Rostrum

Human IgE-independent systemic anaphylaxis

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Anaphylaxis is a rapidly developing, life-threatening, generalized or systemic allergic reaction that is classically elicited by antigen crosslinking of antigen-specific IgE bound to the high-affinity IgE receptor FcεRI on mast cells and basophils. This initiates signals that induce cellular degranulation with release and secretion of vasoactive mediators, enzymes, and cytokines. However, IgE-independent mechanisms of anaphylaxis have been clearly demonstrated in experimental animals. These include IgG-dependent anaphylaxis, which involves the triggering of mediator release by IgG/antigen complex crosslinking of FcγRs on macrophages, basophils, and neutrophils; anaphylaxis mediated by binding of the complement-derived peptides C3a and C5a to their receptors on mast cells, basophils, and other myeloid cells; and direct activation of mast cells by drugs that interact with receptors on these cells. Here we review the mechanisms involved in these IgE-independent forms of anaphylaxis and the clinical evidence for their human relevance. We conclude that this evidence supports the existence of all 3 IgE-independent mechanisms as important causes of human disease, although practical and ethical considerations preclude their demonstration to the degree of certainty possible with animal models. Furthermore, we cite evidence that different clinical situations can suggest different mechanisms as having a primal role in anaphylaxis and that IgE-dependent and distinct IgE-independent mechanisms can act together to increase anaphylaxis severity. As specific agents become available that can interfere with mechanisms involved in the different types of anaphylaxis, recognition of specific types of anaphylaxis is likely to become important for optimal prophylaxis and therapy. (J Allergy Clin Immunol 2016;■■■:■■■-■■■.)

Key words: *Anaphylatoxin, complement, FcεR, FcγR, IgE, IgG, mast cell, basophil, mouse*

Abbreviations used
NSAID: Nonsteroidal anti-inflammatory drug
PAF: Platelet-activating factor
TNP: Trinitrophenyl

Anaphylaxis is a rapidly developing, life-threatening, generalized or systemic allergic reaction.[1] Foods, drugs, and insect stings are the most common causes of this disorder.[2] Classically, anaphylaxis is induced by antigen crosslinking of antigen-specific IgE that has bound to the high-affinity IgE receptor (FcεRI) on mast cells and basophils.[3] Crosslinking of IgE and its receptor induces a signaling cascade that results in mast cell degranulation with release of mediators, including histamine, as well as preformed cytokines and proteases, and synthesis and secretion of additional cytokines, as well as lipid mediators, such as platelet-activating factor (PAF), leukotrienes, and prostaglandins.[4] Passive immunization studies in which mice were sensitized by injecting an antigen-specific IgE antibody, followed by enteral or parenteral exposure to that antigen, support the importance of IgE and mast cells in antigen-induced shock.[5] Indeed, both passive and active immunization studies in which mice were challenged orally with the appropriate antigen have generally demonstrated that genetic or antibody elimination of IgE, mast cells, or the IgE-binding chain of FcεRI (FcεRIα) completely suppresses anaphylaxis development.[6-8] In contrast, studies in which mice were actively immunized with an antigen, followed by parenteral challenge with the same antigen, have often revealed that anaphylaxis can occur in the absence of the classical IgE/FcεRI/mast cell pathway and demonstrated that a disorder that closely resembles IgE-mediated systemic anaphylaxis can be mediated by mechanisms that involve IgG rather than IgE.[9-11] Consistent with this, mice that are passively immunized with an IgG_1, IgG_{2a}, or IgG_{2b} (but not IgG_3) mAb specific for the hapten trinitrophenyl (TNP) have anaphylaxis, which is nearly indistinguishable clinically from IgE-mediated anaphylaxis, when challenged parenterally but not enterally with a TNP-protein conjugate.[5,6,8] These observations, coupled with several human clinical observations, suggest that IgE-independent anaphylaxis might be clinically important. Here we will first review observations that prove the existence of IgG-mediated anaphylaxis in mice and describe differences in the mechanisms behind the classical IgE-mediated pathway and the alternative IgG-mediated pathway in this species, as well as the clinical implications of these differences. Next, we will review observations that support the existence of IgG-mediated anaphylaxis in human subjects, as well as the implications and limitations of these observations. Finally, we will discuss the evidence and its limitations for other antibody-independent mechanisms of anaphylaxis in both mice and human subjects.

