three groups are described: monosensitized to Phl p1 (group I), monosensitized to Phl p5b (group II) and sensitized to both Phl p1 and

Table II - *Prevalence of Phl p1 and Phl p5b sensitization.*

	Total	Phl p1	Phl p5b
Number of results	508	302	206
Number of positive results (> 0.35 kU_A/L)	430	296	134

Table III - *Characteristics of sensitization to Phl p1 and/or Phl p5b.*

	Total Group		Group I	Group II	Group III	
	Phl p1	Phl p5b	Phl p1	Phl p5b	Phl p1	Phl p5b
Total (n)	188	188	68	4	116	116
Gender (f/m)	95/93	95/93	40/28	03-gen	52/64	52/64
Age (years)	25.15 ± 13.40	25.15 ± 13.40	25.97 ± 14.11	35 ± 16.67	24.34 ± 12.82	24.34 ± 12.82
IgE (kU_A/L)						
Mean ± SD	27.59 ± 31.34	20.89 ± 29.40	12.70 ± 21.26	22.43 ± 20.10	37.26 ± 32.90	33.07 ± 31.43
Median	14.75	6.85	4.09	15.50	25.75	21.9
Minimum	0	0	0.48	6.91	0.37	0.6
Maximum	100	100	98.90	51.80	100	100
Q_1	4.15	0.01	1.68	8.43	10.3	8.15
Q_3	39.15	32.70	14.23	43.35	56.92	47.95
Children (n)	59	59	21	1	37	37
Gender (f/m)	20/39	20/39	lug-14	0/1	13/24	13/24
Age (years)	11.44 ± 3.65	11.44 ± 3.65	11.38 ± 3.53	10	11.51 ± 3.81	11.51 ± 3.81
IgE (kU_A/L)						
Mean ± SD	43.25 ± 37.39	31.71 ± 37.11	20.62 ± 30.63	18	57.25 ± 34.34	50.07 ± 35.80
Median	29.90	15.30	7.92	18	52.4	41.6
Minimum	0.20	0	0.48	18	4.59	0.6
Maximum	100	100	98.90	18	100	100
Q_1	8.00	0.01	2.98	18	24.4	18.75
Q_3	91.70	49.00	18	18	100	100
Adults (n)	129	129	47	3	79	79
Gender (f/m)	75/54	75/54	33/14	3/0	61/68	61/68
Age (years)	31.43 ± 11.40	31.43 ± 11.40	32.49 ± 12.01	43.33 ± 0.58	30.34 ± 11.0	30.34 ± 11.0
IgE (kU_A/L)						
Mean ± SD	20.42 ± 25.24	15.94 ± 23.67	9.16 ± 14.46	23.90 ± 24.35	27.90 ± 27.81	25.11 ± 25.77
Median	10.60	5.64	3.28	13	18.1	15
Minimum	0	0	0.49	6.91	0.37	0.65
Maximum	100	100	84.7	51.8	100	100
Q_1	2.92	0.00	1.59	n	7.4	6.52
Q_3	26.75	21.40	11.3	n	33.9	35.4

Group I: monosensitization to Phl p1; group II: monosensitization to Phl p5b; group III: sensitization to both Phl p1 and Phl p5b; f: female; m: male; SD: standard deviation. Descriptive statistic: mean, standard deviation, median, minimum, maximum, Q_1: first quartile and Q_3: third quartile.

Figure 2 - *Graphical representation of IgE levels (kU_A/L) for allergens Phl p1 and Phl p5b (y-axis) in the total group and children and adults' subgroups (x-axis).*

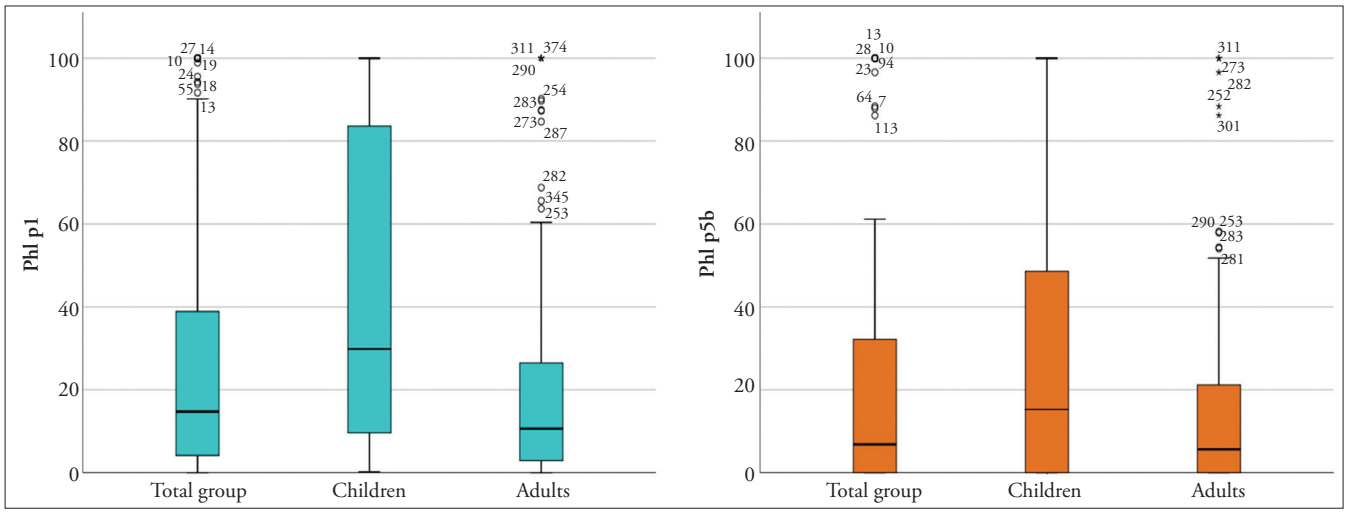


Table IV - *Characteristics of sensitization to Phl p1 and/or Phl p5b according to the area of residence.*

	Total Group		Group I	Group I	Group III	
	Phl p1	**Phl p5b**	**Phl p1**	**Phl p5b**	**Phl p1**	**Phl p5b**
Coastal areas						
Total (n)	123	123	48	3	72	72
Gender (f/m)	66/57	66/57	28/20	02-gen	36/36	36/36
Age (years)	25.45 ± 13.36	25.45 ± 13.36	25.90 ± 13.32	32 ± 19.05	24.88 ± 13.29	24.88 ± 13.29
IgE (kU_A/L)						
Mean ± SD	25.91 ± 31.99	18.76 ± 29.05	12.34 ± 21.35	27.6 ± 21.11	36.04 ± 34.64	30.89 ± 32.36
Median	11.3	5.3	3.42	18	22.7	15
Minimum	0	0	0.49	13	0.37	0.6
Maximum	100	100	98.9	51.8	100	100
Q_1	3.07	0	1.58	N	8.14	6.45
Q_3	32	26.5	13.55	N	57.08	44.88
Inland areas						
Total (n)	65	65	20		44	44
Gender (f/m)	29/36	29/36	12-ago	1/0	16/28	16/28
Age (years)	24.6 ± 13.57	24.6 ± 13.57	26.15 ± 16.24	44	23.45 ± 12.10	23.45 ± 12.10
IgE (kU_A/L)						
Mean ± SD	30.75 ± 30.07	24.92 ± 29.88	13.58 ± 21.57	6.91	39.26 ± 30.12	36.64 ± 29.87
Median	20.1	15.0	4.51	N	29.4	26.7
Minimum	0.01	0.00	0.48	N	1.87	1.03
Maximum	100	100	93.80	N	100	100
Q_1	7.03	0.01	2.04	N	14.95	14.18
Q_3	43.45	41.1	15.43	N	56.83	52.68

Group I: monosensitization to Phl p1; group II: monosensitization to Phl p5b; group III: sensitization to both Phl p1 and Phl p5b; f: female; m: male. Descriptive statistic: mean, standard deviation, median, minimum, maximum, Q_1: first quartile and Q_3: third quartile.

Phl p5b molecular allergens (group III). IgE levels of the total group are also represented in **figure 2**. In the total group of 188 patients, 72 (38.3%) patients were sensitized to only one molecular allergen, while the majority, 116 patients (61.7%), were sensitized for both molecular allergens. Concerning age subgroups, the same pattern was observed (62.7% of the children and 61.2% of the adults were sensitized to both allergens). Levels of IgE to Phl p1 were higher than IgE to Phl p5b, in total group and in both subgroups, children and adults ($p < 0.001$, $p = 0.002$ and $p < 0.001$ respectively). Higher levels of both molecular allergens were found in children when compared to adults (Phl p1 $p < 0.001$ and Phl p5b $p = 0.016$).
Concerning patients of group III (sensitized to both allergens), Phl p1 levels were higher than Phl p5b, besides no statistical difference was found ($p = 0.074$). In children and adult groups, Phl p1 levels were also higher than Phl p5b ($p = 0.162$ and $p = 0.208$, respectively). Comparing children to adult groups, children have higher IgE levels than adults of both Phl p1 and Phl p5b ($p < 0.001$).
Considering the 72 patients with only one sensitization, 68 (94.4%) were only sensitized to Phl p1 (Phl p5b negative, representing 36.2% of the total group) and only four patients (5.6%) were sensitized to Phl p5b (Phl p1 negative, representing 2.1% of the total group). In group I, children also showed higher levels of IgE comparing to adults. Although the sample of group II is small, the 4 patients exclusively sensitized to Phl p5b showed higher IgE levels comparing to IgE levels of group I patients.
Levels of IgE to Phl p1 in group III were significantly higher than in group I ($p < 0.001$).
Considering the area of residence, the 188 patients' group was organized into two groups, according on whether they lived inland or coastal (**table IV**). Either patients living in coastal or inland areas showed IgE levels of Phl p1 higher than IgE to Phl p5b ($p < 0.001$ and $p = 0.009$, respectively). This could be mainly because the number of monosensitized patients to Phl p1. If we focus on group III, IgE to Phl p1 is also higher, but the difference is not significative ($p = 0.106$ for coastal and $p = 0.414$ for inland). Considering group II, only three patients from coastal areas (2.4% of coastal residents) and one patient from inland (1.5% of inland residents) were monosensitized to Phl p5b. No significant differences were found between IgE coastal levels and IgE inland levels.

Discussion and conclusions

Phl p1 and Phl p5 are considered major allergens – allergens recognized by IgE antibodies in over 50% of allergic patients to an allergen source (3). Both are also considered markers of genuine sensitization to timothy grass pollen. Sensitization to Phl p1 is the most prevalent component sensitization, with a sensitization prevalence of > 90% and Phl p5 has a lower sensitization prevalence with specific IgE present in 65-90% of grass pollen allergic patients (9). Besides less prevalent, Phl p5 often has higher IgE levels (3). In our analysis group we found sensitization to Phl p1 in 97.87% of the patients and sensitization to Phl p5b in 63.83%. Similar frequency percentages were found between children and adults for both allergens (respectively 98.3% of children and 97.67% of adults for Phl p1, and 64.40% of children and 63.57% of adults for Phl p5b).
Besides Phl p1 is not the only marker of genuine sensitization, IgE to other markers (Phl p2, Phl p5 and Phl p6) are rarely observed in the absence of IgE to Phl p1 (3). According to our data, the majority of the patients (61.7%) showed sensitization to both genuine markers or monosensitization to Phl p1 (36.2%). We only found four patients (2.1%) with exclusive sensitization to Phl p5b (Phl p1 negative) and these four patients showed, in fact, higher IgE levels to Phl p5b comparing to monosensitization levels to Phl p1. Other authors also showed that monosensitization to Phl p5 is rare (10).
Sensitization to *Phleum pratense* molecules usually start in early childhood with an IgE response to Phl p1 that later grows by involving other grass molecules (10). Hatzler *et al*. studied the progression of IgE sensitization responses against grass pollen showing that in > 75% of cases, Phl p1 sensitization precedes other grass pollen sensitizations (11). In our population, only one child had exclusive sensitization to Phl p5b without any IgE response to Phl p1. Comparing children and adults, except in group II, children showed higher IgE levels.
Our data showed also higher levels of IgE in polysensitized patients comparing to IgE levels of monosensitized patients. According to geographic area we found similar frequencies. Sensitization to Phl p1 was found in 97.56% and 98.46% in cosatal and inland areas respectively. The frequency of Phl p5b sensitization was slightly higher in inland comparing to coastal, with 69.2% *versus* 60.98% of the patients. On the other hand, monosensitization to Phl p1 was lower in inland comparing to coastal, 30.77% *versus* 39.0% of the patients.
There are no other studies about molecular sensitization to pollen in the North-Central region of Portugal. Tavares *et al*. analyzed sensitization to pollens based on skin prick tests (12). Sensitization to grass pollen was detected in 72.0% of the patients with pollen sensitization, of which 27.6% were also sensitized to the pan-allergen profilin (Pho d2). Monosensitization to grass pollen was not associated with sensitization to profilin. Considering the large extent of cross-reactivity between plant profilins, sensitivity to Phl p12 will probably have a similar frequency (*Phleum pratense* profilin, minor allergen).
In the Southern region of Portugal, a characterization of molecular profiles of *Phleum pratense* pollen was made. Almeida *et al*. also found that the majority of patients were sensitized to Phl p1; patients sensitized to both allergens had higher IgE levels; monosensitization frequency to Phl p1 was similar (40.0% *versus* 36.2%), and children also showed higher mean IgE levels than adults (13). On

the other hand, in the North-Central region, mean IgE levels were higher comparing to the South, both for children (43.25 kU/L *versus* 22.49 kU/L and 31.71 kU/L *versus* 20.23 kU/L, respectively, for Phl p1 and Phl p5b) and adults (20.42 kU/L *versus* 10.46 kU/L and 15.96 kU/L *versus* 8.43 kU/L, respectively for Phl p1 and Phl 5b). Sensitization to Phl p5b was more frequent in our data (63.83% *versus* 44.4%) while sensitization to Phl p1 was > 90% in both studies. IgE to Phl p1 seems to be the most important molecular allergen of *Phleum pratense* in Portugal.

Barber *et al*. measured IgE to the different allergens in different regions of Spain (14), including Phl p1 and Phl p5. Spain borders Portugal and, in particular, the provinces of Salamanca and Cáceres border the central region of Portugal (districts of Guarda and Castelo Branco, respectively). The authors found a very high prevalence of Phl p1 (> 76%) with IgE median of 10.83 kU/L in both provinces. In Portuguese inland areas, prevalence of Phl p1 was also very high (97.56%) but the median IgE seems to be higher (20.1 kU/L). Considering sensitization to Phl p5, Spanish border regions showed a prevalence between 26-50% *versus* 69.2% in Portuguese inland areas, with median IgE to Phl p5 of 14.52 kU/L (Salamanca) and 9.31 kU/L (Cárceres) *versus* 15.0 kU/L in Portuguese inland areas.

We acknowledge study limitations. As a retrospective study, it was only possible to analyze the previous available results facing the limitation that not all patients were tested for all allergens. Furthermore, no correlation was made with clinical severity of allergic disease.

In conclusion, our characterization of timothy grass pollen molecular pattern showed that, in the North-Central region of Portugal, Phl p1 sensitization is more common than Phl p5b. Besides both being major and also markers of genuine sensitization, when considering AIT, Phl p1 would be more sensible. IgE to Phl p5b should not be considered alone, not least because monosensitization to Phl p5b is rare. As Phl p5b sensitization can also indicate long-term disease, in children and/or patients with recent onset disease it may still not be detected, which can lead to wrong conclusions.

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Contributions

IN: conceptualization, methodology, investigation, data curation, formal analysis, writing – original draft, writing – review & editing. GL: conceptualization, methodology, writing – review & editing. BT: conceptualization, formal analysis, writing – review & editing. ATB: writing – review & editing. RC: resources, writing – review & editing.

Conflict of interests

The authors declare that they have no conflict of interests.

