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Summary

The aggregation of high-affinity IgE receptors (FcεRI) on mast cells and basophils has long been known to be the critical event that initiates allergic reactions. Monomeric IgE was recently found to induce a variety of effects when binding to FcεRI. Up-regulation of FcεRI required binding only, whereas other responses resulted from FcεRI aggregation. Interestingly, FcεRI aggregation has been understood to generate a mixture of positive and negative intracellular signals. Mast cells/basophils express also low-affinity and, under specific conditions, high-affinity IgG receptors. When co-engaging these receptors with FcεRI, IgG antibodies can amplify or dampen IgE-induced mast cell activation. Based on these findings, FcRs have been proposed to be used as targets and/or tools for new therapeutic approaches of allergies.

Introduction

Cell activation results from the transient displacement of a physiological balance between positive and negative signals delivered by activating and inhibitory receptors. Such receptors are constitutively or inducibly expressed on the membrane of cells involved in allergy. Among these are Fc Receptors (FcRs). They include high- and intermediate-affinity IgE receptors (FcεRI and FcεRII, respectively), high- and low-affinity IgG receptors (FcγRI, and FcγRII, respectively), and high-affinity IgA receptors (FcαRI). The last two years covered by this review have witnessed significant advances concerning FcR-dependent negative signaling. We will focus on FcR-dependent negative regulation of the IgE response, of FcεRI expression, and of FcεRI signaling, and discuss FcR-based new therapeutic approaches of allergies.

Regulation of IgE by FcRs

FcεRII/CD23 was shown to control IgE production and transport.

CD23 has been known to enhance IgG and IgE responses following immunization with Ag and specific IgE [1]. CD23 can also decrease IgE responses. Transgenic mice overexpressing murine CD23 indeed failed to mount an IgE response following *L. major* infection or Ag+alum immunization, whereas increased and sustained IgE antibody responses were observed in $CD23^{-/-}$ mice. Recently, NZB mice were reported to express a mutated CD23 allele and to develop hyper-IgE syndromes and increased IgE responses following immunization [2]. Mutations, located in the C-lectin domain and the stalk region, impair the trimerisation and cell surface expression, and decrease 10-100-fold the affinity of CD23 for IgE.

Besides regulating IgE production, CD23 was suggested to participate in intestinal allergy by regulating the trans-epithelial transport of IgE and IgE-allergen complexes. Murine CD23b and its novel isoform, CD23bΔ5, constitutively endocytose IgE-allergen complexes and free IgE, respectively, enabling their transport from the luminal to the basolateral pole of enterocytes [3]. This isoform resembles human CD23a which is expressed on human enterocytes and could mediate the transport of IgE-allergen complexes towards intestinal mast cells, when constitutively internalized *via* clathrin-coated pits.

Regulation of FcεRI expression

Relative synthesis and degradation rates determine FcεRI expression. FcεRI assembly has been understood to be controlled by the FcRβ subunit while degradation is controlled by IgE.

IL-4 and IL-10 decrease FcεRI expression in mice. Decrease affects FcRβ, but not FcεRIα or FcRγ expression [4]. The expression of murine FcεRI indeed requires all three subunits. If human FcεRI does not require FcRβ, rendering FcεRI expression in other cells possible, FcRβ nevertheless regulates human FcεRI expression in mast cells and basophils. $FcR\beta_T$, a splice variant of FcRβ, blocks the maturation of FcεRIα, prevents its association with FcRγ, and increases its proteasome degradation [5]. $FcR\beta_T$ is a truncated molecule which competes with FcRβ during the assembly of the receptor in the reticulum, leading to an intracellular accumulation of immature, misfolded FcεRIα chains [6]. Based on these data, polymorphisms of the FcRβ gene have been searched for. A SNP resulting in an E237G mutation in the intracytoplasmic domain of FcRβ was identified as a potential risk factor [7,8] and, compared to E237 donors, G237 healthy volunteers had basophils with a higher FcεRI

expression [9]. The E237G substitution had however earlier been shown not to affect the *in vitro* expression/function of FcεRI [10,11]. Two other SNPs, tightly linked to the E237G SNP, were identified in potential binding sites for transcription factors and resulted in a higher promoter activity [9]. This provides a possible functional link between FcRβ polymorphisms, FcεRI expression and atopy.