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MURINE EVIDENCE FOR IgG-MEDIATED ANAPHYLAXIS

Evidence for IgE-independent, IgG-dependent anaphylaxis was provided by studies in which mice were immunized and then parenterally challenged with a potent antigen.[11] In some of these active immunization models, disease developed even if mice were first treated with an anti-IgE mAb but was suppressed if mice were instead treated with the rat IgG_{2b} mAb 2.4G2. This mAb binds to and triggers but then blocks the inhibitory low-affinity IgG receptor FcγRIIB and the stimulatory low-affinity IgG receptor FcγRIII and indirectly blocks the other murine FcγRs, FcγRI and FcγRIV.[11,12] The existence of IgE-independent anaphylaxis in actively immunized mice was demonstrated most conclusively by studies that (1) induced severe anaphylaxis in actively immunized IgE- or FcγRIα–deficient mice but not in actively immunized mice that lacked all stimulatory FcRs (ie, FcRγ-deficient mice) and (2) demonstrated reduced severity or absence of anaphylaxis in actively immunized mice that lacked function of 1 or more of the stimulatory murine FcγRs.[11-14]

Subsequent passive immunization studies demonstrated that an anti-IgE mAb would block anaphylaxis when mice were sensitized with an antigen-specific IgE mAb but not when mice were sensitized with an antigen-specific IgG_1, IgG_{2a}, or IgG_{2b} mAb, whereas reciprocal results were found when passively immunized mice were treated with 2.4G2.[10-12,15,16] The severity of systemic anaphylaxis in these IgG passive immunization models was normal or increased in mice deficient in FcεRIα.[13] In contrast, anaphylaxis in mice passively sensitized with an antigen-specific IgG_1 mAb was totally absent in mice deficient in FcγRIII (the only stimulatory murine FcγR that binds mouse IgG_1), whereas total suppression of anaphylaxis in mice sensitized with an IgG_{2a} mAb (which binds to all 3 stimulatory murine FcγRs) required deletion or blocking of all of these receptors.[10,12] The importance of FcγRs in murine IgG-dependent anaphylaxis was also shown by the unique inability of IgG_3, among the murine IgG isotypes, to mediate anaphylaxis, which correlates with the observation that IgG_3 is the only murine IgG isotype that does not bind to any stimulatory murine FcγR.[17,18]

CLINICAL IMPLICATIONS OF STUDIES OF MURINE IgG-MEDIATED ANAPHYLAXIS

Studies of murine IgG-mediated anaphylaxis by several groups have evaluated the mediators involved, the responsible cell types, and the quantities of antigen required to induce shock. Nearly all studies have identified PAF, rather than histamine, as the mediator most important in IgG-mediated anaphylaxis in actively immunized mice,[11,19,20] although this has not been investigated thoroughly in passively immunized mice. In contrast to agreement about the importance of PAF in IgG-mediated anaphylaxis in actively immunized mice, different studies have identified monocytes/macrophages, basophils, or neutrophils as the critical cell type in IgG-mediated anaphylaxis.[11,19,20] All of these cell types express FcγRIII and FcγRIV in mice, and all are capable of producing PAF in response to appropriate stimuli.[18,20-24] Differences in cell types that appear to be responsible for IgG-mediated anaphylaxis can result from differences in mouse strains used, stimuli that elicit anaphylaxis, endogenous bacterial flora, and/or animal husbandry practices.

Results of studies that compared the doses of antigen required to induce IgE- versus IgG_1-mediated anaphylaxis suggest that the dose of challenge antigen determines when IgG-mediated anaphylaxis can occur. In mice that were passively sensitized with high-affinity IgE or IgG antibodies to TNP, 100- to 1000-fold less TNP-conjugated protein was required to induce shock in IgE- than in IgG-sensitized mice.[15] This was true regardless of the extent of TNP labeling of the TNP-conjugated protein, although less TNP conjugate was required to induce either IgE- or IgG-mediated anaphylaxis when the protein was heavily labeled.[15] These observations are consistent, respectively, with the much higher affinity of FcεRI than FcγRIII, the much higher ratio of cell-bound to serum IgE than IgG, and the better cross-linking of an antigen-specific mAb by an antigen that has multiple copies of the epitope bound by that mAb.

