# Recent knowledge and insights on the mechanisms of immediate hypersensitivityand anaphylaxis : Ige/Fcεri- and non-Ige/Fcεri-dependent anaphylaxis

# Recent knowledge and insights on the mechanisms of immediate hypersensitivity and anaphylaxis: IgE/FcεRI- and non-IgE/FcεRI-dependent

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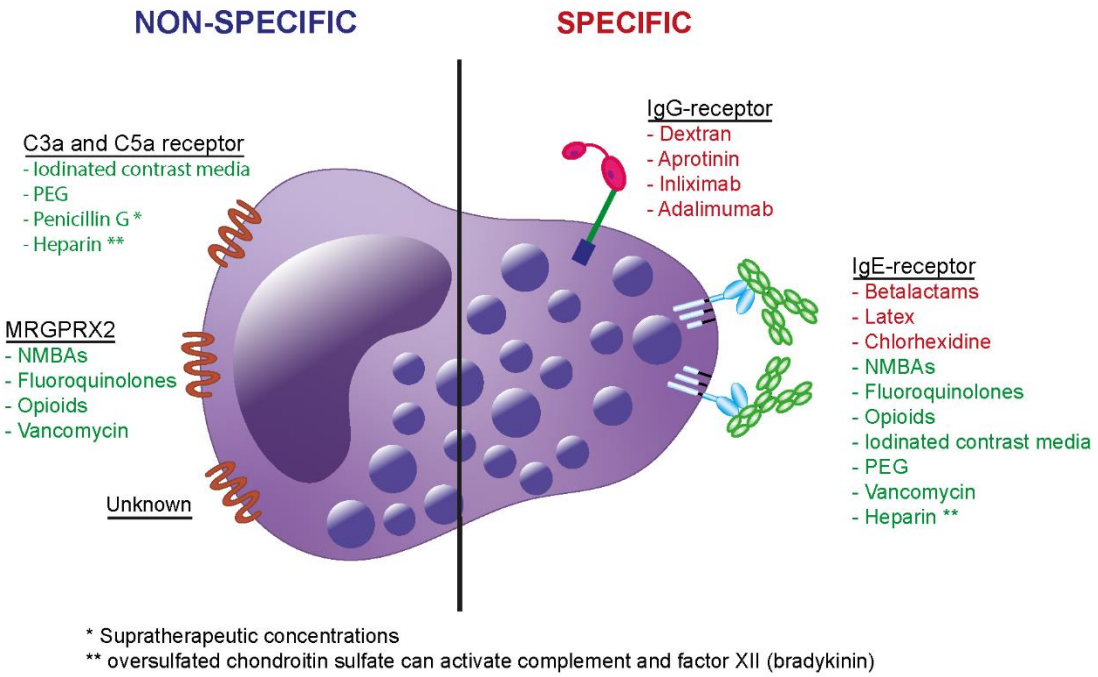
**Graphical abstract**




**Mast cell (MC) and/or basophil activation can occur via non-specific and specific pathways.**

Non-specific activation predominantly involves activation of complement receptors C3aR and C5aR. Another way of non-specific MC activation implies occupation of the Mas-related G protein coupled-receptor X2 (MRGPRX2). Note that MRGPRX2 is not constitutively expressed by basophils. Specific activation of MCs and basophils requires activation of B lymphocytes of the adaptive immune system with production of allergen-specific antibodies.

**Abstract**

Immediate hypersensitivity reactions can pose a clinical and diagnostic challenge, mainly because of the multifarious clinical presentation and distinct underlying – frequently uncertain – mechanisms. Anaphylaxis encompasses all rapidly developing and life-threatening signs and may cause death. Evidence has accumulated that immediate hypersensitivity and anaphylaxis does not necessarily involve an allergen-specific immune response with cross-linking of specific IgE (sIgE) antibodies bound to their high affinity IgE receptor (FcεRI) on the surface of mast cells (MCs) and basophils. Immediate hypersensitive and anaphylaxis can also result from alternative specific and nonspecific MC and basophils activation and degranulation such as complement-derived anaphylatoxins and off-target occupancy of MC and/or basophil surface receptors such as the Mas-related G protein-coupled receptor X2 (MRGPRX2). Degranulation of MCs and basophils results in the release of inflammatory mediators which can be, depending on the underlying trigger, in a different spatiotemporal manner. In addition, hypersensitivity and anaphylaxis can occur entirely independent from MC and basophil degranulation, as is observed in hypersensitivity to nonsteroidal anti-inflammatory drugs (NSAIDs) that divert normal arachidonic acid metabolism by inhibiting the cyclooxygenase (COX)-1 isoenzyme. Finally, one should remember that anaphylaxis might be part of the phenotype of particular – sometimes poorly recognizable - conditions such as clonal MC diseases (e.g. mastocytosis) and MC activation syndrome (MCAS). This review provides a status update on the different molecular mechanisms involved in both sIgE/FcεRI- and non-sIgE/FcεRI-dependent immediate hypersensitivity and anaphylaxis. In conclusion, there is increasing evidence for alternative pathophysiological hypersensitivity and anaphylaxis endotypes that are phenotypically and biologically indistinguishable, which are frequently difficult to diagnose, mainly because of uncertainties associated with diagnostic tests that might not absolutely enable to unveil the underlying mechanism.