An incubation of mast cells with IgE for several hours or days increases FcεRI expression. IgE-induced FcεRI upregulation does not depend on ITAMs and does not involve early signaling events triggered by FcεRI aggregation. It results from a decreased internalization and degradation that constantly removes free FcεRI from the cell surface, resulting in an enhanced half-life at the membrane. The synthesis rate being not affected, FcεRI accumulate on mast cells/basophils which bind even more IgE [12]. Consistent with these observations, the level of FcεRI expression on basophils correlates with the concentration of IgE in blood.

Autonomous regulation of FcεRI signaling

Monomeric IgE and FcεRI signaling

Based on the demonstration that FcεRI aggregation — at least dimerization — was required to trigger mast cell degranulation, it has been accepted that, alone, the binding of IgE to FcεRI induces no detectable response. Recently, however, IgE was reported to increase the survival of mouse mast cells in the absence of IL-3 [13,14] and to induce FcεRI internalization, cytokine secretion and degranulation [15,16]. These responses were all induced by high concentrations (several μg/ml) of monoclonal IgE anti-DNP/TNP, among which highly and poorly "cytokinergic" IgE were distinguished [17]. Mast cell survival and cytokine secretion were found to depend on the FcRγ ITAM [16] and FcεRI internalization required the src kinase Lyn, whereas degranulation, cytokine secretion and survival required also Syk [18]. It thus became thought that the binding of some IgE to FcεRI could trigger intracellular signals similar to those triggered by FcεRI aggregation. As a consequence, a proportion of mast cell and basophil FcεRI being normally occupied by IgE *in vivo*, these would either generate a physiological background response or prime cells for activation. A clarifying finding was that IgE alone-induced mast cell responses were inhibited by the monovalent hapten DNP-lysine [17], demonstrating that IgE was engaged by unknown plurivalent ligand(s) resulting in FcεRI aggregation. A recently proposed explanation is that, differing from physiological conditions where low concentrations of polyclonal IgE with different specificities bind to FcεRI on any given mast cell, high concentrations of monoclonal IgE with the same specificity saturate FcεRI in experimental conditions, thus rendering IgE aggregation easier. As SPE7, the most cytokinergic IgE anti-DNP described, was found to bind to an epitope other than DNP, low-affinity cross-reactions, possibly with cell surface or even IgE-borne epitopes, were proposed to induce minimal receptor aggregation [19].

Differential signaling as a function of the degree and duration of FcεRI aggregation

Interestingly, these experiments revealed that qualitatively different responses were induced as a result of quantitative differences in receptor aggregation.

Low levels of FcεRI aggregation, induced by IgE alone, triggered the secretion of IL-3 or MCP-1, but not degranulation, whereas high levels of receptor aggregation, induced by the same IgE and multivalent antigen, triggered both degranulation and cytokine secretion [16,20,21]. Using FcRγ-deficient mast cells that expressed CD8/FcRγ chimeras and were

challenged with anti-CD8 antibodies, it was found that degranulation required stronger stimulation whereas survival required longer stimulations [21].

When challenged with antigen, histamine release by IgE-sensitized mast cells displays a bell-shaped curve as a function of antigen concentration. This was thought to result from decreased receptor aggregation, due to a competition of antigen for IgE. Intracellular signals do not decrease, however, but increase, as antigen concentration increases, including the phopshorylation of the inositol 5-phosphatase SHIP1, and inhibition observed in antigen excess was abrogated in SHIP1$^{-/-}$ mast cells [22]. SHIP1 therefore determines both the threshold, as proposed earlier, and the upper limit of aggregation that triggers degranulation.

Subcellular compartments and signaling domains

A recent advance in understanding immunoreceptor-dependent (mast) cell activation has been the recognition of membrane signaling domains. Lipid rafts were first understood as platforms where critical membrane-associated signaling molecules are concentrated and into which receptors transiently translocate upon aggregation. Electron microscopy mapping of signaling molecules using immunogold-labeled antibodies unraveled the existence of sub-domains. Primary signaling domains contain receptors and, among other signaling molecules, Syk but not Lyn, whereas secondary signaling domains contain the transmembrane adapter LAT and PLCγ-1, but not receptors [23].