References

1. D'Amato G, Cecchi L, Bonini S, Nunes C, Annesi-Maesano I, Behrendt H, et al. Allergenic pollen and pollen allergy in Europe. Allergy. 2007;62(9):976-90. doi: 10.1111/j.1398-9995.2007.01393.x.
2. Camacho I, Caeiro E, Nunes C, Morais-Almeida M. Airborne pollen calendar of Portugal: a 15-year survey (2002-2017). Allergol Immunopathol (Madr). 2020;48(2):194-201. doi: 10.1016/j.aller.2019.06.012.
3. Matricardi PM, Kleine-tebbe J, Hoffmann J, Valenta R, Ollert M, Ree R Van, et al. EAACI Molecular Allergology User's Guide. Pediatr Allergy Immunol. 2016;27 Suppl 23:1-250. doi: 10.1111/pai.12563.
4. Caeiro E, Camacho IC, Lopes L, Gaspar Â, Todo-Bom A, Ferraz De Oliveira J, et al. Análise das concentrações de pólen de gramíneas na atmosfera de Portugal Continental. Rev Port Imunoalergologia. 2014;22(2):125-38. Available at: http://hdl.handle.net/10400.13/3069.
5. Asero R, Mistrello G, Amato S. Detection of pan-allergens in commercial pollen extracts for allergen immunotherapy. Ann Allergy Asthma Immunol. 2016;117(2):180-5. doi: 10.1016/j.anai.2016.05.010.
6. Tripodi S, Frediani T, Lucarelli S, MacR F, Pingitore G, Di Rienzo Businco A, et al. Molecular profiles of IgE to Phleum pratense in children with grass pollen allergy: Implications for specific immunotherapy. J Allergy Clin Immunol. 2012;129(3):834-9.e8. doi: 10.1016/j.jaci.2011.10.045.
7. Sastre J, Sastre-Ibañez M. Molecular diagnosis and immunotherapy. Curr Opin Allergy Clin Immunol. 2016;16(6):565-570. doi: 10.1097/ACI.0000000000000318.
8. Douladiris N, Savvatianos S, Roumpedaki I, Skevaki C, Mitsias D, Papadopoulos NG. A molecular diagnostic algorithm to guide pollen immunotherapy in southern Europe: towards component-resolved management of allergic diseases. Int Arch Allergy Immunol. 2013;162(2):163-72. doi: 10.1159/000353113.
9. Pablos I, Wildner S, Asam C, Wallner M, Gadermaier G. Pollen Allergens for Molecular Diagnosis. Curr Allergy Asthma Rep. 2016;16(4):31. doi: 10.1007/s11882-016-0603-z.
10. Cipriani F, Mastrorilli C, Tripodi S, Ricci G, Perna S, Panetta V, et al. Diagnostic relevance of IgE sensitization profiles to eight recombinant Phleum pratense molecules. Allergy. 2018;73(3):673-82. doi: 10.1111/all.13338.
11. Hatzler L, Panetta V, Lau S, Wagner P, Bergmann RL, Illi S, et al. Molecular spreading and predictive value of preclinical IgE response to Phleum pratense in children with hay fever. J Allergy Clin Immunol. 2012;130(4):894-901.e5. doi: 10.1016/j.jaci.2012.05.053.
12. Tavares B, Machado D, Loureiro G, Cemlyn-Jones J, Pereira C. Sensitization to profilin in the Central region of Portugal. Sci Total Environ. 2008;407(1):273-8. doi: 10.1016/j.scitotenv.2008.08.013.
13. Almeida E, Caeiro E, Todo-Bom A, Duarte A, Gazarini L. Sensitization to grass allergens: Phl p1, Phl p5 and Phl p7 Phl p12 in adult and children patients in Beja (Southern Portugal). Allergol Immunopathol (Madr). 2019;47(6):579-84. doi: 10.1016/j.aller.2019.04.006.
14. Barber D, De La Torre F, Lombardero M, Antépara I, Colas C, Dávila I, et al. Component-resolved diagnosis of pollen allergy based on skin testing with profilin, polcalcin and lipid transfer protein pan-allergens. Clin Exp Allergy. 2009;39(11):1764-73. doi: 10.1111/j.1365-2222.2009.03351.x.

ORIGINAL ARTICLE

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Anil Chousein[1], Handan Duman Senol[2], Emine Ece Özdoğru[2], Sanem Eren Akarcan[2], Tuba Tuncel[3]

The clinical and laboratory findings of infants with atopic dermatitis during diagnosis and follow-up

[1]Department of Pediatrics, University of Health Sciences Tepecik Training and Research Hospital, Izmir, Turkey
[2]Department of Pediatric Allergy and Immunology, University of Health Sciences Tepecik Training and Research Hospital, Izmir, Turkey
[3]Division of Pediatric Allergy and Immunology, Department of Pediatrics, Faculty of Medicine, Izmir Katip Celebi University, Izmir, Turkey

Key words

Atopic dermatitis; food allergy; infant; prognosis.

Corresponding author
Handan Duman Şenol
Department of Pediatric Allergy and Immunology
University of Health Sciences Tepecik Training and Research Hospital
Erzene Mahallesi Ege Üniversitesi
35040 Bornova, Izmir, Turkey
ORCID: 0000-0002-7152-5053
E-mail: dumanhandan@yahoo.com

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Impact statement

Food allergy should be investigated at patients with early -onset and moderate/severe atopic dermatitis.

Summary

Background. *Atopic dermatitis is a chronic disease that affects patient and parents life worsely. What time will the patient go into remission is not known. Therefore, clinical and laboratory indicators that can indicate remission are needed.* ***Methods.*** *The study was conducted in İzmir Health Sciences University Tepecik Training and Research Hospital. The clinical and laboratory data of patients between January 2014 and December 2019 were scanned from the patient records and the hospital data system.* ***Results.*** *102 patients with a median age of 8 (min 2- max 24) months were included in the study. The median age of onset of the symptoms was 3 (min 1-max 21) months. The patients most frequently (85.2%) presented with eczema and lesions were most common (60.7%) in the extremities. Most of the patients (56.9%) had mild dermatitis. In the 6th month, 26.5% who continued follow-up, had clinical improvement. Food allergy was present in 33.3% of the patients. The most common food allergen was egg (52.9%). Food allergy was associated with the severity of atopic dermatitis ($p = 0.033$), and the symptoms started earlier ($p = 0.002$). There is no relationship between the severity of atopic dermatitis and gender, family history, presence of additional atopic disease, response to treatment, total IgE and eosinophil count ($p > 0.05$); however, it was determined that the symptoms started earlier in patients with moderate/severe atopic dermatitis ($p = 0.002$).* ***Conclusions.*** *Food allergy is more common in the early-onset and moderate/severe atopic dermatitis. Accurate diagnosis of food allergy is necessary to increase the success of treatment and to prevent unnecessary diets.*

Introduction

Atopic dermatitis (AD) is a chronic, recurrent, inflammatory skin disease that affects 2-20% of the population. Among children, nearly 50% begins in the first year of life and the frequency of the disease decreases by older age (1). More than half of the patients develop asthma and allergic rhinitis later in their life.

Atopic dermatitis etiology is complex; genetic, environmental factors, skin barrier dysfunction, and immune dysregulation take place. Both IgE mediated and non-IgE mediated food allergy can be related to AD. It is reported that most patients

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with severe disease have food allergy in epidemiologic studies (2). Immune alteration due to changes in epigenome driven by environmental exposure plays a role both in AD and food allergy (3). Prenatal (mother's nutrition, obesity, microbial exposure, *etc*.), perinatal (type of delivery, antibiotics, season of birth), and postnatal early exposure to food allergens influence the development of AD and food allergy. Diagnosis of AD is made by typical morphological distribution of the lesions and history. The Hanifin-Rajka (H-R) criteria is the most common used diagnostic criteria accepted to be the gold standard for diagnosis. Papules, papulovesicules and, eczematous plaques typically affect the face, and extensor surface of extremities in infants. The diaper area is usually spared (4).

Further investigations for food allergy should be done in the following situations according to the European Academy of Allergy and Clinical Immunology (EAACI) position paper on late eczematous reactions: a) history of an immediate reaction against foods; b) persistent, moderate-to-severe AD, no history of immediate-reactions to food, no suspected eczematous reactions to food; c) food is suspected by patients or parents as a trigger factor of persistent AD (without obvious immediate reactions) (5). Identification of the causative allergen is important to improve symptoms of AD. Cow's milk, egg, wheat and soy are the most common allergens during childhood.

Clinic and severity of AD are highly variable. It is valuable for treatment selection and monitoring the response. SCORing AD (SCORAD) index and the Eczema Area and Severity Index (EASI) are validated scoring systems, that evaluate both clinical signs and area of involvement (6, 7).

Atopic dermatitis is a chronic relapsing disease. Some of the patients go into remission in late childhood, however, in some cases the disease persists into adulthood. A recent review reported 80% of the patients' AD did not persist by 8 years old age and children younger than 2 years had less persistent disease (8). In this study, we aim to evaluate the clinical and laboratory findings of children aged 0-2 years who were diagnosed with atopic dermatitis, and to investigate the existence of a relationship between the findings at admission and follow-up findings.

Materials and methods

The study was performed at Pediatric Allergy Department of Saglik Bilimleri University Izmir Tepecik Training and Research Hospital and was approved by Saglik Bilimleri University Izmir Tepecik Training and Research Hospital Ethics Committee (date: March 23, 2022; No:2020/1-6). Parents signed an informed consent for the research study. Helsinki prtocol was followed.

Patients and study design

The hospital records of the patients with atopic dermatitis from 2016 to 2020 were analyzed retrospectively. The patients diagnosed with atopic dermatitis according to Hanifin-Rajka criteria and who were under two years old at the time of admission, without any other chronic disease were enrolled to the study. The patients with another dermatologic disease and whose follow-up results could not be reached were excluded from the study. Clinical and demographic characteristics (gender, age at onset of symptoms, age at diagnosis, history of family atopy, symptoms, duration, distribution and severity of lesions, comorbid allergic diseases, treatments, treatment responses) and laboratory data (complete blood count, immunoglobulin levels, skin prick test results, specific IgE levels, oral food challenge results) were recorded from the patient file and hospital registry system.

Laboratory tests

Results of laboratory parameters were evaluated according to the patient's age and defined as low, normal, or high. The number of eosinophils was > 500 /mm^3 or > 4% considered eosinophilia. Total serum IgE level > 165 UI/ml was defined as elevated IgE.

Skin prick tests

Skin prick testing was performed using the prick-to-prick method with cow's milk, egg white, egg yolk, wheat, soy, peanut and other suspicious foods. Saline and histamine solutions were used as the negative and positive controls, respectively. The results were evaluated after 15 min. A wheal with a diameter 3 mm greater than the negative control was taken as a positive result. Children with at least one positive skin test were considered as sensitized.

Specific IgE levels

Specific IgE (spIgE) levels were measured by the enzyme-linked immunosorbent assay (ELISA) method for the single allergen (milk, egg white, egg yolk, wheat) and/or multi allergen panel (Food panel 1 contains wheat, codfish, soybean, milk, peanut, egg white spIgE, and Food panel 2 contains almond, hazelnut, peanut, or coconut). Test results above 0.35 kIU/L were considered positive (9).

Elimination diet and oral food challenge tests

For the infants who suspected food allergy (and their mothers for breastfed infants), the elimination diet with suspected food was performed for 2-4 weeks and clinical findings of patients were observed. After the elimination diet, for IgE mediated food allergy, oral food challenge was applied in the hospital. Increasing doses of the offending food at 20 minutes interval were given with a starting dose of 0.1% of the total dose according to PRACTALL guidelines (10). For patients with non-IgE mediated food allergy, the food challenge was applied at home. For completely breast-

fed infants under six months old, elimination diet and oral food challenges were performed by changes in mothers' diet.

Statistical analysis

Statistical analyses were performed using the SPSS program version 24 ("IBM® SPSS® 24 (IBM, Armonk, NY, ABD)"). Shapiro-Wilk and Kolmogorov-Smirnov tests were used for the conformity of the data to normal distribution. The results were expressed as frequency (percentage) for categorical data and mean ± standard deviation (SD) for numerical data with normal distribution or median (minimum-maximum and interquartile range-IQR) for numerical data without normal distribution. Independent sample t-test was used to compare continuous variables in two groups with normal distribution; Mann-Whitney U tests were used to compare variables that did not have normal distribution. The P-values < 0.05 were considered statistically significant.

Results

A total number of 102 patients were enrolled in the study. The median age of the patients was 8 (min. 1, max. 24 months) and 63 (61.8%) of them were male. The median onset age of the symptoms was months 3 (min. 2, max. 21 months). Thiry-two (31.4%) of the patients had a family history of atopy and 17 (16.7%) of them had a co-morbid allergic disease (14 wheezy child, 1 proctocolitis, 1 allergic rhinitis and, 1 eosinophilic esophagitis). Most of the patients (56.8%) had mild dermatitis. Eczema was the most seen symptom at 87 (85.2%) of the patients at the first evaluation. The other symptoms were skin dryness at 68 (67.3%) and pruritus in 52 (50.9%) of the patients. Lesions were seen at extremities mostly. The characteristics of the patients were shown in **table I**.

When laboratory examinations of patients were evaluated, 47 patients (46.1%) had anemia and 20 patients (19%) had eosinophilia. Total serum IgE level was high in 49 (48%) patients. Serum IgA levels were low at 13 patients, IgG levels were low at 13 patients, and IgM levels were low at 7 patients. None of

Table I *- Characteristics of the patients.*

Characteristics	
Gender F/M	39/63
Age median (IQR) (min-max) (months)	8.0 (9.0) (2-24)
Symptom onset age median (IQR) (min-max)	3.0 (5.0) (1-21)
Family history of atopy n (%)	32 (31.4)
Mother history of atopy	18 (17.6)
Father history of atopy	10 (9.8)
Sibling history of atopy	12 (11.8)
Symptoms n (%)	
Eczema	87 (85.2)
Skin dryness	68 (67.3)
Pruritis	52 (50.9)
Mucus stool	3 (2.9)
Urticaria	2 (1.9)
Bloody stool	2 (1.9)
Lesion area n (%)	
Extremities	62 (60.7)
Trunk	41 (40.2)
Cheek	31 (30.3)
Scalp	11 (10.7)
Severity of AD n (%)	
Mild	58 (56.9)
Moderate	42 (41.2)
Severe	2 (1.9)
Comorbid atopic disease n (%)	17 (16.7)

Table II *- Laboratory findings of the patients.*

Laboratory parameters	
Hemoglobin (gr/dL) (mean ± SD)	11.3 ± 0.8
White blood cell /mm^3 (mean ± SD)	7,655 ± 662
Platelet count ± $10^3/mm^3$ (mean ± SD)	384 ± 123
Absolute neutrophil count/mm^3 (mean ± SD)	8,590 ± 739
Absolute lymphocyte count/mm^3 (mean ± SD)	5,130 ± 428
Eosinophil count/mm^3 (mean ± SD)	62 ± 73
Eosinophil (%)	5.1 ± 4.2
IgA (mg/dL) (mean ± SD)	30.7 ± 30.8
IgM (mg/dL) (mean ± SD)	68.6 ± 130.0
IgG (mg/dL) (mean ± SD)	402.7 ± 280.2
Total IgE (IU/mL) (mean ± SD)	109.8 ± 183.7
Skin Prick Test positivity n (%)	
Negative	50 (49.0)
Egg white	46 (45.1)
Egg yolk	36 (35.3)
Milk	16 (15.7)
Wheat	5 (4.2)
Soy	1 (0.8)
Peanut	3 (2.5)
Specific IgE positivity n (%)	
Egg white	29 (28.4)
Egg yolk	19 (18.6)
Milk	12 (11.7)
Wheat	4 (3.9)
Soy	1 (0.9)

Table III - *The treatments and treatment response of the patients.*

Treatment	n (%)
Diet	
No	68 (66.6)
Both mother and child	32 (31.3)
Only child	2 (1.9)
Local corticosteroid	
No	37 (36.2)
Low potency	63 (61.7)
Mediate/High potency	2 (1.9)
Moisturizer	
No	0 (0.00)
Yes	102 (100.0)
Response to treatment	
Complete	39 (38.2)
Partial	63 (61.8)

these patients were diagnosed with immune deficiency at follow-up. Fifty-two (51%) of the patients had skin test positivity with at least one of the tested allergens. The most common sensitivity was against egg white (45.1%). At least one food sensitization was detected in 41 (58.6%) of 69 patients whose sIgE was measured. Complete blood count, skin prick test, and sIgE levels of the patients were shown in **table II**.

Oral food challenge was applied to 63 (61.7%) of the patients: 11 (10.8%) at home, 52 (51%) at the hospital. Based on laboratory tests and oral food challenge results, 46 food allergy was diagnosed in 34 (33.3%) of the patients. Among food allergic patients 19 (52.9%) had an egg allergy, 12 (35.2%) had both milk and egg allergies, 2 (5.8%) had cow's milk allergy. None of the patients had peanut, wheat, or soybean allergy. A patient who had a history of urticaria with potato had positive food challenge with potato.

Moisturizers were recommended for all patients and local steroid treatment was given to 65 (63.7%) patients. None of the patients received systemic treatment. Diet was given to 34 (33.3%) food allergic patients. While treatment response was complete at 39 (38.2%) patients, it was partial at 63 (61.8%) patients. The characteristics of the treatment were shown in **table III**.