**Key words:** Anaphylaxis, mast cells, basophils, IgE, MRGPRX2, pathomechanisms

## Nomenclature

In consonance with the actualized nomenclature of allergy [1], the umbrella term "hypersensitivity reactions" encompasses all unexpected but reproducible symptoms that extend beyond the primary pharmacological activity and can result from the activation of immune cell, inflammatory pathways or both. As such, hypersensitivity reactions are defined without the underlying pathophysiological processes. According to the underlying mechanism, hypersensitivity reactions can be further subdivided into allergic and nonallergic hypersensitivity to indicate a specific (immune) or non-specific (non-immune) mechanism, respectively. The term "anaphylaxis" encompasses all rapidly developing and life-threatening signs and symptoms involving multiple (>2) organ systems, irrespective of the underlying mechanistic endotype and should not be reserved for specific, mainly sIgE/FcεRI-dependent reactions [2]. However, a recent definition from the WAO does not include anymore the need of multiple organ involvement as severe symptoms can present in only one organ system [3]. When a specific immunologic mechanism is demonstratable, the term "allergic anaphylaxis" is applicable. An anaphylactic reaction mediated by cross-linking of allergen-specific IgE (sIgE) bound to its high-affinity IgE receptor (FcεRI) on the surface of mast cells (MCs) and basophils may be called sIgE/FcεRI-dependent anaphylaxis (e.g., as in (possibly exercise-induced) food anaphylaxis and Hymenoptera venom allergy. All other reactions should be reported as nonspecific or nonallergic anaphylaxis. The terms "anaphylactoid" and "pseudoallergic" should be avoided as various alternative specific and nonspecific hypersensitivity mechanisms, that can explain basophil and MC activation and degranulation independent from sIgE/FcεRI-signalling, have also been described. However, many authors still continue to use the obsolete terminology, which, regrettably, do not help harmonization of classification and deepening insights in pathomechanistic studies. In this review and shown in table 1, we provide a status update on the molecular mechanisms of immediate hypersensitivity reactions and anaphylaxis. For a narrative review of the pathophysiology of anaphylaxis, the reader is referred elsewhere [4].

## sIgE/FcεRI- and non- sIgE/FcεRI dependent mast cell and basophil activation and degranulation

The key effector cells of immediate hypersensitivity and anaphylaxis are MCs and basophils. As illustrated in the graphical abstract, activation and degranulation of these cells can be initiated by various specific (immune) and nonspecific (non-immune) mechanisms that occur via distinct complex signalling pathways. The underlying signalling mechanisms responsible for MC and basophil activation and degranulation and inhibition as well as the exocytic pathways exceed the scope of this review and have been thoroughly described elsewhere [5].

### *Specific effector cell activation*

Classically, allergic hypersensitivity involves activation of T-and B-lymphocytes of the adaptive immune system resulting in the synthesis and secretion of sIgE antibodies by plasma cells. These secreted sIgE antibodies bind to the membrane FcεRIs that are present on basophils and MCs. When a specific allergen cross-links sIgE/FcεRI-complexes, downstream signalling finally culminating in a compounded degranulation with exteriorization of aggregates of secretory granules and release of quantifiable mediators is set in motion. Typical examples of IgE-mediated hypersensitivity reactions are allergies against β-lactams (e.g. penicillins and cephalosporins), *Hevea* latex and the biguanide antiseptic chlorhexidine. The presence of sIgE antibodies is a *condition sine qua* but does not always lead to an effective cross-linking of FcεRI complexes with activation and degranulation. Although the requirements for effector cell activation and degranulation are not yet fully unravelled, the number of cross-links

per MC or basophil and the duration of it, as well the number of IgE recognition sites on the allergen (epitopes or antigenic determinants) are critical elements [6].

In contrast, specific activation and degranulation of both effector cells and anaphylaxis can also happen independently from allergen sIgE [7-9]. A specific activation and degranulation mechanism, but sIgE/FcεRI-independent, is allergen-specific cross-linking of IgG/FcγR complexes. However, insight in IgG-mediated anaphylaxis are predominantly obtained in animal models, clinical evidence for IgG-mediated anaphylaxis in humans is restricted to a few observations involving the parenteral administration of large amount of (proteinaceous) allergens. Potential IgG-dependent anaphylaxis has been described with different chimeric, humanized and even fully human monoclonal antibodies such as infliximab [10, 11] and adalimumab [12], and subjects treated with dextrans [13] or aprotinin [14, 15]. However, the pertinence of some of these observations remains uncertain and debatable [15], mainly because of the demonstrable drug-sIgE antibodies in some cases.