Another sub-cellular compartment, the F-actin skeleton, is involved in negative regulation. Actin-disrupting drugs increased aggregation-induced FcεRI and Syk phosphorylation, as well as degranulation, especially when triggered by high concentrations of ligands [24]. Inhibitors of actin polymerization also increased IgE-induced phopshorylation of LAT and PI3 kinase activity, but they did not enhance FcεRI translocation into lipid rafts. Because actin polymerization inhibitors, alone, induced some among these signaling events, F-actin was suggested to set the threshold for degranulation before FcεRI engagement [25]. FcεRI aggregation induces a rapid extracellular Ca^{2+}-dependent initial decrease of the F-actin content over the first two minutes, followed by a slow recovery. The Wiskott-Aldrich syndrome protein (WASP)-interacting protein (WIP) negatively regulates actin polymerization/elongation. F-actin recovery was indeed faster in WIP$^{-/-}$ mast cells. Interestingly, IgE-induced mast cell responses were all impaired in WIP$^{-/-}$ mast cells, and WIP$^{-/-}$ mice were resistant to IgE-induced systemic anaphylaxis, suggesting that F-actin-dependent inhibition was enhanced in WIP$^{-/-}$ mast cells. Cbl-dependent Syk degradation was however also enhanced in WIP$^{-/-}$ mast cells [26].

Signaling molecules in negative regulation

Another recent advance was the understanding that the same signaling molecules concur both to activate and to inhibit mast cells (Fig. 1).

FcRβ amplifies FcRγ-dependent signaling. It also contributes to negative signaling due to an additional tyrosine, located between the two YxxL motifs of its ITAM [27]. MAPK and NF-κB activation and, ultimately, the secretion of cytokines were indeed enhanced when this residue was mutated. Lyn and SHIP1 bound to this tyrosine *in vitro*, less FcRβ coprecipitated with Lyn, and SHIP1 was less phosphorylated following FcεRI engagement, when the additional tyrosine was mutated, suggesting that FcRβ may contribute to recruit SHIP1 and Lyn in signaling complexes.

Lyn initiates positive signaling by phosphorylating FcεRI ITAMs and Syk. It also generates negative signaling. Lyn$^{-/-}$ mast cells were indeed more responsive to IL-3, Stem Cell Factor and IgE+antigen. IgE-induced ITAM phosphorylation, PLC-γ activation and Ca^{2+} mobilization were reduced but persisted longer [28]; PI(3,4,5)P3 levels, and Fyn activity were however markedly enhanced in Lyn$^{-/-}$ mast cells. Increased PI(3,4,5)P3 levels were accounted

for by an impaired activity of SHIP1 [28] and Fyn hyperactivity by an abrogation of Lyn-dependent phosphorylation of Csk-Binding Protein (Cbp) which mediates the recruitment of Csk. Csk phosphorylates regulatory tyrosines in Fyn and thereby inhibits its catalytic activity. Supporting this model, the hyper-responsiveness of $Lyn^{-/-}$ mast cells to IgE was abrogated in $Lyn^{-/-}/Fyn^{-/-}$ BMMCs [29]. Interestingly, the phenotype of $Lyn^{-/-}$ mice was reminiscent of an "allergic" phenotype which could not be accounted for by the hyper-reactivity of mast cells only. As these mice grew older, they displayed increased serum IgE concentrations, an increased FcεRI expression, increased numbers of peritoneal mast cells and eosinophils, and elevated levels of plasma histamine [29]. Most of these allergy-associated traits could be ascribed to a biased isotypic switch toward IgE due to the hyper-responsiveness of $Lyn^{-/-}$ B cells to IL-4, and to the consequences of an increased IgE serum concentration.