When compared the characteristics of patients with and without food allergies, in food allergic patients, atopic dermatitis was more severe (p = 0.033), and the symptoms had begun

Table IV - *The comparison of the characteristics of the patients with food allergy and without food allergy.*

	Patients without food allergy n = 68	Patients with food allergy n = 34	P-value
Gender n (%)			0.666
Female	25 (36.7)	14 (41.1)	
Male	43 (63.2)	20 (58.8)	
Family history of atopy n (%)			0.327
No	44 (64.7)	26 (76.5)	
Yes	24 (35.3)	8 (25.5)	
Comorbid atopic disease n (%)			0.851
No	57 (83.8)	28 (82.3)	
Yes	11 (16.2)	6 (17.7)	
Severity of AD n (%)			0.033
Mild	43 (63.2)	15 (44.1)	
Moderate	25 (36.8)	17 (50.0)	
Severe	0 (0)	2 (5.9)	
Response to treatment n (%)			0.666
Partial	43 (63.2)	20 (58.8)	
Complete	25 (36.8)	14 (41.2)	
Age at onset of AD (median-IQR)	5.2 (4.7)	2.6 (2.2)	0.011

Table V - *The comparison of the characteristics of the patients according to severity of atopic dermatitis.*

	Mild AD (n = 58)	**Moderate/severe AD (n = 44)**	**P-value**
Gender n (%)			0.942
Female	22 (37.9)	17 (38.2)	
Male	36 (62.0)	27 (61.8)	
Family history of atopy n (%)			0.606
No	41 (70.6)	29 (65.9)	
Yes	17 (29.3)	15 (34.1)	
Comorbid atopic disease n (%)			0.474
No	47 (81.0)	38 (86.4)	
Yes	11 (18.9)	6 (13.6)	
Allergic food n (%)			0.615
Cow's milk	1 (1.7)	1 (2.3)	
Egg	10 (17.2)	9 (21.4)	
Cow's milk and egg	4 (6.8)	6 (14.2)	
Treatment response n (%)			0.942
Partial	36 (62.0)	27 (61.4)	
Complete	22 (37.9)	17 (38.6)	
Age at onset of AD (median-IQR)	5.2 (4.2)	3.2 (2.0)	0.017

earlier (p = 0.002). There was no difference for gender, history of family and individual atopy and treatment responses, eosinophil counts and sIgE levels between allergic and nonallergic patients (p > 0.05). The results were shown in **table IV**.

Patients were grouped according to the severity of atopic dermatitis (as mild and moderate/severe) and compared for demographic data and clinical findings. There was no statistically significant difference between the groups in terms of gender, family history, presence of additional atopic disease, allergic food and treatment response (p > 0.05). In the moderate/severe AD group, the onset age of symptoms was younger, and the difference was statistically significant (p = 0.002). The results were shown in **table V**.

Clinical and laboratory findings in the 6th, 12th, 24th, and 36th months were examined to evaluate the changes in the follow-up. It was found that most patients without food allergies did not come for outpatient follow-ups. Among the patients who continued to follow up, AD severity decreased in 51 patients (27 patients in the 6th month, 17 patients in the first year, 6 patients in the second year, 1 patient in the third year). Among 14 patients with cow's milk allergy who continued to follow up, 12 (85.7%) of them recovered in the follow-up period (10 patients in the 6th month, 1 patient in the first year, 1 patient at the second year). Among 22 patients with egg allergy, 15 (68.1%) of them recovered in the follow up period (12 patients in the 6th month, 1 patient in the first year, 1 patient in the second year, and 1 patient in the third year). Also, among the patients who continued to follow up, new food sensitization occurred in 11 patients (9 patients in the 6th month, 1 patient in the first year, 1 patient at the second year). The changes in laboratory and clinical findings during follow-up period were shown in **table VI**.

Table VI - *The changes of the patient's laboratory and clinic findings during follow- up.*

	6th month (n = 53)	**12th month (n = 22)**	**24th month (n = 10)**	**36th month (n = 3)**
Regression of food allergy* n (%)	22 (61.1)	2 (5.5)	2 (5.5)	1 (3.7)
Decreased severity of atopic dermatitis n (%)	27 (50.9)	17 (32.1)	6 (11.3)	1 (1.9)
Development of new sensitization n (%)	9 (16.9)	1 (1.8)	1 (1.8)	0

*A total of 36 food allergies in 25 patients.

Discussion and conclusions

In our study, it was found that mild AD was the most seen pattern in our patients. AD was accompanied by food allergy in 1/3 of the patients, and egg allergy was the most common food allergen. Also, atopic dermatitis had started earlier and was more severe in patients with food allergies. During follow-up, half of the patients had both food allergies and atopic dermatitis findings regressed in the first 6 months, while almost all had recovered at the age of three.

The median age at onset of symptoms was less than 6 months in our study, which is consistent with the literature. Kay *et al*. reported that 60% of 1,104 patients developed the disease younger than 1 year and 3/4 of them were younger than 6 months (11). It was seen that the patients' admissions were 4 months after the onset of the symptoms on average. It was thought that this period between the onset of the disease and the admission of the patients to our outpatient clinic was related to the fact that the patients had to apply to the pediatrician first, the first examination and treatments were started, and the time taken to make an appointment.

Family history of atopy was found in 1/3 of our patients and nearly half of them with maternal history. In a cohort of 4,089 children from Stockholm, AD was detected in 39% of those with AD in one parent, in 50% of those with AD in both parents, and 27% of those without atopic history (12). In a prospective study from Sweden, the incidence rate of AD in any parent of the patients was 58%, and atopy in any parent was 69% (13). Laske *et al*. reported a positive family history for atopic diseases was 60% (14). As it is known that genetic predisposition plays role in the development of allergic diseases, consistent with literature we found the presence of atopy in the family was common in patients with AD.

Weidinger *et al*. reported EASI and SCORAD as the most performed tests in 32 international AD centers with accurate AD severity (15). It was observed that more than half of the patients in our study had mild AD and almost none of the patients had severe AD. Laske *et al*. reported that mild and severe disease were detected in 60% and 54% of boys, 40% and 46% of girls, respectively (14). Roehr *et al*. who used a modified scoring system from Hanifin and Rajka, reported that 62% of the patients had mild, 27% had moderate, and 10% had severe disease (16). Although almost all our patients had mild and moderate atopic dermatitis, which is consistent with the literature, it was noteworthy that the rate of severe disease was much lower than in the literature. In our center, which is a 3rd level hospital, the reason for the low number of patients was that the time between the onset of the complaints of the patients and the admission was long (almost 4 months), and the complaints had partially regressed with the treatments applied during this time.

In our study, more than half of our patients had positive skin prick test and sIgE levels for food. Similar to our results, in a 7-year cohort study conducted in Sweden, 61% of patients younger than 36 months were found to be positive for specific IgE (f × 5 food group) (13).

Double-blind placebo-controlled food challenges (DBPCFCs) are the gold standard tests for diagnosing food allergy. Because our children were under two years old, we used open oral food challenges (OFC). We found that approximately 1/3 of the patients with positive skin test did not have a food allergy and the results of the skin test were compatible with the sensitivity. In a study involving 88 patients with AD in children aged 3 months to 7 years in Greece, food sensitivity was found in 39 patients, and 23 of these patients were diagnosed with food allergy (17). It is known that although the negative predictive value of skin prick test and sIgE is high as 90-95%, its positive predictive value is low (50%) (18). Rowlands *et al*. performed 91 food challenge tests to 19 children with severe AD refractory to multiple therapies. They found a rare relation between foods and eczema and emphasized that diagnosis of food allergy inducing eczema cannot be said without food challenge testing (19). Compatible with the literature, our results are important to show that skin test or sIgE positivity for food is not sufficient for the diagnosis of food allergy and a food challenge test is required.

In our study 1/3 of all AD patients had a diagnosis of food allergy. In a study conducted in South Korea in 2013, the frequency of food allergy in infants with AD was reported to be 37.1% in 0-5-month-old infants and 38.5% in 6-11-month-old infants (20). Mavroudi *et al*. reported that in children aged three months to seven years, food allergy was found in 26% of AD patients (17). The presence of food allergy in our patients with AD was found to be compatible with studies conducted in other countries. In our study, more than half of the patients with food allergy were found to be allergic to egg white and yolk. This was followed by milk allergy in 1/3 of the patients and more than half of the patients had both milk and egg allergies. None of our patients had sensitization to soy, peanut, or wheat. Mavroudi *et al*. reported milk (80%) had the most common sensitization rate in their study, followed by the egg (55%), wheat (20%), cod, and soy (2.5%) (17). Various food sensitizations from various countries were reported according to complementary feeding sources and habit (21, 22). In our country egg and milk are the most common used foods during infancy, so we saw more sensitization with these foods.

When the clinical findings of patients with and without food allergy were evaluated, we found that AD started earlier and more severe in patients with food allergy. In a study by Şengül *et al*., it was reported that the SCORAD index and total IgE levels were higher in children with food sensitivity younger than three months of age (23). In a study conducted in Thessaloniki, it was reported that 47% of patients with mild-to-moderate

AD and 39% of patients with severe AD had a food allergy (17). A population-based study evaluating 619 children aged 3 months, reported that patients with SCORAD index greater than 20, had more association between food sensitization and AD (OR 25.60; 95%CI 9.03-72.57; p < 0 .001) (24).

In our study, it was found that the clinical findings started earlier in the group with severe AD. In an international study conducted 12 countries, (n = 2,048, age range 11.5-25.5 months, mean 17.6 months), a statistically significant relationship was found between early-onset AD and the severity of clinical severity (p < 0.001) (25). Cansever *et al*. also reported that early age of onset was one of the risk factors for the severity of AD (26).

In the treatment of AD, multifaceted, stepwise approach consisting daily shower, moisturizers, avoidance of triggers as irritants and food allergens (for allergic patients) is advised in the literature. Moisturizers were started for all our patients, more than half of them with topical steroids, and all those with food allergies were on a diet. It was observed that all the patients responded to the treatment, but in most the response was partial. In AD patients younger than 12 months of age, significant regression was reported in the lesions of patients who used both moisturizing and topical steroids in the sixth week of treatment (24). In a study conducted with 110 children, it was reported that symptoms regressed in all patients treated with diet (23). Due to the multifactorial nature of the disease, it was thought that all our patients did not respond fully to the treatment.

Our study reveals the data of our region. The most important limitation of this study is its' retrospective design. The lack of a standard follow-up form and the deficiencies in the data led to the low number of patients included in the study. We could not get data especially about prognosis and new atopic disease development.

In conclusion, the presence of skin test and/or sIgE positivity in AD patients does not mean food allergy, and the diagnosis should be made by elimination diet and oral food challenge test. It should not be forgotten that food allergy is more common especially in the early-onset and severe patient group. Accurate diagnosis of food allergy is necessary to increase the success of treatment and to prevent unnecessary diets. The regression of clinical findings in most patients within the first year is also an important finding in terms of prognosis.

Fundings

None.

Contributions

TT, AC: conceptualization. HDS, EEÖ, TT: data curation. AI, TT: formal analysis. HDS, TT: writing – original draft, writing – review & editing. SEA: data curation, formal analysis, writing – review & editing.

Conflict of interests

The authors declare that they have no conflict of interests.

References

1. Illi S, von Mutius E, Lau S, Nickel R, Grüber C, Niggemann B, et al. The natural course of atopic dermatitis from birth to age 7 years and the association with asthma. J Allergy Clin Immunol. 2004;113(5):925-31. doi: 10.1016/j.jaci.2004.01.778.
2. Spergel JM, Paller AS. Atopic dermatitis and the atopic march. J Allergy Clin Immunol. 2003;112(6 Suppl):S118-27. doi: 10.1016/j.jaci.2003.09.033.
3. Lockett GA, Huoman J, Holloway JW. Does allergy begin in utero? Pediatr Allergy Immunol. 2015;26(5):394-402. doi: 10.1111/pai.12408.
4. Yew YW, Thyssen JP, Silverberg JI. A systematic review and meta-analysis of the regional and age-related differences in atopic dermatitis clinical characteristics. J Am Acad Dermatol. 2019;80(2):390-401. doi: 10.1016/j.jaad.2018.09.035.
5. Werfel T, Ballmer-Weber B, Eigenmann PA, Niggemann B, Rancé F, Turjanmaa K, et al. Eczematous reactions to food in atopic eczema: position paper of the EAACI and GA2LEN. Allergy. 2007;62(7):723-8. doi: 10.1111/j.1398-9995.2007.01429.x.
6. Severity scoring of atopic dermatitis: the SCORAD index. Consensus Report of the European Task Force on Atopic Dermatitis. Dermatology. 1993;186(1):23-31. doi: 10.1159/000247298.
7. Hanifin JM, Thurston M, Omoto M, Cherill R, Tofte SJ, Graeber M. The eczema area and severity index (EASI): assessment of reliability in atopic dermatitis. EASI Evaluator Group. Exp Dermatol. 2001;10(1):11-8. doi: 10.1034/j.1600-0625.2001.100102.x.
8. Kim JP, Chao LX, Simpson EL, Silverberg JI. Persistence of atopic dermatitis (AD): A systematic review and meta-analysis. J Am Acad Dermatol. 2016;75(4):681-7.e11. doi: 10.1016/j.jaad.2016.05.028.
9. Eigenmann PA, Oh JW, Beyer K. Diagnostic testing in the evaluation of food allergy. Pediatr Clin North Am. 2011;58(2):351-62. doi: 10.1016/j.pcl.2011.02.003.
10. Sampson HA, Gerth van Wijk R, Bindslev-Jensen C, Sicherer S, Teuber SS, Burks AW, et al. Standardizing double-blind, placebo-controlled oral food challenges: American Academy of Allergy, Asthma & Immunology-European Academy of Allergy and Clinical Immunology PRACTALL consensus report. J Allergy Clin Immunol. 2012;130(6):1260-74. doi: 10.1016/j.jaci.2012.10.017.
11. Kay J, Gawkrodger DJ, Mortimer MJ, Jaron AG. The prevalence of childhood atopic eczema in a general population. Journal of the American Academy of Dermatology. 1994;30(1):35-9. doi: 10.1016/s0190-9622(94)70004-4.
12. Böhme M, Wickman M, Lennart Nordvall S, Svartengren M, Wahlgren CF. Family history and risk of atopic dermatitis in children up to 4 years. Clin Exp Allergy. 2003;33(9):1226-31. doi: 10.1046/j.1365-2222.2003.01749.x.
13. Gustafsson D, Sjöberg O, Foucard T. Development of allergies and asthma in infants and young children with atopic

dermatitis--a prospective follow-up to 7 years of age. Allergy. 2000;55(3):240-5. doi: 10.1034/j.1398-9995.2000.00391.x.
14. Laske N, Niggemann B. Does the severity of atopic dermatitis correlate with serum IgE levels? Pediatr Allergy Immunol. 2004;15(1):86-8. doi: 10.1046/j.0905-6157.2003.00106.x.
15. Weidinger S, Nosbaum A, Simpson E, Guttman E. Good practice intervention for clinical assessment and diagnosis of atopic dermatitis: Findings from the atopic dermatitis quality of care initiative. Dermatol Ther. 2022;35(3):e15259. doi: 10.1111/dth.15259.
16. Roehr CC, Reibel S, Ziegert M, Sommerfeld C, Wahn U, Niggemann B. Atopy patch tests, together with determination of specific IgE levels, reduce the need for oral food challenges in children with atopic dermatitis. J Allergy Clin Immunol. 2001;107(3):548-53. doi: 10.1067/mai.2001.112849.
17. Mavroudi A, Karagiannidou A, Xinias I, Cassimos D, Karantaglis N, Farmaki E, et al. Assessment of IgE-mediated food allergies in children with atopic dermatitis. Allergol Immunopathol (Madr). 2017;45(1):77-81. doi: 10.1016/j.aller.2016.06.006.
18. Sampson HA. The evaluation and management of food allergy in atopic dermatitis. Clin Dermatol. 2003;21(3):183-92. doi: 10.1016/s0738-081x(02)00363-2.
19. Rowlands D, Tofte SJ, Hanifin JM. Does food allergy cause atopic dermatitis? Food challenge testing to dissociate eczematous from immediate reactions. Dermatol Ther. 2006;19(2):97-103. doi: 10.1111/j.1529-8019.2006.00063.x.
20. Kwon J, Kim J, Cho S, Noh G, Lee SS. Characterization of food allergies in patients with atopic dermatitis. Nutr Res Pract. 2013;7(2):115-21. doi: 10.4162/nrp.2013.7.2.115.
21. Gustafsson D, Sjöberg O, Foucard T. Sensitization to food and airborne allergens in children with atopic dermatitis followed up to 7 years of age. Pediatr Allergy Immunol. 2003;14(6):448-452. doi: 10.1046/j.0905-6157.2003.00093.x.
22. Jarmila C, Květuše E, Karel E, Jaroslava V, Josef B. Soy allergy in patients suffering from atopic dermatitis. Indian J Dermatol. 2013;58(4):325. doi: 10.4103/0019-5154.113938.
23. Şengül Emeksiz Z, Cavkaytar Ö, Aksoy İ, Dallar Y, Soyer Ö. Food Hypersensitivity in Atopic Dermatitis During Infancy: Skin Prick Testing for Whom? Asthma Allergy Immunol. 2017;15:32-7. doi: 10.21911/aai.16.
24. Flohr C, Perkin M, Logan K, Marrs T, Radulovic S, Campbell LE, et al. Atopic Dermatitis and Disease Severity Are the Main Risk Factors for Food Sensitization in Exclusively Breastfed Infants. Journal of Investigative Dermatology. 2014;134(2):345-50. doi: 10.1038/jid.2013.298.
25. Hill DJ, Hosking CS, de Benedictis FM, Oranje AP, Diepgen TL, Bauchau V, et al. Confirmation of the association between high levels of immunoglobulin E food sensitization and eczema in infancy: an international study. Clin Exp Allergy. 2008;38(1):161-8. doi: 10.1111/j.1365-2222.2007.02861.x.
26. Cansever M, Oruç Ç. What plays a role in the severity of atopic dermatitis in children? Turk J Med Sci. 2021;51(5):2494-501. doi: 10.3906/sag-2101-194.