*Non-specific effector cell activation*

Mast cell and basophil activation and degranulation can also result from non-specific (antibody-independent) mechanisms. First, activation and degranulation of the effector cells can occur through binding of the anaphylatoxins C3a and C5a to their specific G-protein coupled receptors C3aR and C5aR on the membrane of MCs and basophils. Examples of complement-mediated reactions are reactions to iodinated contrast media, and reactions to over-sulphated chondroitin sulphate contaminated heparin [16, 17]. Second, MC activation and degranulation can also result from the off-target occupation of the Mas-related G protein-coupled receptor X2 (MRGPRX2) [18]. Since the seminal reporting, it seems that occupation of MRGPRX2 by various drug classes might herald an additional mechanism of nonspecific (nonimmune) immediate drug hypersensitivity reactions [19]. This pathway seems to be involved in the majority of reactions towards fluoroquinolones (FQs), opiates such as morphine, and the bradykinin receptor 2 antagonist icatibant. For some drug(s) (classes) the situation seems even more complicated. For example, for curarizing neuromuscular blocking agents (NMBAs) it appears that some of them might predominantly trigger effector cell degranulation via an IgE-dependent pathway, whilst others likely mainly act via the MRGPRX2 (for review: [20, 21]). However, current evidence for activation of the MRGPRX2 receptor almost exclusively comes from preclinical animal and *in vitro* studies. Therefore, translation into clinical relevance in humans should be done with prudently. Presentation of the preclinical animal or *in vitro* data is beyond the scope of this review and have been thoroughly described elsewhere [22, 23]. Data in humans is limited to our attempt to compare the clinics skin test responses and changes in serum tryptase in response to sIgE/FcεRI-dependent and likely MRGPRX2-dependent anaphylaxis to the aminosteroid-derived NMBA rocuronium. It seems that clinical details, acute tryptase, and delta tryptase are indistinguishable. In contrast, skin mast cells that strongly express MRGPRX2 appear to be less sensitive in the likely MRGPRX2-dependent hypersensitivity [24]. Alternatively, it is uncertain whether the opioid receptors are involved in hypersensitivity to some opiates and semisynthetic opioids [25]. As shown in the graphical abstract, a specific drug can activate MCs (and basophils) through distinct pathways. Importantly, the IgE-mediated and MRGPRX2-mediated activation can unite to boost activation and degranulation of MCs [26].

**Hypersensitivity reactions beyond the effector cells**

The role of other activation processes or mediators of cells other than MCs and basophils needs to be further studied. For example, as reviewed by Bruhns and Chollet-Martin, from experimental models of anaphylaxis, it emerges that drug-induced anaphylaxis might also involve other cells such as platelets and neutrophils [27]. However, for the time being, human evidence in this respect is rare and seems restricted to a single report on IgG-dependent neutrophil activation in response to neuromuscular blocking agents (NMBAs). This study demonstrated that concentrations of anti-NMBA IgG and markers of FcγR activation, PAF release, and neutrophil activation correlated with anaphylaxis severity. Neutrophil activation could also be observed in patients without evidence of an IgE-dependent anaphylaxis. Therefore, suggests the existence of an IgG-neutrophil pathway in human NMBA-induced anaphylaxis, which may strengthen anaphylaxis in combination with the IgE pathway or underlie anaphylaxis in the absence of specific IgE [28]. However, confirmation and studies with other drugs are needed to establish whether this mechanism might find universal acceptance.

*Hypersensitivity and anaphylaxis resulting from enzyme interference (mainly inhibition)*

Non-steroidal anti-inflammatory drugs (NSAIDs) are a predominant cause of immediate drug hypersensitivity reactions. These reactions can be classified into two distinct groups according to their onset mechanism. First, most frequently, reactions induced by nonspecific mechanisms (nonallergic reactions) that include different clinical phenotypes such as NSAID-exacerbated respiratory disease, cutaneous disease and urticaria/angioedema, NERD, NECD, and NIUA respectively. The pathogenesis of nonspecific NSAID hypersensitivity syndromes relates to nonselective inhibition cyclooxygenase (COX)-1 and 2 iso-enzymes which results in a blocking of prostaglandin synthesis and increases production of leukotrienes eventually causing NERD, NECD and NIUA. Secondly, much rarer, reactions resulting from specific mechanisms (allergic or selective reactions) are designated as single NSAID-induced urticaria/angioedema or anaphylaxis in which interclass cross-reactivity is almost absent [29].

The angiotensin-converting enzyme (ACE) is a key player of the renin-angiotensin system that converts angiotensin I to angiotensin II. Additionally, it is also responsible for the degradation of bradykinin, which is generated from high molecular weight kininogen by kallikrein via the contact system. Via bradykinin 2 receptors (BK2Rs), bradykinin affects vascular permeability and stimulates the release of substance P. This release causes vasodilation and fluid extravasation into tissues. ACE-inhibitors block the degradation of bradykinin which is thought to be a likely explanation for - sometimes life-threatening and difficult to treat – ACE inhibitor-induced angioedema. Moreover, inhibition of ACE potentiates anaphylactic reactions which is at least partially mediated by direct MC priming [30].

**Anaphylaxis as part of another syndrome**

Triggered or (unprovoked) spontaneous pathologic activation of MCs can result in the rapid onset of the typical symptoms of anaphylaxis. Patients experiencing clinical symptoms compatible with anaphylaxis in concomitance with a significant increase of tryptase (20% + 2 µg/L above the baseline tryptase, according to the international consensus formula [31]) fulfil the criterion of Mast Cell Activation Syndrome (MCAS). Primary MCAS include clonal MC disorders with quantitative and/or qualitative defects in MC biology such as systemic mastocytosis and monoclonal MC activation syndrome (MMAS). The latter defined by KIT mutation(s) (mainly KIT D816V) or aberrant CD25 expression without meeting the WHO criteria for systemic mastocytosis. In primary MCAS, several gain-of-function mutations in KIT lead to altered downstream signalling [32]. Secondary MCAS include

MC activation without an underlying clonal MC disease. Idiopathic MCAS are cases without clonal MC disorders and without an identifiable trigger. Another syndrome is the recently described hereditary alpha-tryptasemia (HaT). Hat can play an additive role in the relative risk for unprovoked or Hymenoptera induced anaphylaxis [33]. Recently, it was demonstrated that baseline serum tryptase (bST) > 8 µg/L is more frequently associated with the occurrence of anaphylaxis in perioperative hypersensitivity [34]. Consequently, studying the contribution of HaT to the severity of anaphylaxis and as an independent risk factor in perioperative settings will be interesting.