The transmembrane adapter LAT organizes signals generated by immunoreceptors and couples them with downstream pathways. FcεRI aggregation in $LAT^{-/-}$ BMMCs triggered a reduced phosphorylation of SLP-76 and PLC-γ, resulting in decreased Ca^{2+} mobilization and MAPK activation and, ultimately, in decreased mediator release and cytokine secretion. LAT contains multiple intracytoplasmic tyrosines which, when phosphorylated by Syk, provide docking sites for SH2 domain-containing cytosolic enzymes and adapters. Based on mutational analysis, one tyrosine was shown to recruit PLC-γ, and the three distal tyrosines Gads, Grap and Grb2. Similar results were obtained in T cells and mast cells [30]. LAT knock-in mice in which either the PLC-γ-binding tyrosine or the adapter-binding three distal tyrosines were mutated exhibited a polyclonal lymphoproliferation of $CD4^+$/TCRαβ or $CD4^+$/TCRγδ T cells, respectively, that secreted exaggerated levels of TH2 cytokines. Consequently, serum IgG1 and IgE were markedly increased, and peripheral tissues were infiltrated with eosinophils [31,32]. LAT mutations therefore unraveled that, besides positive signals, LAT supports negative signals that normally control terminal T cell differentiation and proliferation. Our analysis of IgE-induced biological responses of mast cells derived from the same knock-in mice led to the same conclusion for FcεRI signaling. Thus, the PLC-γ-binding tyrosine had a negative effect on mediator release when adapter-binding tyrosines were mutated and, conversely, adapter-binding tyrosines had a negative effect when the PLC-γ-binding tyrosine was mutated. Importantly, LAT integrates positive and negative signals. Thus, the positive effect of the four distal tyrosines on β-hexosaminidase release was of a lower magnitude than the intense positive effects of either the PLC-γ-binding tyrosine or of adapter-binding tyrosines alone [33].

NTAL is another transmembrane adapter resembling LAT. Both LAT and NTAL are expressed in mast cells. NTAL is phosphorylated upon FcεRI or Kit engagement. It was found to negatively regulate FcεRI signaling in murine mast cells, and to positively regulate FcεRI and Kit signaling in human mast cells. All IgE-induced responses were indeed enhanced in mast cells from NTAL-deficient mice, as well as most intracellular signals [34,35]. Whether NTAL recruits an inhibitory molecule or competes with LAT for signaling molecules remains to be clarified. LAT and NTAL were however found in distinct lipid rafts [34]. By contrast, NTAL siRNA that reduced NTAL expression by 70% decreased IgE- and Stem Cell factor-induced degranulation of human mast cells [36]. Whether these seemingly discordant results illustrate the negative and the positive components of NTAL signaling or are due to differences between the two species is not known.

Regulation of FcεRI signaling by coreceptors and other FcRs

IgE-induced reactions can be modulated by a variety of mast cell membrane proteins under different conditions. These comprise ITIM-containing inhibitory molecules such as

MAFA, gp49B1, SIRPα or PIR-B. Inhibitory molecules without ITIM were recently identified. These are CD200R, an immunoglobulin superfamily member, whose ligation by antibodies or Ig-CD200 fusion proteins inhibits degranulation and cytokine secretion [37] and CD63, a β1-integrin-associated tetraspanin, whose engagement by antibodies inhibits not only mast cell adhesion to fibro/vibronectin, but also IgE-induced degranulation of adherent mast cells by a mechanism that affects the Fyn/Gab2/PI3K pathway [38]. Other molecules that regulate FcεRI signaling are receptors for immunoglobulins other than IgE.

FcαRI

The engagement of FcRγ-associated FcαRI by monomeric ligands, including human serum IgA, was found to inhibit IgE-induced degranulation of RBL-2H3 transfectants expressing FcαRI or FcαRI-FcRγ chimeras. Inhibition required an intact FcRγ ITAM which was phosphorylated following monovalent engagement and recruited the tyrosine phosphatase SHP-1. When administered intraperitoneally into human FcαRI transgenic mice, anti-FcαRI Fab fragments also inhibited bronchial constriction and airway infiltration by inflammatory cells induced by IgE and antigen in a murine model of allergic asthma [39]. Although observed in artificial models, these findings possibly explain that IgA have anti-inflammatory effects and that IgA deficiencies are correlated with increased susceptibility to autoimmune and allergic diseases.