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Laura Romero-Sánchez[1], Andrea Otero[1], María González-Rivas[1], Santiago Lojo[2], Arturo González-Quintela[3], Carmen Vidal[1]

Der p 23 sensitization in patients with house dust mite respiratory allergy

[1]Department of Allergy, Complejo Hospitalario Universitario de Santiago, Faculty of Medicine, University of Santiago de Compostela, Santiago de Compostela, Spain
[2]Department of Biochemistry, Complejo Hospitalario Universitario de Santiago, Santiago de Compostela, Spain
[3]Department of Medicine, Complejo Hospitalario Universitario de Santiago, Faculty of Medicine, University of Santiago de Compostela, Santiago de Compostela, Spain

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Corresponding author
Laura Romero-Sánchez
Department of Allergy
Complejo Hospitalario Universitario de Santiago
Faculty of Medicine
University of Santiago
Rúa Ramón Baltar sn
15706 Santiago de Compostela, Spain
ORCID: 0000-0002-9586-1174
E-mail: laura.romero.sanchez@sergas.es

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Impact statement
Der p 23 was the predominant allergen with a prevalence of sensitization of 83.7%. Asthma was more frequent in patients with positive sIgE against Der p 23.

Summary
Background. *The sensitization profile of patients allergic to house dust mites (HDM) and its molecular diagnosis may determine treatment and evolution of the disease. The present study investigates the prevalence of Der p 23 sensitization and its relation to asthma in a population of HDM-allergic patients.* ***Methods.*** *We conducted a cross-sectional study of 891 patients with HDM allergy with symptoms of rhinitis and 52.1% of them with asthma. Total and specific IgE (sIgE) was measured against Dermatophagoides pteronyssinus and its molecular components and the storage mite Lepidoglyphus destructor using ImmunoCAP. Prevalence of sensitization and levels of sIgE were analyzed according to asthma diagnosis and asthma severity.* ***Results.*** *Der p 23 was the predominant allergen in this population (83.7%), but IgE levels were lower than those of sIgE to Der p 1 and Der p 2. A good correlation was found between sIgE to Der p 23 and the other allergens. A total of 8.2% patients were monosensitized to Der p 23. Asthma was more frequent in patients with positive sIgE against Der p 23 than in patients without this sensitization (p = 0.027). A tendency to increase both total IgE and sIgE was observed in relation to the severity of asthma from intermittent mild asthma to persistent moderate asthma, but a substantial decrease in total IgE and sIgE was detected in more severe asthmatics.* ***Conclusions.*** *Der p 23 might be a prevalent allergen in regions with high rates of HDM exposure and its presence could increase the risk of asthma.*

Introduction

House dust mite (HDM) allergy is one of the most prevalent causes of respiratory allergy. Its clinical manifestations are mainly rhinitis, conjunctivitis and asthma. *Dermatophagoides pteronyssinus* (*D. pteronyssinus*) is the most common cause of HDM allergy worldwide (1). Up to 35 different *D. pteronyssinus* allergens have been described (2), being Der p 1 and Der p 2 the most studied allergens. Der p 23 was identified in 2014 by Weghofer *et al.* as a peritrophin-like protein (3), and from that moment onwards many studies have focused their attention on this allergen and its role in *Dermatophagoides* species sensitization. In the first description of this allergen, up to 74% of patients with *D. pteronyssinus* sensitization presented specific IgE (sIgE) against it (3).

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HDMs are the main cause of respiratory allergic diseases in some regions in Spain, namely in the Northwest (4, 5) and the Canary Island (6, 7) where weather conditions favor its development. The three major allergens of *D. pteronyssinus*, Der p 1, Der p 2 and Der p 23 (2, 3) have been found to be associated with severity of asthma (4, 8) and a sensitization to Der p 23 with a higher prevalence of symptoms of allergic rhinitis (9). In addition to *Dermatophagoides*, other HDM also known as storage mites such as *Lepidoglyphus destructor*, *Tyrophagus putrescentiae*, *Acarus siro*, and *Blomia tropicalis*, among others, are common sensitizers in these regions (4-7, 10).

The aim of the present study was to investigate in a large sample of patients with perennial respiratory allergic disease and a positive skin prick test to *D. pteronyssinus*, the prevalence of sensitization against Der p 1, Der p 2 and Der p 23, and their relationship with clinical parameters such as the presence of asthma and the severity of the disease in the Northwest region of Spain. We also intended to determine the frequency of sensitization to *L. destructor,* a relevant HDM in our region.

Patients and methods

We conducted a cross-sectional study of HDM-allergic patients evaluated at the Department of Allergy of Complejo Hospitalario Universitario de Santiago de Compostela, Spain, between January 2019 and December 2020. The hospital covers an area of approximately 500,000 inhabitants, most of them living in a rural environment. The study was approved by the Institutional Ethics Committee (code 2018/434) and complied with the recommendations of the Declaration of Helsinki. The patients had been referred either from primary care physicians or other specialists of our Hospital (mainly, Ear Nose and Throat physicians) for evaluation of respiratory symptoms. Patients, both adults and children, with a positive skin prick test against *D. pteronyssinus* and symptoms related to indoor exposure were included. None of these patients were previously under immunotherapy with HDM allergens.

Study procedures

Skin prick tests with *Phleum pratense*, *Phragmites communis*, *Cynodon dactylon*, *Plantago lanceolata*, *Parietaria judaica*, *Artemisia vulgaris*, *Betula alba*, *Alnus glutinosa*, *Olea europea*, *Fraxinus excelsior*, *Quercus robur*, *Platanus acerifolia*, *Cupressus sempervirens*, Profilin, Polcalcin, LTP, Latex, *Lepidoglyphus destructor*, *Tyrophagus putrescentiae*, *Alternaria alternata*, *Aspergillus fumigatus*, dog dander and cat dander were routinely performed (ALK-Abelló, Madrid, Spain). Histamine was used as a positive control and saline as a negative control.

Asthma was diagnosed based on Guía Española para el Manejo del Asma (GEMA) 4.4 guidelines and an increase in forced exhaled volume in 1 second (FEV_1) greater than 12% and 200 mL after 4 successive 100-µg puffs of a salbutamol inhaler with a spacer was required in patients with typical symptoms. Asthma severity was reclassified according to GEMA 5.0 (11) and GINA (12) before performing the statistical analysis. For the diagnosis of allergic rhinitis, we followed allergic rhinitis and its impact on asthma (ARIA) (13) recommendations. Spirometry was performed in all patients with asthma, but also in some patients with rhinitis to rule out asthma.

Total serum IgE was measured using latex-enhanced nephelometry in a BN-II System analyzer (Siemens). Allergen sIgE was measured using the ImmunoCAP-250 system (Thermo Fisher Scientific) and included sIgE against *D. pteronyssinus*, nDer p 1, rDer p 2, and rDer p 23. *L. destructor* sIgE was measured in 807 patients with a positive SPT result. Following the manufacturer's instructions, sIgE levels ≥ 0.1 kU_A/L were deemed positive, although for most analyses the classic 0.35 kU_A/L threshold level was used.

Statistical analysis

Non-parametric statistics were used due to the non-normal distribution of numerical variables. The Mann-Whitney test to compare numerical variables between pairs of independent groups. The Jonckheere-Terpstra test was used to analyze trend between ordinal independent variables and numerical depen-

Table I *- Factors associated with severity of asthma in house durst mite-allergic patients.*

Factor	No asthma (n = 435)	Asthma (n = 456)				P-value (trend)
		Grade 1 (n = 172)	Grade 2 (n = 93)	Grade 3 (n = 153)	Grade 4 (n = 38)	
Age (years)	27 (19-38)	27 (19-37)	31 (20-41)	31 (22-40)	40 (29-44)	< 0.001
Sex (male)	202 (46.4)	65 (37.8)	44 (47.3)	62 (40.5)	14 (36.8)	0.174
Smoking (current)	54 (12.5)	24 (14.0)	18 (19.4)	23 (15.0)	6 (15.8)	0.209
Habitat (rural)	254 (58.4)	98 (57.0)	55 (59.1)	82 (56.3)	21 (55.3)	0.381

Age data are medians and interquartile ranges (within parentheses). The remaining data are absolute numbers and percentages (within parentheses). Data about smoking was unavailable for two patients.

Table II *- Specific IgE to mites and their allergen components, and total serum IgE in house dust mite-allergic patients.*

Factor	Whole sample (n = 891)	Age			Sex			Habitat		
		< 18 y (n = 157)	≥ 18 y (n = 734)	P-value	Female (n = 504)	Male (n = 387)	P-value	Urban (n = 381)	Rural (n = 510)	P-value
sIgE to D1, kU_A/L	21.7 (5.65-53.6)	38.8 (10.8-78.5)	19.2 (5.21-47.4)	< 0.001	19.6 (5.22-52.2)	25.2 (6.12-55.3)	0.159	21.0 (5.42-52.6)	22.4 (5.79-55.0)	0.724
sIgE to Der p 1, kU_A/L	5.19 (0.10-17.9)	12.1 (0.05-29.8)	4.86 (0.13-15.3)	0.002	4.89 (0.06-17.9)	5.74 (0.15-17.6)	0.368	5.68 (0.53-17.9)	4.90 (0.04-17.8)	0.187
sIgE to Der p 2, kU_A/L	7.82 (0.99-21.4)	15.8 (1.55-33.6)	6.81 (0.90-19.3)	< 0.001	6.96 (0.70-20.6)	8.81 (1.33-24.0)	0.068	7.15 (0.86-19.5)	8.23 (1.21-23.3)	0.270
sIgE to Der p 23, kU_A/L	3.19 (0.94-7.45)	6.10 (2.46-11.6)	2.86 (0.81-6.40)	< 0.001	3.10 (0.88-7.35)	3.36 (1.04-7.93)	0.421	3.11 (0.93-7.14)	3.21 (0.95-7.91)	0.509
sIgE to D71, kU_A/L	2.73 (0.54-12.2)	2.40 (0.63-10.6)	2.74 (0.52-12.4)	0.800	2.73 (0.46-11.8)	2.71 (0.67-12.7)	0.381	2.06 (0.45-8.50)	4.16 (0.64-16.9)	<0.001
Total IgE, kU/L	198 (83-447)	341 (143-784)	179 (77-400)	< 0.001	180 (76-383)	240 (100-531)	<0.001	169 (72-404)	222 (95-469)	0.019

sIgE: specific IgE. Figures are shown as median and (IQR). Cases with sIgE above the analytical limit (100 kU_A/L) were deemed to have 100 kU_A/L. sIgE to d1, Der p 1, Der p 2, d71, and total IgE were unavailable for 9, 3, 6, 84, and 1 patient, respectively.

dent variables. The Spearman's rank test was used to assess correlations. The chi-square test (with trend analysis, where appropriate) was used to compare proportions. P-values lower than 0.05 were deemed statistically significant.

Results

A total of 891 consecutive patients with HDM allergic rhinitis and/or asthma were included in the study (median age: 29 years (range, 4-69 years); 43.4% male). Of note, 607 patients presented positive skin prick test results to other sensitizations such as pollens, epithelia, fungi and/or food (68.1%), but symptoms were not directly related to them.

Patients with asthma (456/891, 51.2%) were divided into mild intermittent (172, 37.7%), mild persistent (93, 20.4%), moderate (153, 33.5%), and severe (38, 8.3%) asthma. Only 125 patients (14.0%) were current smokers. Gender, current smoking, or exposure to rural environment were not related to asthma severity. However, older patients were more frequently diagnosed with severe asthma (**table I**).

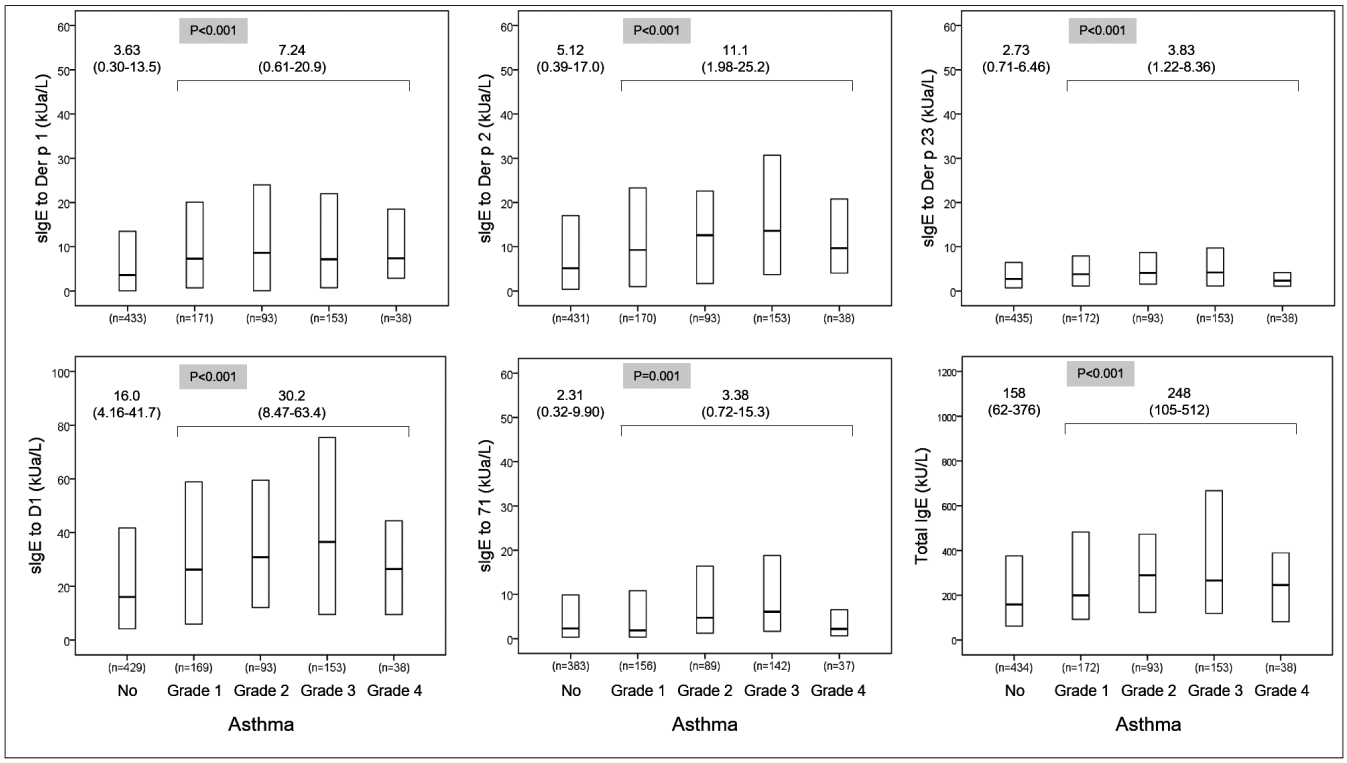
Prevalence of sensitization and levels of specific sIgE in the studied population

Specific IgE to *D. pteronyssinus* was detected in most patients (850/882 samples available, 96.4%). The percentage of patients with sIgE to Der p 23 was higher (746/891, 83.7%) than that of those reacting to Der p 2 (698/885, 78.9%) and Der p 1 (639/888, 72.0%). However, sIgE levels to Der p 2 and Der p 1 were higher than those to Der p 23 (**table II**). Additionally, 79.3% (640/807) of the patients presented sIgE to *L. destructor*, confirming the high prevalence of this HDM in the region. Of the patients studied with sIgE to *D. pteronyssinus* and its molecular components, 73/882 were monosensitive to Der p 23, which represents 8.2% of the sample.

Regarding age, levels of sIgE to *D. pteronyssinus* and its molecular components (Der p 1, Der p 2, and Der p 23) were significantly higher among younger patients (157 children and/or adolescents < 18 years old) than adults (**table II**), following a similar pattern than total IgE (median 341 UI/mL (143-784 UI/mL) *vs* median 179 UI/mL (77-400 UI/mL)), $p < 0.001$. In contrast, no differences were found on sIgE to *L. destructor* between older and younger patients (**table II**). Finally, the levels of sIgE to *L. destructor* were higher among patients living in rural areas ($p < 0.001$), but this habitat seemed not to influence sIgE levels to *D. pteronyssinus* or its molecular components (**table II**).