**Mast cell and basophil degranulation products**

Degranulation of MCs and basophils results in the release of inflammatory mediators. Mast cell mediator can be divided into three classes: 1) preformed (*i.e.* histamine, proteases), 2) new formed or lipid mediators generated within minutes (prostaglandins, leukotrienes, platelet-activating factor (PAF)), 3) de novo-synthetized mediators generated over hours (cytokines, chemokines and growth factors). It remains difficult and challenging to study the specific effect of each single mediator. However, evidence has accumulated that histamine, leukotrienes, and PAF are involved in vasodilation, capillary leak, and bronchospasm. According to the underlying trigger, the mediators can be released in a different spatiotemporal manner. IgE-dependent responses are prolonged and characterized by a slower but sustained calcium response, whereas MRGPRX2-mediated responses are often rapid, with a transient calcium response. In addition, IgE-dependent degranulation involves initial fusion of the granules before fusion with the cell membrane in limited numbers of openings. In contrast, MRGPRX2-mediated degranulation is characterized by direct release of individual, spherical granules over the entire MC membrane, a mechanism called “kiss and run. Regarding the released mediators, IgE-dependent stimulation triggers greater release of cytokines, prostaglandin E2 (PGE2), prostaglandin D2 (PGD2) and vascular endothelial growth factor (VEGF) while in contrast, the PGE2 and PGD2 response is limited and no VEGF is released after a MRGPRX2-mediated stimulation.[35, 36]. However, as indicated , MRGPRX2 signalling has mainly been studied pre-clinically in (knock-out) animal models and *in vitro* settings (reviewed in: [20, 21]), its precise mechanisms and clinical repercussions in the context of immediate drug hypersensitivity and anaphylaxis remains to be elucidated. Except for a higher skin MC responsivity and higher frequency of MC activation in the group with presumed sIgE/FcεRI-dependent rocuronium anaphylaxis, there were no differences with respect to clinic and tryptase between the sIgE/FcεRI-and MRGPRX2 group. Most importantly, acute and delta tryptase level cannot discriminate between the pathomechanistic processes [24]. An observation that was recently confirmed in an *in vitro* model [37]. Moreover, MCs of patients with sIgE/FcεRI-independent anaphylaxis to rocuronium had similar expression of MRGPRX2 and a similar functiinality as MCs from patients with sIgE/FcεRI-mediated anaphylaxis [38]. Alternatively, it is clear that the correct biochemical assessment of MC activation necessitates paired acute and baseline serum tryptase quantification [31, 34, 39]. Unlike a single measurement of acute serum tryptase, only paired analyses enable to depict other conditions such as clonal mast cell disease and HaT that might predesignate for anaphylaxis [40-42].

**Cytokine storm reaction**

Up to here, we described anaphylaxis mainly as the result of specific (allergic) and nonspecific (non-allergic or innate) activation of MCs and basophils and the activation of neutrophils with the release of various mediators acting on different target organs and cells. However, as elegantly reviewed by Castells [43] and Sala-Cunill et al

[44] another endotype with a distinct clinical signature has been described. As shown in Figure 1, these reactions, designated as cytokine storm reactions, result from the synthesis and release of mainly TNF-α, IL-1β and IL-6 by monocytes/macrophages and T-lymphocytes and are phenotypically characterized by fever and chills, rigor, nausea, vomiting, pain, followed by hypotension, desaturation and cardiovascular collapse. Triggers for such cytokine storms include chimeric, humanized, and human monoclonal antibodies and chemotherapy. Note that these compounds can also trigger mixed reactions combining the features of MC and basophil degranulation with cytokine storm-like reactions. Importantly, whatever the endotype, epinephrine and volume substitution are critical for correct acute treatment of severe reactions. Alternatively, it is obvious that cytokine storm-like reactions need other acute bio markers than paired tryptase quantification and will likely be overlooked by conventional tests developed to depict circulating (sIgE) antibodies and/or MC or basophil activation.

## Conclusion

Anaphylaxis is a complex, still incompletely understood, reaction that can involve various cell types, mediators, receptors, and intracellular signaling/degranulation pathways that can be activated alone or in association. The actualized definition of anaphylaxis includes the allergic and nonallergic mechanisms. Although the allergic pathway may lead to the release of mediators that are vasodilators and bronchoconstrictors, the nonallergic mechanisms can lead to the same heterogeneous clinical presentation through alternative modalities of MC and basophil dependent and independent processes such as potent COX-1 inhibition by NSAIDs and accumulation of bradykinin by ACE-inhibition. The allergist and immunologist should also be aware of non-classical masquerades such as cytokine storms triggered by biologicals and chemotherapy, as these reactions require alternative diagnostics and different treatments. Taken together, there is increasing evidence for alternative pathophysiological anaphylaxis endotypes that are phenotypically and biologically indistinguishable and are frequently difficult to document with conventional diagnostics.

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## References


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

[2] Castells MC. Drug Allergy: Phenotypes, Endotypes, and Biomarkers. J Allergy Clin Immunol Pract, 2017; 5: 626-627.