FcγRIIB

FcγRIIB was reported to inhibit IgE-induced mast cell activation 10 years ago. Since then, molecular mechanisms involved in FcγRIIB-dependent negative regulation have been unraveled and the *in vivo* significance of this regulation has been established through the generation of FcR-deficient mice.

When co-aggregated with FcεRI and tyrsosyl-phosphorylated by Lyn, FcγRIIB recruits SHIP1 which is the main intracellular effector of inhibition (reviewed in [40]). FcγRIIB was recently understood to amplify SHIP1-dependent, autonomous regulation of FcεRI. As discussed above, negative regulation of FcεRI signaling involves both SHIP1 and an association with F-actin. How FcεRI recruits SHIP1 is not known as the phosphatase failed to coprecipitate with FcεRI subunits. We recently found that the high-molecular-weight isoform of SHIP1 is constitutively associated with the actin-binding protein, filamin-1 in the F-actin skeleton. Following coaggregation, FcγRIIB and FcεRI rapidly interact with the F-actin skeleton and engage SHIP1 and filamin-1. Later on, filamin-1 and F-actin dissociate while SHIP1 remains associated with FcγRIIB. Filamin-1 therefore functions as a donor of SHIP1 for FcγRIIB which concentrates this phosphatase in the vicinity of FcεRI and thereby extinguish activation signals [41].

FcγRIIB$^{-/-}$ mice are more sensitive not only to IgG- but also to IgE-induced passive systemic anaphylaxis. They are also more sensitive to allergic rhinitis [42]. These mice displayed impaired nasal and oral tolerance, suggesting that FcγRIIB are involved in tolerance to air-borne and food allergens. FcγRIIB expressed on dendritic cells were indeed shown to regulate antigen presentation, cytokine secretion and the generation of regulatory T cells in this model of nasal tolerance [43].

FcγRI

INFγ induces the expression of the high-affinity IgG receptor FcγRI/CD64 on cultured human mast cells. INFγ-treated mast cells were induced to degranulate by human heat-aggregated IgG1 (but not IgG2, IgG3 or IgG4), IgG1 followed by anti-IgG1 antibodies, or anti-CD64 antibodies. FcγRI signals *via* the same FcRγ subunit as FcεRI, and FcγRI triggered

β-hexosaminidase release, PGD2 and LTC4 production, and GM-CSF, IL-3, IL-13 and TNF-α secretion, as FcεRI. FcγRI however triggered more IL-13 and TNF-α than FcεRI whereas FcεRI triggered more IL-3 than FcγRI [44].

FcRs as therapeutic targets/tools in allergies

In view of data reviewed above, antibody-based new therapeutic approaches of allergies have recently been proposed. Some aimed at preventing the interaction of IgE with FcεRI, others at targeting FcεRI with toxic molecules, others at co-engaging FcγRIIB with FcεRI (Fig. 2).

Anti-IgE therapy

Humanized monoclonal antibodies against the FcεRI-binding site of IgE (Omalizumab) have been used in humans, aiming at blocking mast cell sensitization by IgE. Omalizumab proved safe and efficient in seasonal allergic rhinitis and allergic asthma. It decreased asthma-related symptoms and corticosteroid uptake, and improved lung function and quality of life. Serum IgE became undetectable, FcεRI expression was markedly reduced on basophils (within weeks) as a function of IgE levels, on mast cells (within months) and on dendritic cells [45]. Noticeably, Omalizumab treatment markedly increased the sensitivity threshold in peanut-allergic patients [46].

Anti-FcR and FcR ligands

A chimeric fusion protein made of one Cε3 domain and Bak, a proapoptotic molecule, was proposed to target and destroy mast cells/basophils selectively. It was shown to bind to FcεRI-expressing cells selectively, to be slowly internalized (within hours), and to kill cells by apoptosis [47].