Because IgG-mediated anaphylaxis requires a much larger dose of antigen than IgE-mediated anaphylaxis, anaphylaxis induced by means of parenteral administration of a small quantity of antigen (eg, insect sting) is much more likely to be IgE mediated. Similarly, anaphylaxis induced by antigen ingestion (eg, food allergy) always appears to be IgE mediated[6,7] because induction of anaphylaxis in food allergy models requires systemic absorption of ingested antigen and only a very small percentage of ingested antigen is absorbed with all epitopes intact.[7,8] In contrast, both IgE- and IgG-mediated anaphylaxis can be induced by parenteral administration of a relatively large quantity of antigen (eg, infusion of a therapeutic antibody or drug),[15] particularly an antigen that has multiple iterations of an antibody-reactive epitope (eg, a carbohydrate antigen, such as dextran).

The difference in antigen dose requirement for IgE- versus IgG-mediated anaphylaxis allows IgG to act both as a mediator of anaphylaxis and a blocker of IgE-mediated anaphylaxis, depending on antibody and antigen concentrations (Fig 1). In the presence of antigen-specific IgE, antigen-specific IgG antibody will block anaphylaxis that would otherwise be induced by a low dose of antigen by intercepting antigen before it can bind to mast cell–associated IgE and by interacting with the inhibitory receptor FcγRIIB (Fig 1, *A* and *B*)[15,25] but mediate anaphylaxis induced by a higher antigen dose (Fig 1, *C* and *D*). The ability of IgG to both block antigen access to mast cell–associated IgE and to mediate anaphylaxis through IgG/antigen complex binding to stimulatory FcγRs can create the counterintuitive situation in which an intermediate dose of antigen will induce IgG- but not IgE-mediated anaphylaxis in the presence of both antigen-specific IgE and IgG (Fig 1, *C*).[15]

LIMITATIONS OF STUDIES OF MOUSE IgG-MEDIATED ANAPHYLAXIS

Although experimental evidence that IgG-mediated anaphylaxis can occur in mice is unequivocal, there are concerns about the interpretation of studies that identify the importance of different cells and receptors in this process. Nearly all studies that analyze the importance of specific receptors use gene deletion to eliminate specific FcγRs or antibodies to block these receptors, whereas studies that analyze the importance of specific cell types either use antibodies or drugs that eliminate these cell types or transfer a specific cell type to a recipient mouse. Although gene deletion can cause complete deficiency of a specific receptor, the elimination of one receptor under at least some circumstances can increase the expression, signaling capacity, or both of the remaining receptors.[15,24] This can lead investigators to exaggerate the importance of the remaining receptors. IgG mAbs to