[3] Cardona V, Ansotegui IJ, Ebisawa M, El-Gamal Y, Fernandez Rivas M, Fineman S, Geller M, Gonzalez-Estrada A, Greenberger PA, Sanchez Borges M, Senna G, Sheikh A, Tanno LK, Thong BY, Turner PJ, Worm M. World allergy organization anaphylaxis guidance 2020. World Allergy Organ J, 2020; 13: 100472.

[4] Ebo DG, Clarke RC, Mertes PM, Platt PR, Sabato V, Sadleir PHM. Molecular mechanisms and pathophysiology of perioperative hypersensitivity and anaphylaxis: a narrative review. Br J Anaesth, 2019; 123: e38-e49.

[5] Xu H, Bin NR, Sugita S. Diverse exocytic pathways for mast cell mediators. Biochem Soc Trans, 2018; 46: 235-247.

[6] Knol EF. Requirements for effective IgE cross-linking on mast cells and basophils. Mol Nutr Food Res, 2006; 50: 620-4.

[7] Finkelman FD, Khodoun MV, Strait R. Human IgE-independent systemic anaphylaxis. J Allergy Clin Immunol, 2016; 137: 1674-1680.

[8] Reber LL, Hernandez JD, Galli SJ. The pathophysiology of anaphylaxis. J Allergy Clin Immunol, 2017; 140: 335-348.

[9] McLendon K, Sternard BT. Anaphylaxis. In: ed.^eds., StatPearls: Treasure Island (FL), 2021.

[10] Cheifetz A, Smedley M, Martin S, Reiter M, Leone G, Mayer L, Plevy S. The incidence and management of infusion reactions to infliximab: a large center experience. Am J Gastroenterol, 2003; 98: 1315-24.

[11] Steenholdt C, Svenson M, Bendtzen K, Thomsen O, Brynskov J, Ainsworth MA. Severe infusion reactions to infliximab: aetiology, immunogenicity and risk factors in patients with inflammatory bowel disease. Aliment Pharmacol Ther, 2011; 34: 51-8.

[12] Steenholdt C, Svenson M, Bendtzen K, Thomsen O, Brynskov J, Ainsworth MA. Acute and delayed hypersensitivity reactions to infliximab and adalimumab in a patient with Crohn's disease. J Crohns Colitis, 2012; 6: 108-11.

[13] Novey HS, Pahl M, Haydik I, Vaziri ND. Immunologic studies of anaphylaxis to iron dextran in patients on renal dialysis. Ann Allergy, 1994; 72: 224-8.

[14] Umeda Y, Fukumoto Y, Miyauchi T, Imaizumi M, Shimabukuro K, Mori Y, Takemura H. [Anaphylactic shock related to aprotinin induced by anti-aprotinin immunoglobulin G antibody alone; report of a case]. Kyobu Geka, 2007; 60: 69-71.

[15] Kober BJ, Scheule AM, Voth V, Deschner N, Schmid E, Ziemer G. Anaphylactic reaction after systemic application of aprotinin triggered by aprotinin-containing fibrin sealant. Anesth Analg, 2008; 107: 406-9.

[16] Kishimoto TK, Viswanathan K, Ganguly T, Elankumaran S, Smith S, Pelzer K, Lansing JC, Sriranganathan N, Zhao G, Galcheva-Gargova Z, Al-Hakim A, Bailey GS, Fraser B, Roy S, Rogers-Cotrone T, Buhse L, Whary M, Fox J, Nasr M, Dal Pan GJ, Shriver Z, Langer RS, Venkataraman G, Austen KF, Woodcock J, Sasisekharan R. Contaminated heparin associated with adverse clinical events and activation of the contact system. N Engl J Med, 2008; 358: 2457-67.

[17] Arroyave CM, Tan EM. Mechanism of complement activation by radiographic contrast media. Clin Exp Immunol, 1977; 29: 89-94.

[18] Tatemoto K, Nozaki Y, Tsuda R, Konno S, Tomura K, Furuno M, Ogasawara H, Edamura K, Takagi H, Iwamura H, Noguchi M, Naito T. Immunoglobulin E-independent activation of mast cell is mediated by Mrg receptors. Biochem Biophys Res Commun, 2006; 349: 1322-8.