Other chimeric molecules aiming both at targeting FcεRI-bearing cells and at exploiting the inhibitory properties of FcγRIIB were described. A human IgG1 Fc fragment (hinge-Cγ2-Cγ3) fused to a human IgE Fc fragment (Cε2-Cε3-Cε4) was first reported to inhibit IgE-induced human mast cell and basophil activation [48]. The same human IgG1 Fc fragment fused to the cat allergen Fel-d1 was then reported to inhibit Fel-d1-induced activation of human mast cells and basophils sensitized with serum from patients allergic to Fel-d1, and Fel-d1-induced anaphylaxis in human FcεRIα transgenic mice [49]. Evidence that FcγRIIB-dependent regulation was involved is however lacking. Rather, competition (with IgE for binding to FcεRI in the first case, and with allergen for binding to IgE in the second case) was most probably responsible for inhibition induced by these molecules. Supporting this interpretation, IgE-induced Syk phosphorylation was impaired by both molecules. Another bispecific molecule, constructed by chemically crosslinking one Fab' fragment of an anti-human IgE and one Fab' fragment of an anti-human FcγRII antibody was proposed as a proof-of-concept for the co-engagement of IgE-loaded FcεRI with FcγRIIB as a therapeutic strategy in allergic diseases. Antigen-induced histamine release was inhibited when human mast cells and basophils, sensitized with human IgE antibodies, were incubated with this molecule prior challenge [50].

Conclusion

IgE-induced FcεRI aggregation has long been thought to generate positive signals only, leading to mast cell activation and allergic symptoms. During the last years, FcεRI signaling has been understood to consist of a mixture of positive and negative signals, whose

integration determines mast cell responses. Interestingly, signaling molecules previously thought to contribute activation signals only were also found to contribute negative signals. These include FcR subunits, tyrosine kinases and adapters.

IgE and FcεRI have long been known to be the key players in allergic reactions. It now appears that IgG and, possibly, IgA may significantly interfere, positively and negatively, with IgE-induced reactions by engaging activating and inhibitory FcRs. FcRs other than FcεRI therefore provide new targets/tools for novel therapeutic approaches of allergies. Understanding better fundamental mechanisms used by signaling molecules and FcRs *in vivo* is a requirement for such treatments to be safer and more efficient.

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Comments on selected references :

****5. Donnadieu *et al.*** A novel isoform of the human FcRβ subunit, $FcR\beta_T$, was identified as a competitor of FcRβ for the association to FcεRIα chain. $FcR\beta_T$ prevents the expression of the FcεRI complex at the cell surface, by associating with immature, misfolded α-chains. This important work unravels a new mechanism for controlling human FcεRI expression at the mast cell/basophil membrane

* **9. Nishiyama *et al.*** The controversy involving a SNP in the coding region of the FcRβ gene, previously linked to allergic symptoms and FcεRI expression level, and the finding that the resulting mutation does not by itself modulate this expression, is solved in this work. Indeed, authors describe additional SNPs in the FcRβ gene promoter region, linked to the previously described SNP, leading to enhanced promoter activity and thereby enhanced FcεRI expression in atopic individuals.

17. Kitaura *et al. Recent surprising findings on the biological effect of monomeric IgE binding to mast cells are partially unravelled in this study. Monomeric IgE-induced mast cell cytokine secretion, survival and signalling were indeed abolished in the presence of monovalent hapten. These biological effects are therefore the result of FcεRI aggregation induced by IgE self-aggregation or aggregation of IgE at the membrane of these mast cells.

** **29. Odom *et al.*** This work dissects the mechanisms accounting for the allergic-like phenotype of Lyn-deficient mice. It shows how this src kinase phosphorylates an adapter which recruits a kinase which inactivates src kinases by phoshorylating a regulatory tyrosine of these enzymes. It also shows how, by dampening IL-4 signaling in B cells, Lyn prevents high titers of IgE from being synthesized and, as a consequence, controls mast cell numbers and FcεRI expression.

** **39. Pasquier *et al.*** An ITAM-bearing receptor that has been known to activate cells upon aggregation by multivalent ligands was found to inhibit cell activation induced by other receptors upon ligation by monovalent ligands. Monovalent binding induced a weak phopshorylation of ITAMs which recruited the tyrosine phosphatase SHP-1. The significance of this finding will be major when it has been reproduced in more physiological models.