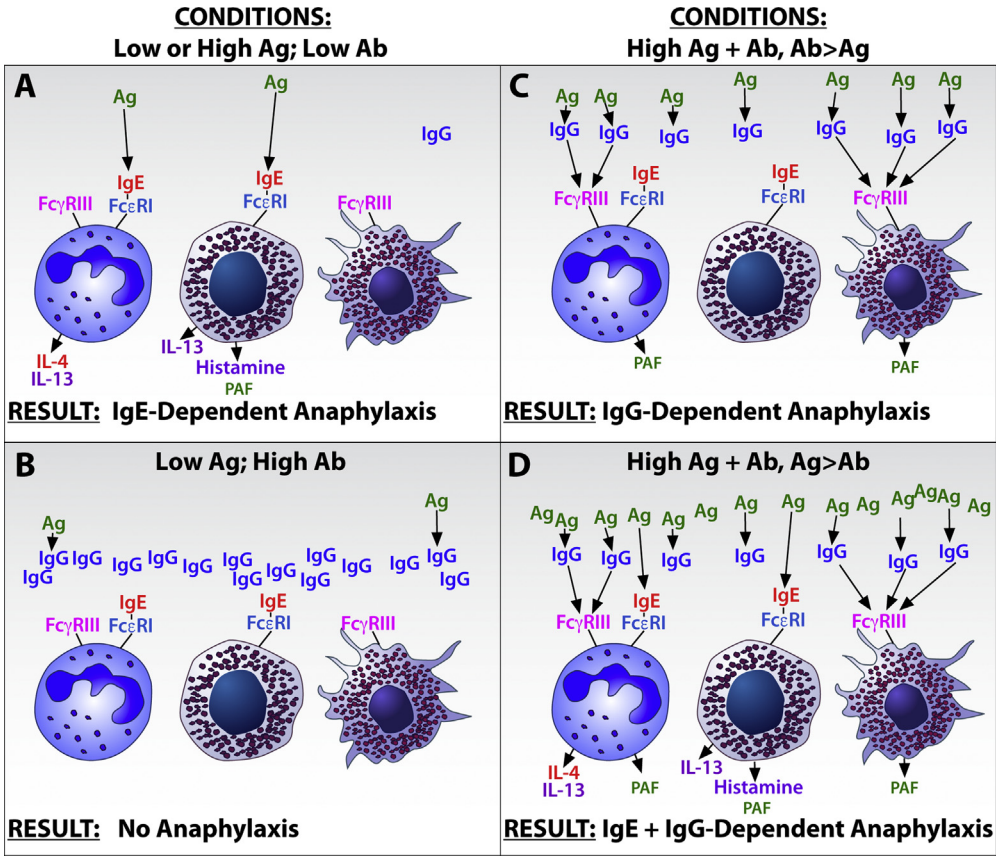

FIG 1. Relative concentrations of antigen *(Ag)* and antibody *(Ab)* determine the roles of IgE and IgG antibodies in the setting of antibody-mediated anaphylaxis. IgE-mediated anaphylaxis requires considerably less antibody and antigen than IgG-mediated anaphylaxis. Consequently, when antibody levels are low **(A)**, only IgE-mediated anaphylaxis can occur. When antigen levels are low but antibody levels are high **(B)**, IgG "blocking" antibodies prevent IgE-mediated anaphylaxis by intercepting antigen before it can bind to FcεRI-associated IgE and by binding to the inhibitory receptor FcγRIIB, but the quantity of IgG/antigen complexes is too low to trigger IgG-mediated anaphylaxis. Consequently, anaphylaxis does not occur. When antigen and antibody levels are both high but antibody levels are in excess to antigen levels **(C)**, IgG antibodies block the binding of antigen to FcεRI-bound IgE, but IgG/antigen complexes can bind to FcγRs; consequently, only IgG-mediated anaphylaxis occurs. When antigen and antibody levels are both high but antigen levels are in excess **(D)**, IgG/antibody complexes are sufficient to trigger IgG-mediated anaphylaxis, and enough antigen escapes IgG blockade to bind to FcεRI-associated IgE and trigger IgE-mediated anaphylaxis.

a specific receptor might influence nontargeted receptors by signaling through the targeted receptor, binding of the Fc part of the IgG mAb to nontargeted receptors (eg, mAb 2.4G2, which binds to FcγRIIB and FcγRIII, at least partially suppresses the expression and function of FcγRI and FcγRIV[12]), or both. Techniques used to delete macrophages, such as silica, clodronate liposomes, and gadolinium, can activate these cells before killing them[26]; cytokines or mediators produced by the activated cells might influence the ability of the remaining cell types to contribute to anaphylaxis. Antibodies used to delete specific cell types, including neutrophils, basophils, and platelets, can activate and deplete complement on binding the targeted cell type or form immune complexes with cell membrane antigens that bind to FcγRs on other cells; both can influence the ability of these other cells to contribute to anaphylaxis. In this regard, for example, we have found that IgG-mediated anaphylaxis is suppressed when platelets are eliminated with IgG anti-platelet antibodies but not when platelets are eliminated with neuraminidase.[11] Antibodies that appear to eliminate a cell type, when that cell type is studied in one organ (eg, blood), might actually cause redistribution of that cell type to another organ (eg, spleen). Finally, because cells, such as neutrophils, can be partially activated by using *in vitro* purification procedures, it is possible that transfer studies with purified neutrophils[19] exaggerate the importance of this cell type in IgG-mediated anaphylaxis. Although none of these techniques are used to evaluate whether IgG-mediated anaphylaxis exists in human subjects, concerns about their use in mice affects hypotheses about which receptors and cell types contribute to putative human IgG-mediated anaphylaxis.