[19] McNeil BD, Pundir P, Meeker S, Han L, Undem BJ, Kulka M, Dong X. Identification of a mast-cell-specific receptor crucial for pseudo-allergic drug reactions. Nature, 2015; 519: 237-41.
[20] Porebski G, Kwiecien K, Pawica M, Kwitniewski M. Mas-Related G Protein-Coupled Receptor-X2 (MRGPRX2) in Drug Hypersensitivity Reactions. Front Immunol, 2018; 9: 3027.
[21] Ali H. Revisiting the role of MRGPRX2 on hypersensitivity reactions to neuromuscular blocking drugs. Curr Opin Immunol, 2021; 72: 65-71.
[22] McNeil BD. MRGPRX2 and Adverse Drug Reactions. Front Immunol, 2021; 12: 676354.
[23] Kolkhir P, Ali H, Babina M, Ebo D, Sabato V, Elst J, Frischbutter S, Pyatilova P, Maurer M. MRGPRX2 in drug allergy: What we know and what we do not know. J Allergy Clin Immunol, 2022.
[24] Ebo DG, Van der Poorten ML, Elst J, Van Gasse AL, Mertens C, Bridts C, Garvey LH, Horiuchi T, Sabato V. Immunoglobulin E cross-linking or MRGPRX2 activation: clinical insights from rocuronium hypersensitivity. Br J Anaesth, 2021; 126: e27-e29.
[25] Blunk JA, Schmelz M, Zeck S, Skov P, Likar R, Koppert W. Opioid-induced mast cell activation and vascular responses is not mediated by mu-opioid receptors: an in vivo microdialysis study in human skin. Anesth Analg, 2004; 98: 364-70, table of contents.
[26] Babina M, Wang Z, Li Z, Franke K, Guhl S, Artuc M, Zuberbier T. FcepsilonRI- and MRGPRX2-evoked acute degranulation responses are fully additive in human skin mast cells. Allergy, 2022.
[27] Bruhns P, Chollet-Martin S. Mechanisms of human drug-induced anaphylaxis. J Allergy Clin Immunol, 2021; 147: 1133-1142.
[28] Jönsson F, de Chaisemartin L, Granger V, Gouel-Chéron A, Gillis CM, Zhu Q, Dib F, Nicaise-Roland P, Ganneau C, Hurtado-Nedelec M, Paugam-Burtz C, Necib S, Keita-Meyer H, Le Dorze M, Cholley B, Langeron O, Jacob L, Plaud B, Fischler M, Sauvan C, Guinnepain MT, Montravers P, Aubier M, Bay S, Neukirch C, Tubach F, Longrois D, Chollet-Martin S, Bruhns P. An IgG-induced neutrophil activation pathway contributes to human drug-induced anaphylaxis. Sci Transl Med, 2019; 11.
[29] Dona I, Salas M, Perkins JR, Barrionuevo E, Gaeta F, Cornejo-Garcia JA, Campo P, Torres MJ. Hypersensitivity Reactions to Non-Steroidal Anti-Inflammatory Drugs. Curr Pharm Des, 2016; 22: 6784-6802.
[30] Nassiri M, Babina M, Dolle S, Edenharter G, Rueff F, Worm M. Ramipril and metoprolol intake aggravate human and murine anaphylaxis: evidence for direct mast cell priming. J Allergy Clin Immunol, 2015; 135: 491-9.
[31] Valent P, Akin C, Arock M, Brockow K, Butterfield JH, Carter MC, Castells M, Escribano L, Hartmann K, Lieberman P, Nedoszytko B, Orfao A, Schwartz LB, Sotlar K, Sperr WR, Triggiani M, Valenta R, Horny HP, Metcalfe DD. 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: 215-25.
[32] Cruse G, Metcalfe DD, Olivera A. Functional deregulation of KIT: link to mast cell proliferative diseases and other neoplasms. Immunol Allergy Clin North Am, 2014; 34: 219-37.
[33] Lyons JJ, Chovanec J, O'Connell MP, Liu Y, Selb J, Zanotti R, Bai Y, Kim J, Le QT, DiMaggio T, Schwartz LB, Komarow HD, Rijavec M, Carter MC, Milner JD, Bonadonna P, Metcalfe DD, Korosec P. Heritable risk for severe anaphylaxis associated with increased alpha-tryptase-encoding germline copy number at TPSAB1. J Allergy Clin Immunol, 2021; 147: 622-632.
[34] Ebo DG, De Puysseleyr LP, Van Gasse AL, Elst J, Poorten MV, Faber MA, Mertens C, Van Houdt M, Hagendorens MM, Sermeus L, Vitte J, Moise M, Garvey LH, Castells MC, Tacquard C, Mertes PM, Schwartz LB, Sabato V. Mast Cell Activation During Suspected Perioperative Hypersensitivity: A Need for Paired Samples Analysis. J Allergy Clin Immunol Pract, 2021; 9: 3051-3059.e1.
[35] Karhausen J, Abraham SN. How mast cells make decisions. J Clin Invest, 2016; 126: 3735-3738.