Der p 23 monosensitization seemed not to be related to age, gender, smoking habit, alcohol intake, concomitant asthma or living in a rural environment (data not shown). Patients monosensitized to Der p 23 presented lower levels of both total IgE and sIgE to *D. pteronyssinus* than patients with oth-

***Figure 1** - Serum concentrations of serum specific IgEs and total IgE in patients with house dust mite allergy, stratified by the presence of asthma and asthma severity.*



Both bars and numbers represent medians and interquartile ranges. All specific IgE and total IgE are higher in patients with asthma than in patients without asthma (P-values in the figure reflect the comparison between asthmatics and non-asthmatics). Within patients with asthma, specific IgE and total IgE tend to increase with asthma severity to some extent − from severity 1 (mild intermittent) to severity 3 (moderate) −, especially Der p 2 (p = 0.012), D1 (p = 0.041), L. destructor (p < 0.001) and total IgE (p = 0.032). However, in the most severe cases of asthma, all specific IgE tended to stabilize or decrease.

er *D. pteronyssinus* IgE sensitizations (123 (48-271) UI/mL *vs* 210 (83-463), p = 0.001, for total IgE and 1.84 (0.76-4.54) kUa/L *vs* 25.8 (8.36-56.9), p < 0.001 for *D. pteronyssinus*, respectively).

A significant correlation was found between specific sIgE to Der p 23 and sIgE to *D. pteronyssinus* (ρ = 0.769, p < 0.001), Der p 1 (ρ = 0.644, p < 0.001), and total sIgE (ρ = 0.628, p < 0.001). A lower but significant correlation was found between sIgE to Der p 23 and Der p 2 (ρ = 0.593, p < 0.001) and *L. destructor* (ρ = 0.345, p < 0.001).

Total and specific sIgE in patients with asthma

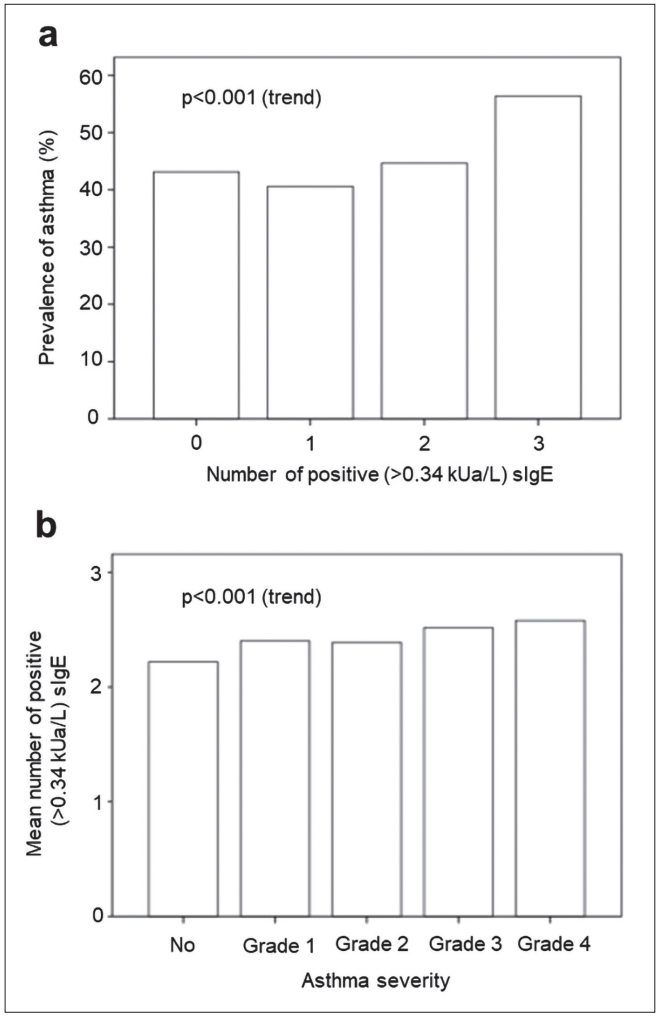
Levels of total IgE and sIgE to *D. pteronyssinus*, Der p 1, Der p 2, Der p 23, and *L. destructor* allergens were significantly higher in patients with asthma than in patients with only rhinitis (**figure 1**). A tendency to increase both total IgE and sIgE was observed in relation to the severity of asthma from intermittent mild asthma to persistent moderate asthma. However, a substantial decrease in total IgE and sIgE was detected in more severe asthmatics (**figure 1**).

The risk of asthma showed a trend to be higher in patients with more positive sIgE results. Thus, the more positive sIgE determinations, the higher risk for suffering from asthma (**figure 2a**). Along this line, asthma was more frequently diagnosed in patients with positive sIgE against Der p 23 than in patients without Der p 23 sensitization (52.8% *vs* 42.8%, p = 0.027). A similar trend was seen in relation to the severity of asthma (p < 0.001) (**figure 2b**). Finally, a forced spirometry was performed in 495 patients (456 asthmatic patients and 39 with rhinitis) but no differences in FEV1 values or in FEV1/FVC were observed depending on the *D. pteronyssinus* sensitization profile (data no shown).

Discussion and conclusions

House dust mites (HDM) are a major cause of respiratory allergy and of perennial asthma worldwide. Der p 23, a gut-derived peritrophin present in the outer membrane of mite feces, has been

Figure 2 *- Patients in whom all three sIgE determinations against D. pteronyssinus allergens (Der p 1, Der p 2 and Der p 23) were available.*



(a) *The mean number of positivities (sIgE > 0.35 kUA/L) against these allergens tended to increase the risk of asthma;* ***(b)*** *The mean number of positivities (sIgE > 0.35 kUA/L) against these allergens tended to increase with asthma severity: 2.22 in non-asthmatics (n = 430), 2.41 in grade 1 asthmatics (n = 170), 2.38 in grade 2 asthmatics (n = 93), 2.51 in grade 3 asthmatics (n = 153), and 2.57 in grade 4 asthmatics (n= 38) (p < 0.001 in the trend test).*

recognized as a major allergen (3, 8, 15, 16, 17-20). Even though sIgE-array platforms have included a wide spectrum of HDM allergens (8, 14), only Der p 1, Der p 2, Der p 23, and Der p 10 (tropomyosin) are currently available for the component-resolved diagnosis of HDM allergy on ImmunoCAP. Our results, using InmunoCAP in the largest sample published to date, are similar to those previously reported in terms of prevalence of sensitization (6, 8, 14, 15, 17), but we report a high rate of monosensitization to Der p 23 (6, 8, 15-18, 20, 21). The clinical relevance of Der p 23 is only partially defined, and the prevalence and relevance of the exclusive sensitization to it has received little attention so far. In this population, patients monosensitized to Der p 23 were similar to the others with respect to age, gender, smoking habit, rural or urban environment and prevalence of rhinitis or asthma. However, when Der p 23 sensitization accompanies other *D. pteronyssinus* allergen sensitizations, the risk of asthma seems to increase. All our patients were primarily diagnosed by means of a positive SPT to *D. pteronyssinus* and most of them were confirmed by a positive serum sIgE to *D. pteronyssinus* whole extract. No positive responses to Der p 23 were found in patients with a negative serum sIgE to *D. pteronyssinus* but these patients have lower levels of total IgE and sIgE to *D. pteronyssinus*.

The fact that sIgE levels decline with age have been previously reported (6, 21). In this population, total and sIgE levels to *D. pteronyssinus*, Der p 1, Der p 2 and Der p 23 were also lower in adults compared with children and adolescents. Specific IgE to *L. destructor* seems to follow a different pattern since it did not vary according to age. Mean specific IgE levels to Der p 2 and Der p 1 allergens were higher than those elicited by Der p 23 in terms of quantity in all group ages, but with a close correlation among them and the crude extract of *D. pteronyssinus*.

Considering the clinical expression of the allergic disease (rhinitis with or without asthma), the risk of suffering from asthma has a tendency to increase with the number of allergens recognized by patients which is in accordance with previous results (9, 18, 22). Letran *et al.* recently found no relationship between Der p 23 sensitization and asthma. Furthermore, they showed a lower prevalence of asthma when Der p 23 sensitization was present (19). In our sample, asthma was more frequently diagnosed in patients with positive sIgE against Der p 23 than in patients without Der p 23 sensitization, giving Der p 23 a role in asthma development when Der p 23 sensitization accompanies Der p 1 and Der p 2. A similar tendency was shown in relation to the severity of asthma. However, when sIgE levels are analyzed, the interpretation changes. While higher levels of total IgE and sIgE to all allergens may be detected in patients with mild intermittent asthma to moderate asthma, a significant drop in all these IgE determinations was detected at the most severe asthma level. It is true that more severe asthmatics were also the eldest, but the small size of this group cannot allow to be sure if the effect of this drop in IgE is due to age or the severity of asthma. No similar findings have been reported elsewhere. Severity of asthma could probably be related to bronchial remodeling or pathology where allergy is less relevant, but more studies are needed to define this cause.

Although the large sample of patients described in our study make our results robust, it has some limitations that we would like to highlight. Firstly, we are based on retrospective data, so we cannot draw predictive conclusions because we do not have follow-up information. Second, pulmonary function parameters were performed only in those patients who reported bronchial symptoms, and some patients who only reported rhinitis may

have been underdiagnosed. Thirdly, we determined specific IgE to Der p 1, Der p 2 and Der p 23, so the monosensitive concept is exclusively applied to these three allergens. Fourthly, the nasal provocation tests were not performed in those patients who had symptoms with exposure to HDMs and had negative SPT, so that a certain number of patients with local allergic rhinitis could be excluded from our study. And finally, we have taken 0.35 kU/L as the cut-off point for specific IgE positivity. The median sIgE level at Der p 23 in our monosensitive patients was 0.27, so these patients were not detected with the cut-off point used.

In conclusion, our study confirms the high prevalence of sensitization to Der p23 and its clinical implication in asthma. This sensitization could be of clinical relevance as patients who recognize sIgE to Der p 23 present asthma more frequently than those who do not. Specific IgE levels to all allergens increase with asthma severity from mild intermittent to moderate asthma but a drop may be detected in the most severe grade of the disease.

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Contributions

LR-S: writing – original draft, data curation, software. AO, MG-R: data curation, software. AG-Q: formal analysis. SL: investigation. CV: project administration, writing – original draft, supervision. All authors: writing – review & editing.

Conflict of interests

The authors declare that they have no conflict of interests.

Data availability

The datasets generated and/or analyzed during the current study are available in the RUNA Repositorio de Saúde repository: http://hdl.handle.net/20.500.11940/16141.

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References

1. Calderón MA, Linneberg A, Kleine-Tebbe J, De Blay F, Hernandez Fernandez De Rojas D, Virchow JC, Demoly P. Respiratory allergy caused by house dust mites: What do we really know? J Allergy Clin Immunol. 2015;136(1):38-48. doi: 10.1016/j.jaci.2014.10.012.
2. Liu XY, Yang KY, Wang MQ, Kwok JS, Zeng X, Yang Z, et al. High-quality assembly of Dermatophagoides pteronyssinus genome and transcriptome reveals a wide range of novel allergens. J Allergy Clin Immunol. 2018;141(6):2268-71.e8. doi: 10.1016/j.jaci.2017.11.038.
3. Weghofer M, Grote M, Resch Y, Casset A, Kneidinger M, Kopec J, et al. Identification of Der p 23, a peritrophin-like protein, as a new major Dermatophagoides pteronyssinus allergen associated with the peritrophic matrix of mite fecal pellets. J Immunol. 2013;190(7):3059-67. doi: 10.4049/jimmunol.1202288.
4. Vidal C, Lojo S, Juangorena M, Gonzalez-Quintela A. Association between asthma and sensitization to allergens of Dermatophagoides pteronyssinus. J Investig Allergol Clin Immunol. 2016;26(5):304-9. doi: 10.18176/jiaci.0048.
5. Vidal C, Boquete O, Gude F, Rey J, Meijide LM, Fernández-Merino MC, et al. High prevalence of storage mite sensitization in a general adult population. Allergy. 2004;59(4):401-5. doi: 10.1111/j.1398-9995.2003.00430.x.
6. González-Pérez R, Pineda F, Poza-Guedes P, Castillo M, Matheu V, Sánchez-Machín I. Molecular allergen profiling of dual mite sensitization in severe allergic rhinitis. J Investig Allergol Clin Immunol. 2020;30(6):421-9. doi: 10.18176/jiaci.0439.
7. González-Pérez R, Poza-Guedes P, Pineda F, Forstenlechner P, Castillo M, Mederos-Luís E, et al. Mite molecular profile in the Th2-Polarized Moderate-to-Severe Persistent Asthma Endotype Subjected to High allergen exposure. Int Arch Allergy Immunol. 2021;182(1):21-31. doi: 10.1159/000510118.
8. Jiménez-Feijoo R, Pascal M, Moya R, Riggioni C, Domínguez O, Lózano J, et al. Molecular diagnosis in house dust mite-allergic patients suggests that Der p 23 is clinically relevant in asthmatic children. J Investig Allergol Clin Immunol. 2020;30(2):127-32. doi: 10.18176/jiaci.0431.
9. Zidarn M, Robič M, Krivec A, Šilar M, Resch-Marat Y, Vrtala S, Kopač P, Bajrović N, Valenta R, Korošec P. Clinical and immunological differences between asymptomatic HDM-sensitized and HDM-allergic rhinitis patients. Clin Exp Allergy. 2019;49(6):808-18. doi: 10.1111/cea.13361.
10. Vidal C, Chomón B, Pérez-Carral C, González-Quintela A. Sensitization to Lepidoglyphus destructor, Tyrophagus putrescentiae, and Acarus siro in patients allergic to house dust mites (Dermatophagoides spp.). J Allergy Clin Immunol. 1997;100(5):716-8. doi: 10.1016/s0091-6749(97)70181-7.
11. Plaza V, Blanco M, García G, Korta J, Molina J, Quirce S; en representación del Comité Ejecutivo de GEMA. Highlights of the Spanish Asthma Guidelines (GEMA), version 5.0. Arch Bronconeumol (Engl Ed). 2021;57(1):11-2. English, Spanish. doi: 10.1016/j.arbres.2020.10.003.
12. Global initiative for asthma. Guía de bolsillo para el manejo y la prevención del asma. 2019;1-32. Available at: https://ginasthma.org/wp-content/uploads/2019/07/GINA-Spanish-2019-wms.pdf.
13. Brożek JL, Bousquet J, Agache I, Agarwal A, Bachert C, Bosnic-Anticevich S, et al. Allergic Rhinitis and its Impact on Asthma (ARIA) guidelines-2016 revision. J Allergy Clin Immunol. 2017;140(4):950-8. doi: 10.1016/j.jaci.2017.03.050.
14. López-Rodríguez R, Rial MJ, Esteban-Gorgojo I, Veleiro **P**érez B, López-Araújo GA, Pérez-Quintero O, Carballas C, Parra A, Pineda F. Serodominance profile in a dust mite complex region. Int Arch Allergy Immunol. 2022;14:1-9. doi: 10.1159/000523869.
15. Kowal K, Pampuch A, Siergiejko G, Siergiejko Z, Swiebocka E, Schlachter CR, et al. Sensitization to major Dermatophagoides pteronyssinus allergens in house dust mite allergic patients from North Eastern Poland developing rhinitis or asthma. Adv Med Sci. 2020;65(2):304-9. doi: 10.1016/j.advms.2020.05.003.

16. Stranzl T, Ipsen H, Christensen LH, Eiwegger T, Johansen N, Lund K, Andersen PS. Limited impact of Der p 23 IgE on treatment outcomes in tablet allergy immunotherapy phase III study. Allergy. 2021;76(4):1235-8. doi: 10.1111/all.14200.
17. Limão R, Spínola Santos A, Araújo L, Cosme J, Inácio F, Tomaz E, et al. Molecular profile of sensitization to Dermatophagoides pteronyssinus dust mite in Portugal. J Investig Allergol Clin Immunol. 2021;32(1):33-9. doi: 10.18176/jiaci.0533.
18. Muddaluru V, Valenta R, Vrtala S, Schlederer T, Hindley J, Hickey P, et al. Comparison of house dust mite sensitization profiles in allergic adults from Canada, Europe, South Africa and USA. Allergy. 2021;76(7):2177-88. doi: 10.1111/all.14749.
19. Letrán A, García I, Espinazo-Romeu M, Moreno-Aguilar C, Moreno F. Cut-off value of D. pteronyssinus specific IgE in double negative patients Der p 1 and Der p 2 and its clinical repercussion. Sci Rep. 2021;11(1):23585. doi: 10.1038/s41598-021-03005-4.
20. Celi G, Brusca I, Scala E, Villalta D, Pastorello E, Farioli L, et al. House dust mite allergy in Italy-diagnostic and clinical relevance of Der p 23 (and of minor allergens): A real-life, multicenter study. Allergy. 2019;74(9):1787-9. doi: 10.1111/all.13776.
21. Eder K, Becker S, Gellrich D, Zieglmayer P, Gröger M. The Role of Der p 23 Sensitization: An analysis of 474 patients sensitized to mite. Int Arch Allergy Immunol. 2020;181(9):689-98. doi: 10.1159/000508522.
22. Posa D, Perna S, Resch Y, Lupinek C, Panetta V, Hofmaier S, et al. Evolution and predictive value of IgE responses toward a comprehensive panel of house dust mite allergens during the first 2 decades of life. J Allergy Clin Immunol. 2017;139(2):541-9.e8. doi: 10.1016/j.jaci.2016.08.014.