THEORETICAL CONSIDERATIONS ABOUT THE POSSIBILITY OF HUMAN IgG-MEDIATED ANAPHYLAXIS

Because the kinds of experiments that have proved the existence of IgG-mediated anaphylaxis in mice would not be appropriate in human subjects, even when possible, evidence

for human IgE-mediated anaphylaxis is typically anecdotal and correlative rather than definitive. In addition, important differences between mice and human subjects, including differences in the properties of their IgG isotypes, differences in their FcγRs, differences in cellular FcγR distribution, and differences in the properties of FcγR-expressing cells themselves, raise questions about the applicability of observations about IgG-mediated anaphylaxis in mice to human subjects. However, consideration of each of these differences does not provide a reason for thinking that human IgG-mediated anaphylaxis is unlikely.

Human IgG_1 and IgG_3 and possibly IgG_4 bind to human FcγRs, with an affinity range similar to what is observed in mice.[24] Although human subjects lack FcγRIV, they express FcγRI and FcγRIII and have the stimulatory FcγRs FcγRIIA and FcγRIIC, which are not present in mice.[24] Activation of human basophils, monocyte/macrophages, and neutrophils can cause these cells to produce PAF, which has been associated with human anaphylaxis.[27,28] In addition, as noted earlier, human neutrophils can mediate IgG-dependent anaphylaxis when infused into mice.[19]

Differences in cellular FcγR expression can actually make it more likely for IgG to mediate anaphylaxis in human subjects than in mice because although both human and mouse mast cells express a stimulatory FcγR (FcγRIII in mice and FcγRIIA and possibly FcγRIIC in human subjects), human mast cells express relatively little or no inhibitory FcγRIIB, whereas mouse mast cells express relatively large amounts of this receptor.[24,29,30] Similarly, the much larger number of granules in human than in murine basophils[31] makes degranulation of these cells more likely to induce anaphylaxis in the former species. Taken together, there is no reason to believe that differences in IgG isotypes, FcγRs, cellular distribution of these receptors, or the physiology of FcγR-expressing cells make IgG less likely to mediate anaphylaxis in human subjects than in mice.

CLINICAL EVIDENCE FOR HUMAN IgG-MEDIATED ANAPHYLAXIS

Several clinical observations support the importance of IgG-mediated anaphylaxis in human subjects, although each of these observations is open to alternate conclusions. Multiple researchers have described anaphylaxis in patients who were treated with a biologic therapeutic and had IgG but not detectable IgE antibody that was specific for that therapeutic.[32] This has been reported in transfused and intravenous immunoglobulin-treated IgA-deficient subjects (who had IgG anti-IgA antibodies)[33]; subjects treated with a variety of chimeric, humanized, and even fully human mAbs[34]; subjects treated with dextran[35] or aprotinin[36]; and von Willebrand factor–deficient subjects who have been infused with von Willebrand factor.[37] It is noteworthy that all of these examples of putative IgG-mediated anaphylaxis involve the parenteral administration of a large quantity of an antigen, precisely the condition that favors IgG-mediated anaphylaxis in mice. However, it remains possible that the subjects who had anaphylaxis might have had cell-bound, FcεRI-associated, therapeutic-specific IgE without detectable antigen-specific IgE in serum. This is possible because the high-affinity FcεRI for IgE and the relatively low level of FcεRI crosslinking required to induce mast cell degranulation allow sufficient IgE to bind to mast cells to mediate their activation, even when serum IgE levels are very low. Similarly, other evidence that supports the existence of IgE-independent anaphylaxis, such as anaphylaxis without evidence of basophil activation, anaphylaxis in the absence of increased serum tryptase levels,[38] and anaphylaxis in subjects with negative skin test results might be explained by a lack of sensitivity of the tests used, the small time window in which a test reflective of IgE-mediated anaphylaxis remains positive, or restrictions in the location and properties of mast cells responsible for IgE-mediated anaphylaxis (eg, antigen-specific IgE might be bound to vascular mast cells but not to skin mast cells).