[36] Gaudenzio N, Sibilano R, Marichal T, Starkl P, Reber LL, Cenac N, McNeil BD, Dong X, Hernandez JD, Sagi-Eisenberg R, Hammel I, Roers A, Valitutti S, Tsai M, Espinosa E, Galli SJ. Different activation signals induce distinct mast cell degranulation strategies. J Clin Invest, 2016; 126: 3981-3998.
[37] Elst J, van der Poorten MM, Van Gasse AL, Mertens C, Hagendorens MM, Ebo DG, Sabato V. Tryptase release does not discriminate between IgE- and MRGPRX2-mediated activation in human mast cells. Clin Exp Allergy, 2022.
[38] Elst J, Maurer M, Sabato V, Faber MA, Bridts CH, Mertens C, Van Houdt M, Van Gasse AL, van der Poorten MM, De Puysseleyr LP, Hagendorens MM, Van Tendeloo VF, Lion E, Campillo-Davo D, Ebo DG. Novel Insights on MRGPRX2-Mediated Hypersensitivity to Neuromuscular Blocking Agents And Fluoroquinolones. Front Immunol, 2021; 12: 668962.
[39] Vitte J, Sabato V, Tacquard C, Garvey LH, Michel M, Mertes PM, Ebo DG, Schwartz LB, Castells MC. Use and Interpretation of Acute and Baseline Tryptase in Perioperative Hypersensitivity and Anaphylaxis. J Allergy Clin Immunol Pract, 2021; 9: 2994-3005.
[40] Sabato V, Van De Vijver E, Hagendorens M, Vrelust I, Reyniers E, Fransen E, Bridts C, De Clerck L, Mortier G, Valent P, Ebo D. Familial hypertryptasemia with associated mast cell activation syndrome. J Allergy Clin Immunol, 2014; 134: 1448-1450 e3.
[41] Sabato V, Chovanec J, Faber M, Milner JD, Ebo D, Lyons JJ. First Identification of an Inherited TPSAB1 Quintuplication in a Patient with Clonal Mast Cell Disease. J Clin Immunol, 2018; 38: 457-459.
[42] Lyons JJ. Hereditary Alpha Tryptasemia: Genotyping and Associated Clinical Features. Immunol Allergy Clin North Am, 2018; 38: 483-495.
[43] Castells M. Diagnosis and management of anaphylaxis in precision medicine. J Allergy Clin Immunol, 2017; 140: 321-333.
[44] Sala-Cunill A, Guilarte M, Cardona V. Phenotypes, endotypes and biomarkers in anaphylaxis: current insights. Curr Opin Allergy Clin Immunol, 2018; 18: 370-376.
[45] Van Gasse AL, Ebo DG, Faber MA, Elst J, Hagendorens MM, Bridts CH, Mertens CM, De Clerck LS, Romano A, Sabato V. Cross-reactivity in IgE-mediated allergy to cefuroxime: Focus on the R1 side chain. J Allergy Clin Immunol Pract, 2020; 8: 1094-1096 e1.
[46] van der Poorten MM, Van Gasse AL, Hagendorens MM, Faber MA, De Puysseleyr L, Elst J, Mertens CM, Sabato V, Ebo DG. Serum specific IgE antibodies in immediate drug hypersensitivity. Clin Chim Acta, 2020; 504: 119-124.
[47] Ebo DG, Bridts CH, Rihs HP. Hevea latex-associated allergies: piecing together the puzzle of the latex IgE reactivity profile. Expert Rev Mol Diagn, 2020; 20: 367-373.
[48] Ebo DG, Bridts CH, Stevens WJ. IgE-mediated anaphylaxis from chlorhexidine: diagnostic possibilities. Contact Dermatitis, 2006; 55: 301-2.
[49] Elst J, Moonen N, van der Poorten MM, Faber MA, Van Gasse AL, Garvey LH, Bridts CH, De Puysseleyr LP, Mertens C, Hagendorens MM, Sabato V, Ebo DG. The passively sensitized mast cell activation test is a reliable diagnostic for chlorhexidine allergy. J Allergy Clin Immunol Pract, 2021; 9: 3826-3828 e2.
[50] Ebo DG, Venemalm L, Bridts CH, Degerbeck F, Hagberg H, De Clerck LS, Stevens WJ. Immunoglobulin E antibodies to rocuronium: a new diagnostic tool. Anesthesiology, 2007; 107: 253-9.
[51] Leysen J, De Witte L, Sabato V, Faber M, Hagendorens M, Bridts C, De Clerck L, Ebo D. IgE-mediated allergy to pholcodine and cross-reactivity to neuromuscular blocking agents: Lessons from flow cytometry. Cytometry B Clin Cytom, 2013; 84: 65-70.
[52] Pinnobphun P, Buranapraditkun S, Kampitak T, Hirankarn N, Klaewsongkram J. The diagnostic value of basophil activation test in patients with an immediate hypersensitivity reaction to radiocontrast media. Ann Allergy Asthma Immunol, 2011; 106: 387-93.
[53] Brockow K, Romano A, Aberer W, Bircher AJ, Barbaud A, Bonadonna P, Faria E, Kanny G, Lerch M, Pichler WJ, Ring J, Rodrigues Cernadas J, Tomaz E, Demoly P, Christiansen C, European