CASE REPORT

Eur Ann Allergy Clin Immunol Vol 56, N.2, 86-88, 2024

Federica Rivolta[1], Alessandra Chiei Gallo[2], Andrea Sangalli[2], Valerio Pravettoni[1]

Acetylsalicylic acid desensitization in an allergic pregnant woman post-vascular scaffolds implantation

[1]UOC General Medicine Immunology and Allergology, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy
[2]Allergy and Clinical Immunology Residency, University of Milan, Milan, Italy

Key words

Acetylsalicylic acid; desensitization; hypersensitivity drug reactions; NSAID hypersensitivity; pregnancy.

Corresponding author
Federica Rivolta
Department of General Medicine Immunology and Allergology
IRCCS Foundation Ca' Granda Ospedale Maggiore Policlinico
via Francesco Sforza 28
20122 Milan, Italy
ORCID: 0000-0002-4693-5632
E-mail: federica.rivolta@policlinico.mi.it

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Summary

The use of acetylsalicylic acid (ASA) desensitization for patients with coronary artery disease (CAD) is growing, but no data are available on desensitization protocol in patients with ASA sensitivity and CAD during pregnancy. This case report shows that ASA desensitization protocol during pregnancy could be safe and effective in a tertiary center with a multidisciplinary team.

Impact statement

ASA desensitization protocol during pregnancy could be safe and effective in a tertiary center with a multidisciplinary team.

Introduction

Non-steroidal anti-inflammatory drugs (NSAIDs), including acetylsalicylic acid (ASA), are associated with adverse effects, ranging from mild gastritis to life-threatening reactions. The overall prevalence of hypersensitivity reactions ranges from 0.6-6%, but no prevalence data are available in pregnant women (1).

The mechanism of action of NSAIDs is the inhibition of the cyclooxygenase (COX) enzymes. There are two main COX isoforms, COX-1 (which is expressed constitutively in all human cell types and is involved in protective physiologic functions) and COX-2 (which is an inducible enzyme expressed in many inflammatory cells in presence of appropriate stimuli) (2).

ASA, a strong COX-1 inhibitor preventing platelet aggregation, is a key-treatment for patients with coronary artery disease (CAD) and is also a cornerstone component of dual antiplatelet therapy (DAPT) with a $P2Y_{12}$ receptor blocker. However, hypersensitivity or intolerance may restrict its use (3).

In literature only four cases on ASA desensitization during pregnancy have been published, especially in women diagnosed with antiphospholipid syndrome (4).

Since there are no commercially available tests to detect NSAIDs hypersensitivity, a diagnostic strategy successfully used for decades is to challenge patients with past mild reactions to NSAIDs. However, ASA desensitization protocol is mandatory in case of positive challenge, or for safety reasons in history of severe reaction, or unstable pathology with compelling need for

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aspirin (5). Increasing doses of ASA under medical supervision are subsequently administered to the patient inducing a state of temporary tolerance (6). After a successful ASA-desensitization, the patient, in order to maintain tolerance, must assume the drug daily (4).

In patient suffering from an established cardiovascular disease or a high-risk patient, a daily therapy with ASA can reduce the risk of subsequent adverse cardiovascular events such as myocardial infarction, stroke, and vascular death (2). For this reason, ASA sensitivity is particularly problematic for individuals who need an urgent or emergency neurologic or cardiac procedure, such as artery stenting (coronary, carotid, or other) or following the diagnosis of an ischemic neurologic event.

Case presentation

We describe a case of a 42-year-old pregnant woman, with a past medical history of arterial hypertension, type 2 diabetes mellitus, dyslipidemia and overweight, affected by CAD, with prior percutaneous coronary intervention (PCI) with two overlapping bio-resorbable vascular scaffolds (Absorb) for unstable angina. After PCI she started DAPT with Clopidogrel and Indobufen.

She was referred to our tertiary center during the fifth month of pregnancy for an allergological evaluation. She was no longer in DAPT because Indobufen was interrupted by her cardiologist when her pregnancy became known (9 months after PCI). Then, a multidisciplinary team of obstetricians and experts of the Thrombosis and Coagulation Department recommended to restart DAPT with Clopidogrel and ASA as the best choice, considering the high risk of late stent thrombosis related to stent type (Absorb), number and procedural techniques (two stents in overlap), diabetes and pregnancy. Unfortunately, the patient had a positive history of hypersensitive reaction to ASA: some years before she had experienced an adverse reaction consisting in labial and periocular angioedema immediately (about 10 minutes) after ASA intake. Nevertheless, she tolerated other NSAID such as Indobufen, Ibuprofene and Ketoprofen. In this setting it was conceivable assuming patient reaction as a single NSAID-induced urticaria/angioedema (SNIUA) (7). Even if desensitization procedures are widely used in cardiological clinical practice, they are not recommended during pregnancy for the risk of anaphylactic reaction that could lead to fetal and maternal damages. However, a recent Food & Drug Administration (FDA) warning reported an increased rate of major adverse cardiac events observed in patients with bio-absorbable device (8).

After a consultation with a multidisciplinary team (anesthesiologists, obstetricians, cardiologists and hematologist), considering the relative low risk of an anaphylactic reaction *versus* the high risk of a stent thrombosis (11%), our Outpatients Allergy Department decided to perform an ASA desensitization treatment, followed by constant administration of the same drug at a disaggregating dose (100 mg/daily) (8, 9).

All procedures were in accordance with the ethical standards of the responsible committee on human experimentation (institutional and national) and with the Helsinki Declaration of 1975, as revised in 2013. The patient signed an informed consent and started a desensitization protocol in an intensive care unit of the Obstetrical Department with maternal and, if necessary, fetal monitoring.

The first day ten increasing doses of acetylsalicylic acid at 30-minute intervals were orally given to the patient, until the cumulative dose of 100.1 mg was reached, using Cortellini *et al.* desensitization protocol (**table I**) (5). The following day, the full dose of 100 mg was administered successfully and from that day, the patient assumed the same dose every day. No reaction was observed

Table I *- ASA desensitization protocol by Cortellini et al. [8].*

Number of doses (every 20 minutes)	mL of solution (10 mg/mL)	Dose of ASA (mg)	Cumulative dose (mg)
1	0 (placebo)	--	--
2	0.01	0.1	0.1
3	0.10	1	1.1
4	0.20	2	3.1
5	0.30	3	6.1
6	0.40	4	10.1
7	0.50	5	15.1
8	1.00	10	25.1
9	1.50	15	40.1
10	2.50	25	65.1
11	3.50	35	100.1

during the desensitization procedure and the following days. No antihistamine premedication was administered due to the possibility to mask any eventual muco-cutaneous reaction, a possible red flag of a more severe reaction.
The patient underwent, as planned, cesarean section scheduled on the 34th week, giving birth to a healthy child. DAPT was interrupted, stopping the administration of clopidogrel alone, from 5 days before to 1 day after the delivery.
Mother and newborn were both asymptomatic and in good general condition at a six-month follow-up after delivery.

Discussion and conclusions

Since the worldwide increasing use, NSAIDs are the most important drug involved in hypersensitivity reactions. According to the nomenclature of the EAACI/AAAAI task force, drug hypersensitivity reactions are classified as allergic and non-allergic (6).
ASA-hypersensitivity constitutes a serious problem for subjects with NSAID sensitivity and concomitant CAD, so ASA desensitization has to be considered, given the excellent clinical efficacy, low risk profile, and cost-effectiveness (10).
Moreover, during pregnancy, low-dose of ASA is indicated for prevention of preeclampsia, fetal growth restriction, stillbirth or obstetric complications related to antiphospholipid syndrome (9).
Based on the patient's clinical history and presentation (an immediate reaction after ASA intake), after a multidisciplinary evaluation, considering the greater risk of stent thrombosis compared to that of ASA reaction, we decided to use a desensitization protocol as the best approach in a pregnant patient with previous allergic reactions to ASA. This because ASA in association with clopidogrel is up to now considered the real effective therapy in patients affected by CAD with medicated stent (11).
Since an ASA challenge was not performed before pregnancy, desensitization has been carried out in a multidisciplinary setting with access to resuscitation facilities and an intensive care unit for maternal and fetal monitoring (4).
Even if desensitization procedures could be at risk during pregnancy, this case shows that, in a patient with a history of mild reaction to ASA, a desensitization protocol during pregnancy may be safe and effective in a tertiary center with a multidisciplinary team, thus making it a valuable option, when the drug is mandatory for the patient. Nonetheless, given the lack of international standardization protocols and limited published data available, more studies are necessary to establish the risks and benefits for ASA-desensitization protocols in ASA-sensitive pregnant women.

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Contributions

VP, FR: resources. ACG, AS: writing – original draft, data curation. All authors: writing - review & editing.

Conflict of interests

The authors declare that they have no conflict of interests.

References

1. Kowalski ML, Makowska JS, Blanca M, Bavbek S, Bochenek G, Bousquet J, et al. Hypersensitivity to nonsteroidal anti-inflammatory drugs (NSAIDs) - classification, diagnosis and management: review of the EAACI/ENDA(#) and GA2LEN/HANNA*. Allergy. 2011;66(7):818-29. doi: 10.1111/j.1398-9995.2011.02557.x.
2. Cheong Z, Tan CYL, Lim CP, Soong JL, Chong CJM, Chan AKW. Patient characterization and predictors of aspirin desensitization response. Asia Pac Allergy. 2021;11(2):e20. doi: 10.5415/apallergy.2021.11.e20.
3. Lambrakis P, Rushworth GF, Adamson J, Leslie SJ. Aspirin hypersensitivity and desensitization protocols: implications for cardiac patients. Ther Adv Drug Saf. 2011;2(6):263-70. doi: 10.1177/2042098611422558.
4. Benito-Garcia F, Pires I, Lima J. Aspirin Desensitization: Implications for Acetylsalicylic Acid-Sensitive Pregnant Women. Medicina (Kaunas). 2021;57(4):390. doi: 10.3390/medicina57040390.
5. Cortellini G, Romano A, Santucci A, Barbaud A, Bavbek S, Bignardi D, et al. Clinical approach on challenge and desensitization procedures with aspirin in patients with ischemic heart disease and nonsteroidal anti-inflammatory drug hypersensitivity. Allergy. 2017;72(3):498-506. doi: 10.1111/all.13068.
6. Cernadas JR, Brockow K, Romano A, Aberer W, Torres MJ, Bircher A, et al. General considerations on rapid desensitization for drug hypersensitivity - a consensus statement. Allergy. 2010;65(11):1357-66. doi: 10.1111/j.1398-9995.2010.02441.x.
7. Doña I, Blanca-López N, Cornejo-García JA, Torres MJ, Laguna JJ, Fernández J, et al. Characteristics of subjects experiencing hypersensitivity to non-steroidal anti-inflammatory drugs: patterns of response. Clin Exp Allergy. 2011;41(1):86-95. doi: 10.1111/j.1365-2222.2010.03651.x.
8. Center for Devices and Radiological Health. "FDA Investigating Increased Rate of Major Adverse Cardiac Events Observed in Patients Receiving Abbott Vascular's Absorb GT1 Bioresorbable Vascular Scaffold (BVS) - Letter to Health Care Providers." U.S. Food and Drug Administration. Available at: www.fda.gov/medical-devices/letters-health-care-providers/fda-investigating-increased-rate-major-adverse-cardiac-events-observed-patients-receiving-abbott.
9. American College of Obstetricians and Gynecologists. ACOG Committee Opinion No. 743: Low-Dose Aspirin Use During Pregnancy. Obstet Gynecol. 2018;132(1):e44-e52. doi: 10.1097/AOG.0000000000002708.
10. Gollapudi RR, Teirstein PS, Stevenson DD, Simon RA. Aspirin Sensitivity: Implications for Patients with Coronary Artery Disease. JAMA. 2004;292(24):3017-23. doi: 10.1001/jama.292.24.3017.
11. Camarero TG, de la Torre Hernández JM. Antithrombotic Treatment After Coronary Intervention: Agreement and Controversy. Eur Cardiol. 2020;15:1-8. doi: 10.15420/ecr.2019.25.2.

Maria Relvas[1], Joana Silva[2], Ana Luísa Matos[1], Francisca Alves[1], Margarida Gonçalo[1,3]

Concomitant evaluation of D-dimer and C-reactive protein in chronic spontaneous urticaria may show divergent values

[1]Department of Dermatology, Centro Hospitalar e Universitário de Coimbra, Coimbra, Portugal
[2]Department of Occupational Health, Centro Hospitalar e Universitário de Coimbra, Coimbra, Portugal
[3]Faculty of Medicine, University of Coimbra, Coimbra, Portugal

Key words

Chronic spontaneous urticaria; D-dimer; C-reactive protein; biomarkers; urticaria activity score.

Corresponding author
Maria Relvas
Department of Dermatology
Centro Hospitalar e Universitário de Coimbra
Praceta Prof. Mota Pinto
3004-561 Coimbra, Portugal
ORCID: 0000-0002-2639-7494
E-mail: mariavrelvas@gmail.com

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To the Editor,

two chronic spontaneous urticaria (CSU) subtypes with distinct pathomechanisms have been described: type IIb autoimmunity (IgG anti-FceRI/anti-IgE) and type I autoallergy (IgE anti-self/anti-thyroperoxidase/anti-interleukin-24) (1). In type IIb there is a higher female predominance, more angioedema, concomitant autoimmune diseases, positive basophil activation test (BAT) and autologous serum skin test (ASST), and a slow response to omalizumab, whereas in type I, response to this agent is usually fast (1). Recently, other concomitant pathomechanisms have been described, namely IgE anti-FceRI and immune-complexes with IgE, which may explain some overlap between the two subtypes and unexpected responses to omalizumab or in the ASST/BAT (2). Biomarkers that characterize these subtypes are still under evaluation, although high IgG anti-thyroperoxidase (anti-TPO) antibodies, low total serum IgE, basopenia and eosinopenia correlate better with type IIb autoimmunity (1). Other serum biomarkers, namely interleukin-6 (3), interleukin-17 (4), D-dimer (5) and C-reactive protein (C-RP) (6) seem to correlate with disease activity, but they are seldom studied simultaneously.

We conducted a retrospective study between 2019 and 2021 in our tertiary dermatology center, where we analysed data from 88 CSU patients, with the objective of evaluating the correlation between serum D-dimer and C-RP values, by comparing patients who had concordant levels of these biomarkers (both elevated or both normal) with those that had divergent values (one high and the other low). We also correlated these biomarkers with disease activity and with other markers that indicate CSU subtypes (type I and type IIb). Patients with pure chronic inducible urticaria or angioedema without wheals were excluded.