* **49. Zhu *et al.*** A bifunctional molecule made of a cat allergen coupled with the Fc portion of a human IgG1 was shown to inhibit IgE-induced reactions *in vitro* and *in vivo*. The mechanism proposed for inhibition may not be prominent but, whatever the mechanism, this innovative molecule proved quite efficient.

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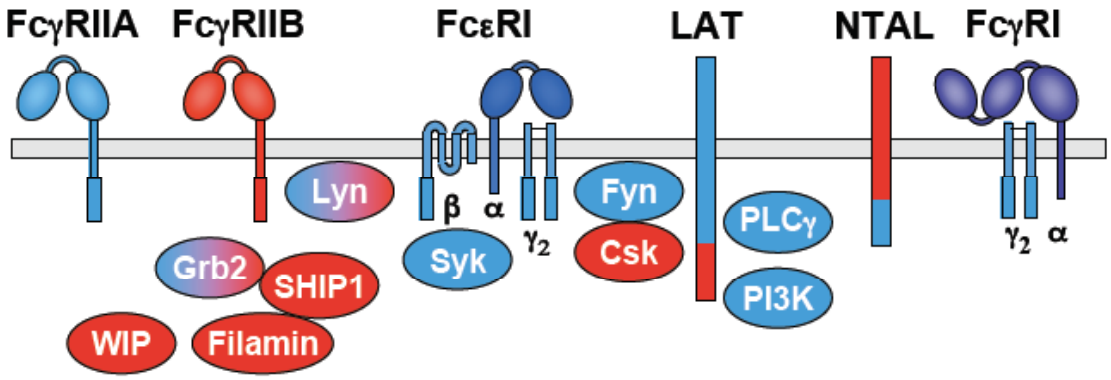

Fig. 1: FcRs and signaling molecules involved in positive and negative regulation of human mast cell/basophil activation.

FcRs, transmembrane and cytosolic adapters and intracellular molecules expressed by human mast cells/basophils are represented in blue when involved in cell activation, and in red when involved in inhibition of cell activation. Molecules involved in both pathways are represented in both colours.

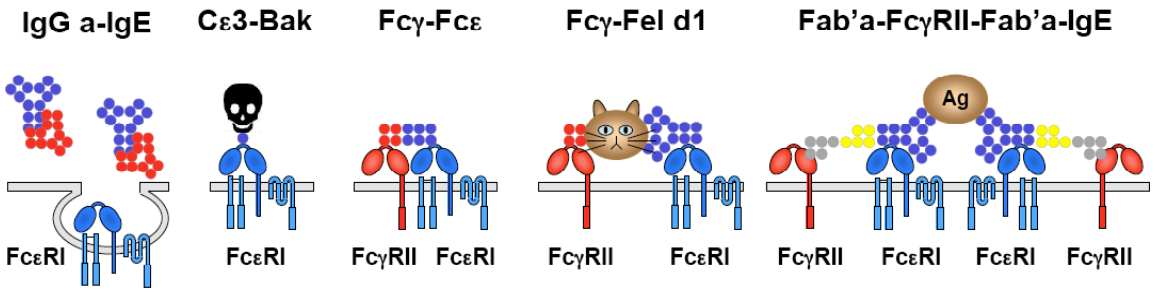

Fig. 2: FcR-related therapeutic tools for allergy.

IgG a-IgE: When binding to circulating IgE, anti-IgE antibodies prevent IgE-induced up-regulation of FcεRI expression on mast cells/basophils.

Cε3-Bak: When binding to FcεRI, the pro-apoptotic molecule Bak fused to a single Cε3 domain is internalized and induces apoptosis of mast cells/basophils selectively.

Fcγ-Fcε: While the Fcε portion binds to FcεRI, competing with IgE antibodies, the Fcγ portion could bind to FcγRII and crosslink the two receptors.

Fcγ-Fel d1: While the major cat allergen Fel d1 is recognized by FcεRI-bound IgE antibodies, competing with natural allergen, the Fcγ portion could bind to FcγRII and crosslink the two receptors.

Fab'a-FcγRII-Fab'a-IgE: This bispecific antibody crosslinks FcεRI-bound IgE antibodies and FcγRII, and inhibits subsequent antigen-induced activation of mast cells/basophils.