Additional evidence in favor of IgG-mediated anaphylaxis comes from a study of subjects treated with the chimeric mAb infliximab, which demonstrated that the presence of IgG anti-infliximab antibody levels of 8 μg/mL or greater was associated with a relative risk of anaphylaxis of 2.4.[39,40] Although this association suggests that IgG might have been involved in anaphylaxis pathogenesis, it is also possible that higher IgG antibody levels were a marker for higher IgE antibody levels.

Because PAF is more strongly associated with IgG- than IgE-mediated anaphylaxis in mice, reports that serum PAF levels are higher in patients undergoing anaphylaxis than in a control group of patients and that serum concentrations of PAF acetylhydrolase, the enzyme that breaks down PAF, correlate inversely with anaphylaxis severity,[27,28] are also consistent with the existence of human IgG-mediated anaphylaxis. However, because PAF can also be produced by mast cells and basophils in response to IgE crosslinking[41,42] and because human myeloid cells other than mast cells and basophils can express FcεRIα,[24] these observations might instead reflect a role for PAF in human IgE-mediated anaphylaxis. This alternative explanation is somewhat refuted by evidence that human neutrophils, monocyte/macrophages, and basophils can produce PAF in response to FcγR crosslinking and that human neutrophils can mediate anaphylaxis in mice[19]; however, it could be argued that there is no direct evidence that the amount of PAF or other mediators produced by these cells is sufficient to induce anaphylaxis in human subjects.

One last intriguing piece of evidence in favor of human IgG-mediated anaphylaxis comes from a study demonstrating increased frequency of a gain-of-function allele of the stimulatory FcγR FcγRIIA in patients with common variable immunodeficiency who have IgG anti-IgA antibodies and anaphylaxis after intravenous immunoglobulin infusion.[43] However, the effect of this elegant work is limited by the small number of patients studied, the possibility that the mechanism that associates increased FcγRIIA activity with anaphylaxis might be indirect (eg, increased FcγRIIA function might promote an IgE response), and the lack of other reported associations of FcγR polymorphisms with human anaphylaxis.

Taken together, these observations appear to make the existence of human IgG-mediated anaphylaxis highly likely, particularly when anaphylaxis occurs in the presence of relatively high titers of specific IgG antibody and undetectable specific IgE in subjects who have been injected or infused with relatively large quantities of the recognized antigen. However, we concede that these observations do not provide absolute proof of the existence or clinical importance of human IgG-mediated anaphylaxis.

MURINE COMPLEMENT-MEDIATED ANAPHYLAXIS

Studies in mice demonstrate that C3a and C5a, small peptides derived from C3 and C5, respectively, and known as anaphylatoxins, can activate mast cells and other myeloid cells[44]; however, there is a lack of convincing evidence that they are either required

for antibody-mediated anaphylaxis or can produce shock in the absence of other factors in this species. Passive anaphylaxis studies have demonstrated that ligation of mast cell C3a or C5a receptor is required for the induction of skin swelling by injected C3a and C5a, respectively, and that both anaphylatoxins stimulate mast cell degranulation.[45] Studies in which mice were injected with soluble peanut extract demonstrated that components of that extract activated complement through both the classical and the lectin pathways.[46,47] C3a produced in this manner could stimulate hypothermia through a macrophage- and PAF-dependent process but only when mice were treated with a β-adrenergic antagonist, a long-acting formulation of IL-4, or both to increase their responsiveness to PAF.[46] Perhaps more importantly, complement activation by peanut extract acted synergistically with IgE-mediated mast cell activation to cause shock in the absence of exogenous β-adrenergic antagonist or IL-4.[46] This suggested that combined IgE-dependent mast cell activation and complement activation by peanuts might be one explanation for the severity of peanut-induced anaphylaxis. The suggestion that complement activation exacerbates anaphylaxis induced by other mechanisms but is insufficient to independently induce murine systemic anaphylaxis is also consistent with the inability of antigen-specific mouse IgG_3, which efficiently activates complement but does not bind to FcγRs, to sensitize mice to have anaphylaxis after relevant antigen challenge. However, murine studies might underestimate the importance of complement-derived anaphylatoxins in human anaphylaxis because complement components are less capable of inducing anaphylaxis in another rodent (the rat) than in some larger mammals, such as dogs and pigs.[48]