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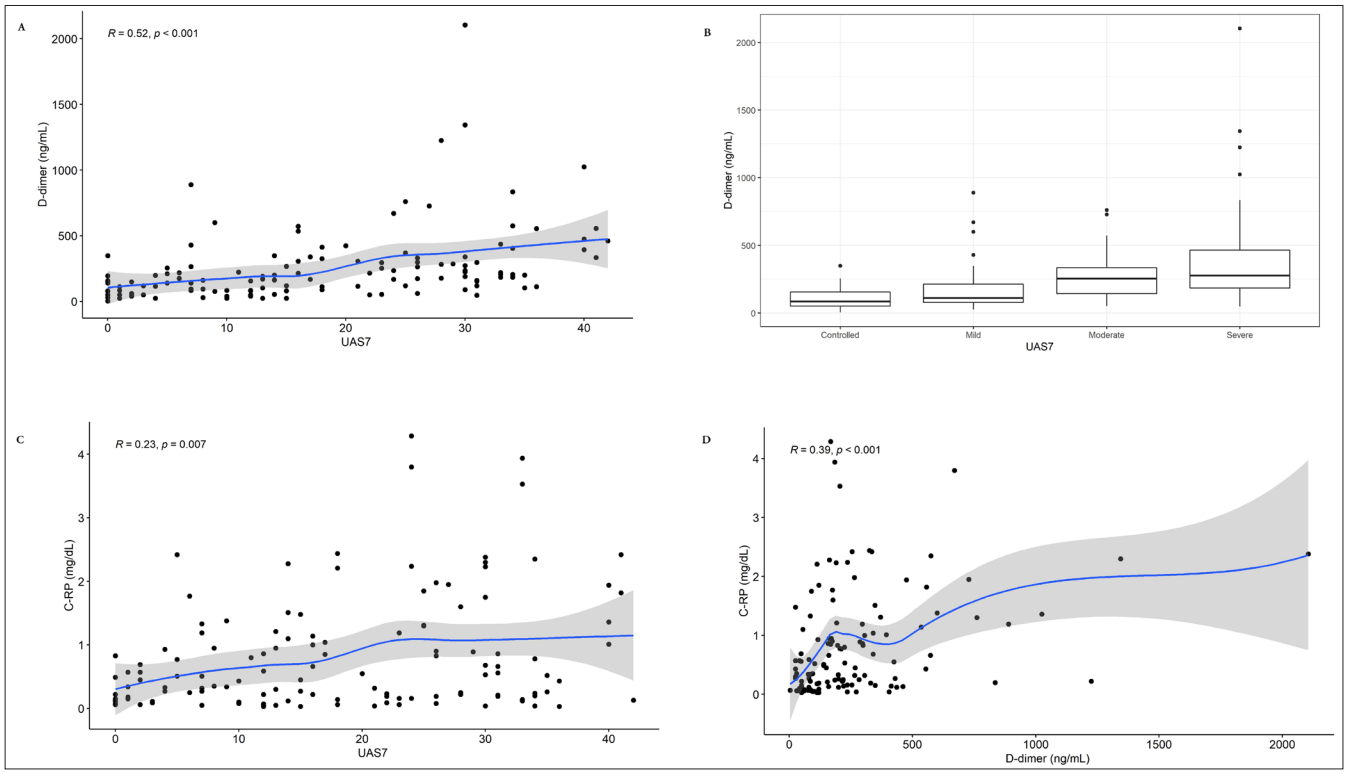
Clinical parameters included demographic data, CSU duration, concomitant angioedema, associated inducible urticaria, history of occasional wheals progressing to residual bruising and disease activity measured by Urticaria Activity Score over the last 7 days (UAS7; controlled, mild, moderate or severe for UAS7 between 0-6, 7-15, 16-27 or 28-42, respectively), evaluated the week before blood collection for the study. Venous blood was collected in the morning before the consultation and, for patients on omalizumab, 4-8 weeks after the last administration, depending on disease control and, consequently the interval between administrations. Parameters evaluated in the same blood sample included total serum IgE (normal < 100 UI/mL), eosinophil and basophil absolute counts, IgG anti-TPO and anti-nuclear antibodies (ANA, positive > 1:160), D-dimer (normal < 230 ng/mL) and C-RP (normal < 0.5 mg/dL) serum levels.

Statistical analysis was performed using R statistical software (version 4.0.5). Quantitative variables were tested for normality using Shapiro-Wilk test; group comparisons were made using Mann-Whitney test and Kruskal Wallis test. For pairwise comparisons P-values were presented with Bonferroni correction for multiple comparisons. Correlations between variables were made using Spearman correlation coefficient. The Chi-Square test of

A		B			
Patients' data	**Total n (%)**	**High D-dimer n (%) (n = 33)**	**High C-RP n (%) (n = 40)**	**Totally concordant n (%) (n = 49)**	**Totally divergent n (%) (n = 31)**
Age in years, mean ± SD	45 ± 15.8	50 ± 14.7	44.5 ± 14.4	43.6 ± 16.8	47.7 ± 14.3
Female	74 (84.1)	26 (78.8)	35 (87.5)	43 (87.8)	23 (74.2)
Angioedema	59 (67.1)	25 (75.8)	27 (67.5)	31 (63.3)	23 (74.2)
Concomitant inducible urticaria	39 (44.3)	/	/	/	/
Symptomatic dermographism	8 (9.1)	/	/	/	/
Delayed pressure urticaria	31 (35.2)	/	/	/	/
Cold urticaria	2 (2.3)	/	/	/	/
Heat urticaria	1 (1.1)	/	/	/	/
Solar urticaria	1 (1.1)	/	/	/	/
Residual bruising	21 (23.8)	11 (33.3)	9 (22.5)	9 (18.4)	7 (22.6)
ANA positive (> 1:160)	22 (25)	10 (30.3)	9 (22.5)	12 (24.5)	7 (22.6)
Anti-TPO positive	9 (11.4)	2 (6.1)	2 (5.0)	7 (14.3)*	1 (3.2)*
UAS7, median (IQR)	16 (7-29.5)	26 (19-33.5)	22 (13-30)	16 (8-26)	21 (13-30)
Controlled (0-6)	13 (14.8)	1 (3)	4 (10.0)	10 (20.4)	3 (9.7)
Mild (7-15)	24 (27.3)	4 (12.1)	11 (27.5)	13 (26.5)	9 (29.0)
Moderate (16-27)	24 (27.3)	13 (39.4)	12 (30.0)	15 (30.6)	7 (22.6)
Severe (28-42)	27 (30.7)	15 (45.5)	13 (32.5)	11 (22.4)	12 (38.7)
Low IgE (< 25 UI/mL)	21 (23.9)	7 (21.2)	14 (35)	12 (24.5)	6 (19.4)
Eosinopenia (< 100/mL)	25 (28.4)	11 (33.3)	10 (25)	18 (36.7)**	5 (16.1)**
Basopenia (0/mL)	26 (29.5)	11 (33.3)	25 (62.5)	17 (34.7)	8 (25.8)
		Median (IQR)			
CSU duration (months)	16.5 (5-60)	26 (6.5-78)	23 (8-69)	13 (5-54)	24 (5-60)
D-dimer (ng/mL)	181 (85-306)	405 (277.5-567.5)	188.5 (150-388)	150 (78.5-344)	185 (150-306)
C-RP (mg/dL)	0.45 (0.15-1.16)	0.55 (0.18-1.72)	1.32 (0.73-2.15)	0.27 (0.09-1.2)	0.83 (0.2-1.3)
Total IgE (UI/mL)	78 (20-255.5)	67 (27.8-261.0)	96 (31-265.5)	75 (25-249)	86 (24.8-275.3)
Eosinophils (mL)	130 (90-200)	110 (75-195)	140 (92-200)	100 (80-180)	170 (100-200)
Basophils (mL)	20 (0-50)	10 (0-95)	2 (2-40)	10 (0-45)	30 (10-80)

ANA: anti-nuclear antibodies; anti-TPO: anti-thyroperoxidase antibodies; C-RP: C-reactive protein; CSU: chronic spontaneous urticaria; IgE: immunoglobulin E; IQR: interquartile range; UAS7: urticaria activity score over 7 days; SD: standard deviation; *p = 0.142; **p = 0.052.

Figure 1 - (A) *The correlation between CSU activity and D-dimer serum levels;* ***(B)*** *Significant higher D-dimer values in patients with moderate-severe CSU versus mild disease;* ***(C)*** *The correlation between CSU activity and C-RP serum levels;* ***(D)*** *The correlation between D-dimer and C-RP serum levels.*



**Modified from Shaker et al. (72); subjectivity of studies regarding scoring anaphylaxis.*

independence was used to determine association between categorical variables. When more than 20% of cells have expected frequencies lower than 5, we used Fisher's exact test. We performed a local regression, a non-parametric approach that fits multiple regressions in local neighbourhood, using the loess () function. A P-value < 0.05 was considered significant.

Patients' demographic, clinical and laboratory data is summarized in **table IA**. From the 88 cases, 44.3% (n = 39) had one or more associated inducible forms of urticaria, including symptomatic dermographism (n = 8), delayed pressure urticaria (n = 31), cold (n = 2), heat (n = 1) and solar (n = 1) urticarias.

Patients were under treatment with second-generation H_1-antihistamines (n = 58) or omalizumab for 4-42 months (n = 30), with some in the latter group occasionally taking H_1-antihistamines on-demand when their disease was not completely controlled.

Median UAS7 was 16 (IQR 7-29.5) and 58% (n = 51) demonstrated moderate-severe disease (UAS7 > 15). Median D-dimer levels were 181 ng/mL (IQR: 85-306), with 33 patients (37.5%) exhibiting values above normal. D-dimer levels correlated with disease activity (p < 0.0001) (**figure 1A**), with significantly higher values in patients with moderate and severe CSU (median 254 (IQR: 120-240) and 277 (IQR 185-571.5), respectively) compared to mild disease (median 111.5 (IQR: 71.5-219.5); p = 0.026 and p = 0.01, respectively) (**figure 1B**). C-RP levels (median 0.45 mg/dL (IQR: 0.15-1.16)) were elevated in 40 patients (45.5%) and correlated with disease activity (p = 0.007) (**figure 1C**), but mean values of this marker for each of the four groups of urticaria activity (from controlled to severe) were not significantly different. Regarding the subgroup of patients with associated inducible forms of urticaria, we also found a correlation between D-dimer and UAS7 (p < 0.001) and C-RP and UAS7 (p = 0.141).

Median total serum IgE was 78 UI/mL (IQR: 20-255.5), with 23.9% of the patients showing low IgE levels (< 25 UI/mL). We did not find any correlation between D-dimer or C-RP and IgE levels.

Among patients with isolated high D-dimer (n = 16), isolated high C-RP (n = 23) or elevation of both biomarkers (n = 17), respec-

tively, 87.5%, 52.2% and 76.5%, demonstrated moderate-severe disease. Within the 51 patients with moderate-severe disease, both D-dimer and C-RP were normal in 11 of them (21.6%).

We did not find any association between high D-dimer or high C-RP and concomitant angioedema, history of bruising, eosinophil or basophil counts, IgE levels, anti-TPO or ANA positivity (**table IB**).

There was significant statistical correlation between D-dimer and C-RP levels ($r = 0.39$, $p < 0.001$) (**figure 1D**). In 57 patients (64.8%) both biomarkers were totally concordant (n = 49), either both normal or both elevated, or partially concordant (n = 8), meaning that only one biomarker was raised, but this elevation did no exceed 20% of the upper limit (*e.g.* 266 ng/mL for D-dimer and 0.6 mg/dL for C-RP). The remaining 31 patients (35.2%) showed totally divergent values. We found no significant differences in clinical or laboratory parameters between the groups with totally concordant or divergent values, although the percentage of patients with anti-TPO antibodies and eosinopenia was higher in the first group (**table IB**).

Our study confirms that both D-dimer and C-RP levels significantly correlate with CSU activity, as in previous studies (5, 6), and there is a positive correlation between these two biomarkers. Nevertheless, they fail to detect around 20% of moderate-severe CSU cases and assessing them, either separately or concomitantly, did not increase their sensitivity as markers of moderate-severe CSU activity.

Apart from CSU activity, there was no significant correlation between elevation of D-dimer or C-RP levels and the remaining clinical or laboratory parameters which are considered to target the two different CSU subtypes. Otherwise, as increasing overlap has been found between these subtypes (1, 7), these biomarkers may also be unable to distinguish them.

Interestingly, around one-third of the patients exhibited totally divergent D-dimer/C-RP levels. This may suggest the involvement of different pathways of inflammation or coagulation/fibrinolysis, either related to an individual response to mast cell degranulation or to specific CSU subtypes. Although we found no significant differences in the studied parameters between the patients with totally concordant or totally divergent D-dimer/C-RP values, the first group showed more frequently anti-TPO antibodies and eosinopenia, which, along with low IgE levels and basopenia, have been associated with type IIb autoimmune CSU (1, 8).

Limitations of this study include the reduced number of patients and their different therapeutic regimens, lack of ASST/BAT results, subjectivity of UAS7 as marker for CSU activity and other possible concomitant factors that may influence D-dimer and C-RP levels.

In conclusion, D-dimer and C-RP serum levels are sensitive biomarkers for CSU activity, but their concomitant use does not seem to increase their sensitivity in detecting higher disease activity. Nevertheless, we could not explain the divergent D-dimer and C-RP results in around one-third of the patients, which do not clearly correlate with the two main pathomechanisms recognized in CSU. More studies are needed to understand and define the particular group of patients who demonstrate a totally divergent response concerning these two biomarkers.

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Contributions

MR: conceptualization, investigation, methodology, resources, visualization, writing – original draft, writing – review & editing. JS: methodology, formal analysis, writing – original draft, writing – review & editing. ALM, FA: investigation, resources. MG: conceptualization, methodology, resources, supervision, writing – original draft, writing – review & editing.

Conflict of interests

The authors declare that they have no conflict of interests.

References

1. Kolkhir P, Kovalkova E, Chernov A, Danilycheva I, Krause K, Sauer M, et al. Autoimmune Chronic Spontaneous Urticaria Detection with IgG Anti-TPO and Total IgE. J Allergy Clin Immunol Pract. 2021;9(11):4138-46. doi: 10.1016/j.jaip.2021.07.043.
2. Asero R, Marzano AV, Ferrucci S, et al. Co-occurrence of IgE and IgG autoantibodies in patients with chronic spontaneous urticaria. Clin Exp Immunol, 2020;200(3):242-9. doi: 10.1111/cei.13428.
3. Kasperska-Zajac A, Sztylc J, Machura E, Jop G. Plasma IL-6 concentration correlates with clinical disease activity and serum C-reactive protein concentration in chronic urticaria patients. Clin Exp Allergy. 2011;41(10):1386-91. doi: 10.1111/j.1365-2222.2011.03789.x.
4. Grzanka A, Damasiewicz-Bodzek A, Kasperska-Zajac A. The relationship between circulating concentrations of interleukin 17 and C reactive protein in chronic spontaneous urticaria. Allergy Asthma Clin Immunol. 2017;13:25. doi: 10.1186/s13223-017-0197-6.
5. Asero R, Tedeschi A, Riboldi P, Griffini S, Bonanni E, Cugno M. Severe chronic urticaria is associated with elevated plasma levels of D-dimer. Allergy. 2008;63(2):176-80. doi: 10.1111/j.1398-9995.2007.01514.x.
6. Kolkhir P, Altrichter S, Hawro T, Maurer M. C-reactive protein is linked to disease activity, impact, and response to treatment in patients with chronic spontaneous urticaria. Allergy. 2018;73(4):940-8. doi: 10.1111/all.13352.
7. Schoepke N, Asero R, Ellrich A, Ferrer M, Gimenez-Arnau A, E H Grattan C, et al. Biomarkers and clinical characteristics of autoimmune chronic spontaneous urticaria: Results of the PURIST Study. Allergy. 2019;74(12):2427-36. doi: 10.1111/all.13949.
8. Santiago L, Ferreira B, Ramos L, Gonçalo M. IgE levels are negatively correlated with clinical response to ciclosporin in chronic spontaneous urticaria. Br J Dermatol. 2019;180(1):199-200. doi: 10.1111/bjd.17005.

Gennaro Liccardi[1], Matteo Martini[2,3], Maria Beatrice Bilò[2,3], Manlio Milanese[4], Maria Vittoria Liccardi[5], Gloria Pane[6], Raffaele Zeccolini[7], Paola Rogliani[1,6]

Bronchospasm and iodinated contrast media (ICM): an accurate evaluation is mandatory

[1]Department of Experimental Medicine, Postgraduate School of Respiratory Medicine, University of Rome Tor Vergata, Rome, Italy
[2]Department of Clinical and Molecular Sciences, Marche Polytechnic University, Ancona, Italy
[3]Allergy Unit, Department of Internal Medicine, University Hospital Ospedali Riuniti, Ancona, Italy
[4]Division of Pulmonology, S. Corona Hospital, Pietra Ligure, Savona, Italy
[5]Psychologist, Naples, Italy
[6]Respiratory Medicine Unit, Department of Experimental Medicine, University of Rome Tor Vergata, Rome, Italy
[7]Postgraduate School of Radiodiagnostics, University Luigi Vanvitelli, Naples, Italy

Key words

Bronchial asthma; bronchospasm; iodinated contrast media; hypersensitivity; psychological stress.