Network of Drug A, the Eigodh. Skin testing in patients with hypersensitivity reactions to iodinated contrast media - a European multicenter study. Allergy, 2009; 64: 234-41.
[54] Salas M, Gomez F, Fernandez TD, Dona I, Aranda A, Ariza A, Blanca-Lopez N, Mayorga C, Blanca M, Torres MJ. Diagnosis of immediate hypersensitivity reactions to radiocontrast media. Allergy, 2013; 68: 1203-6.
[55] Stone CA, Jr., Liu Y, Relling MV, Krantz MS, Pratt AL, Abreo A, Hemler JA, Phillips EJ. Immediate Hypersensitivity to Polyethylene Glycols and Polysorbates: More Common Than We Have Recognized. J Allergy Clin Immunol Pract, 2019; 7: 1533-1540 e8.
[56] Zhou ZH, Stone CA, Jr., Jakubovic B, Phillips EJ, Sussman G, Park J, Hoang U, Kirshner SL, Levin R, Kozlowski S. Anti-PEG IgE in anaphylaxis associated with polyethylene glycol. J Allergy Clin Immunol Pract, 2021; 9: 1731-1733 e3.
[57] Alvarez-Arango S, Yerneni S, Tang O, Zhou L, Mancini CM, Blackley SV, Keet CA, Blumenthal KG. Vancomycin Hypersensitivity Reactions Documented in Electronic Health Records. J Allergy Clin Immunol Pract, 2021; 9: 906-912.
[58] Cesana P, Scherer K, Bircher AJ. Immediate Type Hypersensitivity to Heparins: Two Case Reports and a Review of the Literature. Int Arch Allergy Immunol, 2016; 171: 285-289.
[59] Anders D, Trautmann A. Allergic anaphylaxis due to subcutaneously injected heparin. Allergy Asthma Clin Immunol, 2013; 9: 1.
[60] Hamad I, Hunter AC, Szebeni J, Moghimi SM. Poly(ethylene glycol)s generate complement activation products in human serum through increased alternative pathway turnover and a MASP-2-dependent process. Mol Immunol, 2008; 46: 225-32.
[61] Merkel OM, Urbanics R, Bedocs P, Rozsnyay Z, Rosivall L, Toth M, Kissel T, Szebeni J. In vitro and in vivo complement activation and related anaphylactic effects associated with polyethylenimine and polyethylenimine-graft-poly(ethylene glycol) block copolymers. Biomaterials, 2011; 32: 4936-42.
[62] Elst J, Sabato V, Faber MA, Bridts CH, Mertens C, Van Houdt M, Van Gasse AL, Hagendorens MM, Van Tendeloo V, Maurer M, Campillo-Davo D, Timmermans JP, Pintelon I, Ebo DG. MRGPRX2 and Immediate Drug Hypersensitivity: Insights From Cultured Human Mast Cells. J Investig Allergol Clin Immunol, 2021; 31: 489-499.
[63] Chompunud Na Ayudhya C, Amponnawarat A, Roy S, Oskeritzian CA, Ali H. MRGPRX2 Activation by Rocuronium: Insights from Studies with Human Skin Mast Cells and Missense Variants. Cells, 2021; 10.
[64] Fukuoka Y, Schwartz DL, Ward BR. Activation of human skin mast cells by vancomycin via MrgX2: Comparison to the effects of brimonidine. The Journal of Immunology, 2021; 206: 23.10-23.10.
[65] Sarkar P, Nicholson G, Hall G. Brief review: angiotensin converting enzyme inhibitors and angioedema: anesthetic implications. Can J Anaesth, 2006; 53: 994-1003.
[66] Stojiljkovic L. Renin-angiotensin system inhibitors and angioedema: anesthetic implications. Curr Opin Anaesthesiol, 2012; 25: 356-62.

454 **Table and legends of figures**

| **Mechanisms of immediate hypersensitivity and anaphylaxis** | | | | |
|---|---|---|---|---|
| **Specific, antibody-dependent, effector cell activation/degranulation** | | | | |
| **Type** | **Effector cells** | **Receptor** | **Causing agents (not exhaustive)** | **Comments** |
| sIgE-mediated | Basophils<br>Mast cells | FcεRI | B-lactams [45, 46]<br>*Hevea* Latex [47]<br>Chlorhexidine [48, 49]<br>NMBAs [50]<br>Opioids [51]<br>Iodinated contrast media [52-54]<br>PEG [55, 56]<br>Vancomycin [57]<br>Heparin ** [58, 59] | Some compounds can also trigger effector cell degranulation by alternative processes (as indicated below e.g. by activation of MRGPRX2). |
| IgG-mediated | Basophils<br>Mast cells | FcγR | Infliximab [10, 11]<br>Adalimumab [12]<br>Dextrans [13]<br>Aprotinin [14, 15] | Some observations remains debatable as drug sIgE was still detectable. |
| IgG-mediated | Platelets<br>Neutrophils | FcγR | NMBAs [27, 28] | Human evidence is restricted to a single report on IgG-dependent neutrophil activation in response to NMBAs. |
| **Non-specific effector cell activation/degranulation** | | | | |
| **Type** | **Effector cells** | **Receptor** | **Causing agents** | **Comments** |
| Complement-mediated | Basophils<br>Mast cells | C3aR<br>C5aR | Iodinated contrast media [17]<br>PEG [60, 61]<br>Heparin ** [16] | |

| | | | | |
|---|---|---|---|---|
| MRGPRX2-mediated | Mast cells | MRGPRX2 | NMBAs [38, 62, 63]<br>Fluoroquinolones [38, 62]<br>Opiates [62]<br>Vancomycin [64]<br>Icatibant [36] | Human literature is limited and clinical evidence is still lacking. Most data is gathered in animal models or transfected cell lines. |
| **Effector cell independent** | | | | |
| **Type** | | | **Causing agents** | **Mechanism** |
| Enzyme interference | | | NSAIDS [29] | Inhibition of COX leading to a blocking of prostaglandin synthesis and increased production of leukotrienes. |
| Enzyme interference | | | ACE-inhibitors [65, 66] | Blocking the degradation of bradykinin. |
| Cytokine-storm-reactions | | | Monoclonal antibodies (e.g. rituximab) [43, 44]<br>Chemotherapy [43, 44] | Cytokine release by monocytes/macrophages and T-lymphocytes. |
| *** In supratherapeutic concentrations**<br>**** oversulfated chondroitin sulfate can activate complement and factor XII (bradykinin)**<br>**NMBAs, neuromuscular blocking agents; PEG, polyethylene Glycol; NSAIDs, nonsteroidal anti-inflammatory drugs; ACE, angiotensin-converting enzyme** | | | | |

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**Figure 1: Release of cytokines by monocytes/macrophages and T-lymphocytes**

Cytokine storm reactions, result from the synthesis and release of mainly TNF-α, IL-1β and IL-6 by monocytes/macrophages and T-lymphocytes. Triggers for these reactions include chimeric, humanized, and human monoclonal antibodies and chemotherapy.