These observations suggest that antibody-mediated anaphylaxis should be less severe in the absence of C3 or anaphylatoxin receptors when anaphylaxis is mediated by a complement-activating isotype, such as mouse IgG_{2a}. Even IgE-mediated anaphylaxis might be expected to be less severe in the absence of complement or anaphylatoxin receptors if IgE-mediated mast cell activation results indirectly in anaphylatoxin production. However, it is also possible that decreased stimulation of the G protein–dependent anaphylatoxin receptors increases the responsiveness of other G protein–dependent receptors, such as the histamine receptors, that mediate anaphylaxis (this would be analogous to the increased signaling through FcγRIII that is observed in the absence of FcεRIα[10]). Studies are required to evaluate these possibilities.

COMPLEMENT-MEDIATED ANAPHYLAXIS: OBSERVATIONS IN HUMAN SUBJECTS

The potential for complement-mediated human anaphylaxis is suggested by studies showing expression of 1 or both anaphylatoxin receptors on human mast cells, basophils, other myeloid cells, and vascular endothelial cells.[49-52] A role for complement in human antibody-mediated anaphylaxis is suggested by a correlation between the severity of anaphylaxis and serum anaphylatoxin levels, although the risk associated with increased anaphylatoxin levels is not as high as the risk associated with increased tryptase or histamine levels.[53] Complement can have a particularly important anaphylaxis-enhancing role in vespid toxin–induced anaphylaxis, in which complement activation by proteases in vespid toxins is likely to exacerbate disease caused by IgE antibodies to vespid toxin antigens.[54] There is also considerable clinical evidence for the induction of anaphylaxis by agents that directly activate complement in the absence of agent-specific IgE or IgG antibodies. This can be observed in association with hemodialysis (particularly during the first use of a new dialysis membrane), protamine neutralization of heparin and liposomal drug infusion, and infusions of drugs that are dissolved or suspended in certain lipid vehicles, such as Cremophor EL (BASF Corporation, Florham, NJ), and polyethylene glycol infusion.[48] Two limitations of these correlative studies are that (1) complement might not be the only factor activated that could contribute to shock (activation of the contact/kinin system is just one alternative possibility) and (2) no human studies have been performed to try to prevent anaphylaxis in any of these situations with inhibitors of complement activation or anaphylatoxin receptors. Taken together with mouse data, the most likely interpretation of the clinical studies is that acute complement activation can induce anaphylaxis, particularly when other factors are present (eg, FcεRI or FcγR crosslinking or pre-existing vasculopathies) that can add to or synergize with anaphylatoxin effects.

ANTIBODY- AND COMPLEMENT-INDEPENDENT ANAPHYLAXIS

Several drugs have been associated with anaphylaxis in susceptible subjects in the absence of a direct immunoglobulin-mediated mechanism or complement activation. These drugs include oversulfated heparin, aspirin, and nonsteroidal anti-inflammatory drugs (NSAIDs), antibiotics (including vancomycin and the fluoroquinilones), opiates, and drugs used in general anesthesia, particularly neuromuscular blocking agents.[38,55-57] Different mechanisms for anaphylaxis induction have been implicated for these different drugs in *in vitro* studies with human cells and plasma and *in vivo* animal studies. Oversulfated heparin directly activates the kinin system, with increased production of bradykinin.[57] NSAIDs, including aspirin, block COXs, which are essential for prostaglandin production. This results in decreased levels of prostaglandin E_2, which can suppress anaphylaxis, and increased levels of cysteinyl leukotrienes, which, among other effects, increase pulmonary smooth muscle contraction and vascular permeability.[38] Aspirin, unlike other NSAIDs, has also been reported to increase FcεRI-mediated basophil activation by enhancing phosphorylation of the signaling molecule Syk.[56] Vancomycin activates mast cells to release histamine and other mediators through a mechanism that is calcium-, phospholipase C–, and phospholipase A_2–dependent but otherwise unknown.[58] Opiates also induce histamine release through a mechanism that involves binding to central opioid receptors.[38] Fluoroquinolone antibiotics and nicotinic receptor antagonist nonsteroidal neuromuscular blocking agents, such as tubocurarine, which have a tetrahydroisoquinoline motif, directly activate mast cells by binding to MRGPRX2, a G protein–coupled receptor.[55] Taken together, these observations provide considerable reason to believe that the direct effects of these drugs on mast cells and basophils contribute to anaphylaxis. However, antibody/FcR interactions can also contribute to the ability of at least some of these drugs to induce anaphylaxis. With the exception that opioid-induced anaphylaxis has been reversed in human subjects by means of administration of opioid receptor antagonists,[38,59] the importance of direct mast cell activation for anaphylaxis induction by these drugs has not been proved in human subjects *in vivo*. For anaphylaxis associated with other drugs, such as