Corresponding author
Gennaro Liccardi
Department of Experimental Medicine
Postgraduate School of Respiratory Medicine
University of Rome Tor Vergata
via Cracovia 50
00133 Rome, Italy
ORCID: 0000-0002-0433-1339
E-mail: gennaro.liccardi51@gmail.com

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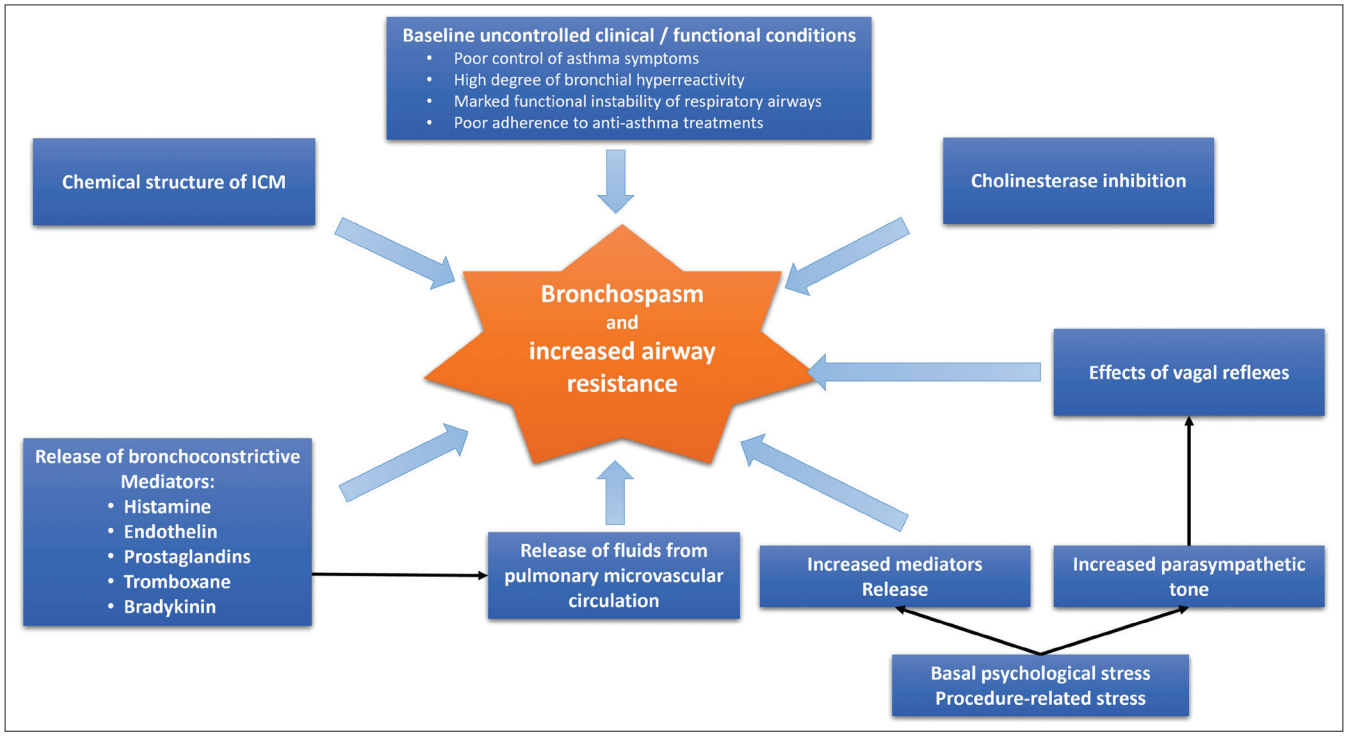
To the Editor,

we have greatly appreciated the editorial *Iodinated contrast media hypersensitivity reactions: is it time to re-evaluate risk factors?* (doi: 10.23822/EurAnnACI.1764-1489.245) and three articles about the relevant topic of hypersensitivity reactions to iodinated contrast media (ICM), published on *European Annals of Allergy and Clinical Immunology* on March 2022 (*Hypersensitivity reactions to iodinated contrast media in Italy: a retrospective study. Characteristics of patients and risk factors* – doi: 10.23822/EurAnnACI.1764-1489.225; *Isn't it time to consider oncological status as a new risk factor of iodinated contrast media hypersensitivity?* – doi: 10.23822/EurAnnACI.1764-1489.242; *Anaphylactic shock to iodinated contrast media: not so rare after all* – doi: 10.23822/EurAnnACI.1764-1489.210).

With this letter we wish to stress 1) the importance of respiratory symptoms (with or without concomitant manifestations) as adverse events to ICM, and 2) the need for an accurate clini-

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Figure 1 *- Factors and mechanisms increasing the risk of ICM induced bronchospasm in asthmatics.*



Modified from Liccardi et al. (15).

cal evaluation of asthmatics undergoing radiological procedures with ICM, aiming at 3) deciding when and which preventive pharmacological approach should be recommended.

We consider bronchospasm as a respiratory adverse event to ICM. According to the Ring and Messmer scale commonly adopted for reaction severity, respiratory symptoms are included into II and III grade and are associated to other multiorgan clinical manifestations (1-4). However, bronchospasm can be the only or prevalent symptom in particular circumstances, particularly in non-controlled asthma (5). Moreover, asthma severity is a risk factor for acute adverse events to contrast agents (6), and this is true also within Step 1 of GINA classification. The apparently unexplainable higher risk in Step 1, reported by Kobayashi *et al.* (6), could be justified by the fact that their study enrolled much more GINA Step 1 asthmatics than patients belonging to the other Steps and, above all, none of them were taking inhaled corticosteroids. On the contrary, from a general point of view, there is a direct correlation between the degree of asthma severity and female sex, and the risk of developing anaphylactic reactions (7).

Lung is a key target for water soluble ICM, and this is reflected by bronchospasm, that constitutes the most common adverse event, among respiratory reactions, to ICM (8), with a reported occurrence of 23% and 5% cases of moderate and severe reactions, respectively (9). Although symptomatic/severe bronchospasm seems to be rare, sub-clinical bronchospasm assessed by a fall in FEV_1 values is quite common, particularly in patients with an history of allergy and/or asthma (10). Furthermore, a bronchospasm can develop both during systemic reactions (*e.g.*, IgE/non IgE-mediated anaphylaxis) and as isolated events (10). Although low osmolar ICM have been reported to induce a reduced frequency of bronchospasm, Wilson and Davies (11) found that both high osmolar and low osmolar non-ionic ICM produce a comparable fall in FEV_1 and FVC, the latter an expression of air trapping (theoretically more dangerous). Other studies have shown percentages of bronchospasm between 0.18 and 4% with non-ionic ICM for computer tomography and fluorescein angiography, and also with magnetic resonance contrast medium (RCM)-gadolinium chelates (12).

The pathogenesis of ICM-induced bronchospasm is complex and not completely understood, but it is likely that this could be multifactorial (**figure 1**): the chemical structure itself, the release of bronchoconstrictor mediators (such as histamine, endothelin, prostaglandins, thromboxane and bradykinin), and the cholinesterase inhibition may play a role (13). Some of these released mediators can also cause leakage of fluids from the lung microvasculature which further contributes to increase airways

resistance (13). Recently, the role of stress as predisposing/triggering factor in asthma has been extensively studied. Individuals with stress due to psychological disorders or to the fear of ICM administration might experience vagal hypertone and, consequently, increased risk of bronchospasm (14).

An accurate evaluation of respiratory status is of essence. Despite this well-known relevance of respiratory adverse events to ICM, little attention has been paid by healthcare professionals to the risk of bronchospasm in allergic/asthmatic patients requiring ICM. In fact, no recent articles have been published on this topic to suggest diagnostic evaluations or pharmacological preventive treatments (*e.g.*, $ß_2$ agonists), with consequent poor awareness about this risk.

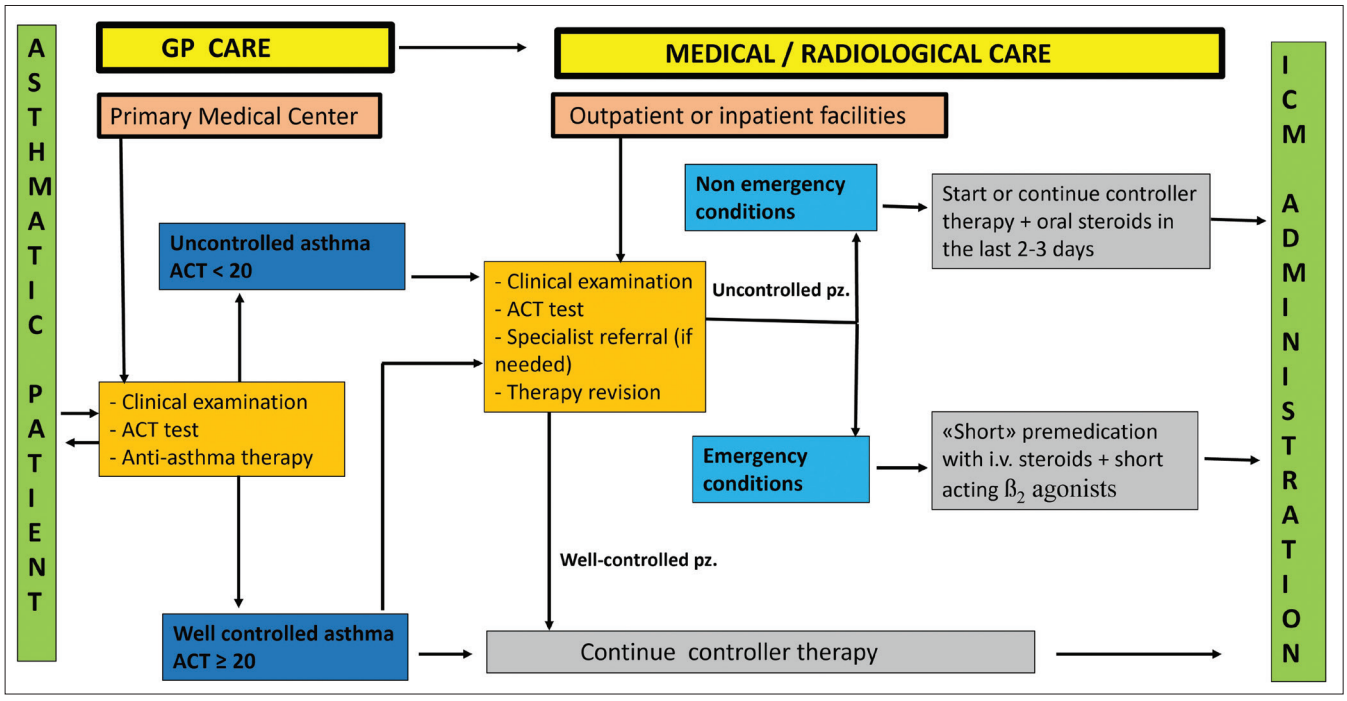
This is true either in inpatient care (and, consequently, in the controlled environment of the hospital setting) and in outpatient care (*e.g.*, private radiology centers).

Bearing in mind the risk of potentially fatal respiratory adverse events, all patients suffering from persistent (moderate/severe) bronchial asthma should undergo an accurate evaluation, before ICM administration. This should include a thorough clinical assessment, ideally performed by pulmonologists/allergists or by internists trained in respiratory medicine, aimed at optimizing asthma control (15), as it is likely that an optimal control of asthma symptoms could be useful in reducing the risk of bronchospasm in asthmatic patients (8). Evaluation of Asthma Control Test (ACT) score could be very useful to detect uncontrolled patients potentially at higher risk of bronchospasm. In patients suffering from seasonal allergic asthma the examination with ICM could be postponed, if possible, outside the peak of the pollen season or after adequate pharmacologic treatment.

Approach to prevent ICM-induced bronchospasm is needed. Patients at risk of respiratory adverse events should be treated with an optimal anti-inflammatory and reliever therapy to reduce bronchial hyperreactivity and consequently the risk of bronchospasm. It is likely that subclinical bronchospasm might determine greater obstructive effects in airways of asthmatics with uncontrolled asthma (11). Although uncontrolled asthma, does not represent a contraindication to perform ICM administration in emergency setting, it is quite likely that ICM administration would be safer in asthmatic patients that are usually well-controlled, during their daily life, when it is possible to perform a clinical evaluation and stepping up the therapy in case of poor control.

For this reason, general practitioners (GP) play an important preventive role, as they manage asthmatic patients in "Real Life", leveraging on their long-term knowledge of patients' medical history and their constant commitment in managing and following up their chronic conditions (16). This high level of knowledge of the specific patient cannot be reached by the radiologist, as he/she may have just one single contact with the patient (*i.e.*, when the radiologic procedure is performed) (16). **Figure 2** shows a possible flow-chart for the management of the risk of ICM – induced bronchospasm, for different healthcare

Figure 2 *- Suggested flow-chart (from GP to radiologist/internist) in managing asthmatic patient before ICM administration.*



professionals and settings (GPs, radiologists, and pulmonologists/internists, working in inpatient or outpatient care).
It is well known that asthmatic patients undergoing administration of water soluble ICM have an increased risk of potentially severe bronchospasm. It has been shown that the most important risk factor for bronchospasm is represented by a high degree of bronchial hyperreactivity with airway instability, not adequately controlled by long-term anti-inflammatory treatments. Therefore, an accurate clinical evaluation of asthmatics (especially those with a relevant degree of asthma severity) should be recommended, before ICM administration. Guidelines shared by pulmonologists, allergists and radiologists should be developed in the future for a better evaluation and management of these patients.

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Contributions

All authors contributed equally to this work.

Conflict of interests

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References

1. Voltolini S, Cofini V, Murzilli F, Bignardi D, Borro M, Calamari M, et al. Hypersensitivity reactions to iodinate contrast media in Italy: a retrospective study. Characteristics of patients and risk factors. Eur Ann Allergy Clin Immunol. 2022;54(2):60-7. doi: 10.23822/EurAnnACI.1764-1489.225.
2. Nucera E, Parrinello G, Gangemi S, Buonomo A, Aruanno A, Lohmeyer FM, et al. Contrast Medium Hypersensitivity: A Large Italian Study with Long-Term Follow-Up. Biomedicines. 2022;10(4):759. doi: 10.3390/biomedicines10040759.
3. Torres MJ, Trautmann A, Böhm I, Scherer K, Barbaud A, Bavbek S, et al. Practice parameters for diagnosing and managing iodinated contrast media hypersensitivity. Allergy. 2021;76(5):1325-39. doi: 10.1111/all.14656.
4. Costantino MT, Romanini L, Gaeta F, Stacul F, Valluzzi RL, Passamonti M, et al SIRM-SIAAIC consensus, an Italian document on management of patients at risk of hypersensitivity reactions to contrast media. Clin Mol Allergy. 2020;18:13. doi: 10.1186/s12948-020-00128-3.
5. Liccardi G, Salzillo A, De Blasio F, D'Amato G. Control of asthma for reducing the risk of bronchospasm in asthmatics undergoing general anesthesia and/or intravascular administration of radiographic contrast media. Curr Med Res Opin. 2009;25(7):1621-30. doi: 10.1185/03007990903010474.
6. Kobayashi D, Takahashi O, Ueda T, Arioka H, Akaishi Y, Fukui T. Asthma severity is a risk factor for acute hypersensitivity reactions to contrast agents: a large-scale cohort study. Chest. 2012;141(5):1367-8. doi: 10.1378/chest.11-3143.
7. González-Pérez A, Aponte Z, Vidaurre CF, Rodríguez LA. Anaphylaxis epidemiology in patients with and patients without asthma: a United Kingdom database review. J Allergy Clin Immunol. 2010;125(5):1098-104.e1. doi: 10.1016/j.jaci.2010.02.009.
8. Morcos SK. Review article: Effects of radiographic contrast media on the lung. Br J Radiol. 2003;76(905):290-5. doi: 10.1259/bjr/54892465.
9. Bertrand P, Rouleau P, Alison D, Chastin I. Use of peak expiratory flow rate to identify patients with increased risk of contrast medium reaction. Results of preliminary study. Invest Radiol. 1988;23 Suppl 1:S203-5. doi: 10.1097/00004424-198809001-00039.
10. Littner MR, Ulreich S, Putman CE, Rosenfield AT, Meadows G. Bronchospasm during excretory urography: lack of specificity for the methylglucamine cation. AJR Am J Roentgenol. 1981;137(3):477-81. doi: 10.2214/ajr.137.3.477.
11. Wilson AR, Davies P. Ventilatory function during urography: a comparison of iopamidol and sodium iothalamate. Clin Radiol. 1988;39(5):490-3. doi: 10.1016/s0009-9260(88)80208-3.
12. Dillman JR, Strouse PJ, Ellis JH, Cohan RH, Jan SC. Incidence and severity of acute allergic-like reactions to i.v. nonionic iodinated contrast material in children. AJR Am J Roentgenol. 2007;188(6):1643-7. doi: 10.2214/AJR.06.1328.
13. Poros B, Katchanov J, Curta A, Weig T. Schweres nichtkardiales Lungenödem durch Permeabilitätsstörung nach i.v.-CT-Kontrastmittel-Gabe mit konsekutiver venovenöser extrakorporaler Membranoxygenierung [Severe noncardiogenic pulmonary edema due to permeability disorder after i.v. administration of CT contrast medium with subsequent venovenous extracorporeal membrane oxygenation]. Anaesthesist. 2020;69(8):579-82. German. doi: 10.1007/s00101-020-00804-8.
14. Liccardi G, Calzetta L, Milanese M, Bilò MB, Liccardi MV, Matera MG, et al. Anxiety and depression in adolescents with asthma and in their parents. Is an increased basal cholinergic tone a possible further reason to explain the negative impact on asthma control? Monaldi Arch Chest Dis. 2020 8;90(1). doi: 10.4081/monaldi.2020.1233
15. Liccardi G, Salzillo A, Piccolo A, De Napoli I, D'Amato G. The risk of bronchospasm in asthmatics undergoing general anaesthesia and/or intravascular administration of radiographic contrast media. physiopatology and clinical/functional evaluation. Eur Ann Allergy Clin Immunol. 2010;42(5):167-73.
16. Liccardi G, Infantino A, Paggiaro P, Fabbri LM, Salzillo A, Infantino R, et al. GPs' role in reducing the risk of bronchospasm in asthma patients undergoing general anesthesia and/or intravascular administration of radiographic contrast media. Prim Care Respir J. 2011;20(3):333-5. doi: 10.4104/pcrj.2011.00044.

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