TABLE I. Etiologic mechanisms of anaphylaxis and their distinguishing characteristics

Type	Inciting agents	Cells	Receptors	Mediators
IgE mediated	Food allergy	Mast cells	FcεRI	Histamine
	Insect sting allergy	Basophils		PAF
	Drug allergy			
IgG mediated	Biologicals	Macrophages	FcγRIII	PAF
	Drugs	Neutrophils	FcγRI	Histamine
	Dextrans	Basophils	FcγRIV (mouse)	
	Aprotinin		FcγRIIA (human)	
	Transfusions			
Complement mediated	Lipid incipients	Macrophages	C3aR	PAF
	Micellar drugs	Mast cells	C5aR	Histamine
	Liposome			
	Other nanoparticles			
	Polyethylene glycol			
	Cellulose membranes			
Direct mast cell activation*	NSAIDs, including aspirin	Mast cells	MRGPRX2	Cysteinyl leukotrienes
	Vancomycin	Other myeloid cells	Other receptors	Histamine
	Opiates			
	Local anesthetics			
	Fluoroquinolone antibiotics			
	Neuromuscular blockers			
	Octreotide			
	Leuprolide			

*Mechanisms differ for different agents.

iodinated radiologic contrast media, the relative roles of IgE antibodies, IgG antibodies, complement, and direct effects on myeloid cells are still debated.[38] It also remains to be determined whether any anaphylaxis-associated drugs cause disease solely through direct effects on mast cells or as part of a 2-hit or multihit mechanism and why some subjects are much more susceptible than others.

CONCLUSION

Although it is currently impossible to prove beyond a doubt that non–IgE-mediated anaphylaxis is clinically relevant, considerable evidence supports the occurrence and clinical importance of human IgE-independent anaphylaxis that is mediated by IgG, complement, or direct basophil and mast cell activation. Clinical situations that have been associated with the 4 different putative types of anaphylaxis are summarized in Table I, which also summarizes the cells, receptors, and mediators that are thought to contribute to the pathogenesis of each form of this disorder. IgG-mediated anaphylaxis should be suspected when there are large infusions of antigen and high titers of IgG antibody specific for the infused antigen, whereas complement-mediated anaphylaxis and direct mast cell/basophil activation should be suspected in patients who have received drugs, biologicals, or excipients that are known to have the appropriate complement or mast cell/basophil–activating properties. Identifying the probable cause of a specific episode of anaphylaxis is likely to increase in importance as therapeutics that block a specific pathway become available; pretreatment with such therapeutics can also be useful as prophylaxis for patients who require specific anaphylaxis-associated drugs. Finally, it is important that a single episode of anaphylaxis might involve more than 1 mechanism. Simultaneous occurrence of IgG- and IgE-mediated anaphylaxis has been demonstrated in mice,[60] as has synergy between anaphylatoxin- and IgE-mediated anaphylaxis[46]; IgG/antigen complexes that bind to stimulatory FcγRs can also initiate anaphylaxis by activating complement, and direct mast cell activation by drugs is likely to act additively or synergistically with antibody- or complement-dependent activation of these and other myeloid cells to increase anaphylaxis severity.

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