



Immunopharmacological modulation of mast cells

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Mast cells produce a wide spectrum of mediators and they have been implicated in several physiopathological conditions (e.g. allergic reactions and certain tumors). Pharmacologic agents that modulate the release of mediators from mast cells has helped to elucidate the biochemical mechanisms by which immunological and non-immunological stimuli activate these cells. Furthermore, the study of surface receptors and signaling pathways associated with mast cell activation revealed novel pharmacologic targets. Thus, the development of pharmacologic agents based on this new wave of knowledge holds promise for the treatment of several mast cell-mediated disorders.

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Introduction

Mast cells and basophils are the only cells that express the tetrameric ($\alpha\beta\gamma_2$) high affinity receptor for IgE (Fc ϵ RI) and synthesize histamine in humans, although they differ morphologically, ultrastructurally, immunologically, and pharmacologically [1]. Mast cells originate from CD34⁺ pluripotent cells present in bone marrow, in fetal liver and in peripheral blood [2–5]. Mature mast cells are distributed throughout mucosal and connective tissues where they are often adjacent to blood and lymphatic vessels, near or within nerves, and beneath epithelial surfaces that are exposed to the environment (e.g. skin, airways and the gastrointestinal and genitourinary systems) [6].

In response to IgE-dependent and IgE-independent stimuli mast cells can synthesize and release a wide spectrum of mediators, namely: preformed mediators (e.g. histamine, serotonin, proteases); newly synthesized lipid mediators (e.g. prostaglandin D₂ [PGD₂] and leukotriene C₄ [LTC₄]); cytokines/growth factors (e.g. IL-3, IL-13, VEGF-A, GM-CSF, TNF) and chemokines (e.g. CCL2/MCP-1 and IL-8/CXCL8) [7]. Therefore, in theory, mast cells may be involved in several pathophysiological conditions [8]. Results obtained with new mouse models of mast cell deficiency suggest that mast cells play nonredundant roles in IgE-mediated responses [9–11]. *In vitro* and *in vivo* data suggest that mast cells may foster the growth of certain tumors [12–14] but should also be involved in IgE-mediated tumor cell killing [15]. Recent evidence suggests that mast cell and their mediators can enhance resistance to animal venoms [16,17^{**},18,19^{**},20]. Finally, mast cells have been involved in the acute and chronic response to pathogens [21]. Consequently, elucidation of the intrinsic signaling pathways underlying mast cell activation and their pharmacological modulation is poised to give rise to novel strategies for the treatment of a wide spectrum of disorders.

Several pharmacologic modulators of mast cell functions have been identified. Notably, no one drug that can solely and specifically inhibit mast cell activation has yet been discovered. Furthermore, significant differences have been documented between mast cells isolated from different species or even from different anatomical sites and between the pharmacological effects of compounds of the same class of drugs [22,23]. Therefore, caution should be exercised in extrapolating results from *in vitro* experiments to the clinical setting in terms of the clinical efficacy of certain drugs in different mast cell-mediated disorders.

Mast cells and their pharmacological modulation have been the topic of several excellent reviews [6^{*},24,25]. In this brief review we shall concentrate on pharmacological and immunological agents that modulate the release of mediators from mast cells that are already therapeutically in use or appear to hold promise for the treatment of inflammatory diseases.

β_2 -Adrenoceptor agonists and cyclic AMP phosphodiesterase inhibitors

Adrenergic β_2 -agonists are the treatment of choice for reversal of acute bronchospasm in bronchial asthma [26].

Several *in vitro* studies have shown that β_2 -agonists are very effective at preventing the release and generation of mediators from human lung (HLMC) and skin tissues (HSMC) mast cells, and from cord blood-derived (hCBMC) and peripheral blood-derived mast cells (hPBMC) [27–30]. Mechanistically, β_2 -agonists induce the activation of adenylyl cyclase and the intracellular accumulation of cyclic AMP (cAMP), thus downregulating the functions of mast cells [27].

One factor that may limit the therapeutic effectiveness of β_2 -agonists is the development of tolerance. Two long-acting β_2 -agonists, formoterol and indacaterol, inhibit the IgE-dependent responses of HLMC. However, long-term incubation of mast cells with these long-acting β_2 -agonists lead to a reduction in the ability of these drugs to prevent the release of histamine [31]. In addition, stem cell factor (SCF), a growth factor critical for mast cells growth, differentiation, and survival [6^{*}] that also enhances IgE-dependent HLMC degranulation and cytokine production [32,33], attenuates β_2 -agonist-induced inhibition of histamine release from HLMC [34]. The latter observation is relevant because SCF can be immunologically released from HLMC *in vitro* [35] and its expression is markedly increased in asthmatic airways [36,37]. Therefore, the presence of SCF in the airways of asthmatic patients may attenuate the beneficial effects of β_2 -agonists.

Phosphodiesterases (PDEs) are a family of enzymes that catalyze the metabolism of intracellular cyclic nucleotides [38]. There are, at present, 11 PDE families and 21 isoforms with numerous splice variants characterized by differences in structure, substrate specificity, inhibitor selectivity, tissue and cell distribution, and subcellular distribution. A variety of PDE inhibitors are available as pharmacological probes: standard non-selective inhibitors of PDE activity such as theophylline and 3-isobutyl-1-methylxanthine (IBMX); and selective inhibitors of individual PDE isozymes. Non-selective PDE inhibitors inhibit the activation of HLMC and HSMC [39,40], whereas selective PDE4 inhibitors (e.g. rolipram, Ro20-1724 and RP73401) are ineffective. Thus, the nature of PDE(s) that regulate mast cell activation is still uncertain.

Cromones

Cromones (sodium cromoglycate [DSCG] and nedocromil sodium) have lost popularity in the treatment of allergic disorders, mainly due to their limited therapeutic effect and short duration of action [26]. However, cromones are very well tolerated, unlike the commonly used glucocorticoids, which renders them interesting therapeutic agents. Cromones inhibit mast cell activation, although their mechanism of action has not yet been completely characterized and may involve at least the reduction of calcium influx, which is necessary for mast

cell degranulation [41,42]. Recently, it has been demonstrated that DSCG and nedocromil inhibit histamine and PGD₂ release by inducing PKC-dependent phosphorylation and release of annexin A₁ [43^{*}], a mechanism common to other cell types [44,45]. Interestingly, DSCG inhibits mast cell activation and mediator release in rats but not in mice [46]. The latter results emphasize the pharmacological differences that exist between mast cells from different species.

Recently, two novel cromones, meta-azocromone (MAC) and para-azocromone (PAC), were found to be almost 100 times more powerful than DSCG in suppressing β -hexosaminidase release from the LAD2 mast cell line [47^{*}], which renders them interesting candidates as anti-allergic drugs.

Glucocorticoids

Glucocorticoids are one of the therapeutic mainstays in most inflammatory and immune disorders [48] and can modulate mast cell activity at different levels [49^{*}]. Glucocorticoids reduce Fc ϵ RI expression [50–52] and inhibit mast cell degranulation through a nongenomic mechanism [53]. In addition, glucocorticoids decrease the number of mast cells in human inflamed tissue biopsies [54,55]. Although the latter effect was initially attributed to the ability of glucocorticoids to reduce the expression of SCF by resident cells (e.g. fibroblasts) [56], it was subsequently reported that prolonged treatment of lung fibroblasts with glucocorticoids stimulates, rather than inhibits, the expression of SCF [57]. Thus, further studies are required to understand how glucocorticoids modulate the survival and the activity of mast cells in inflamed tissues.

The fact that human mast cells are a major source of cytokines and chemokines [7] raised the important question as to whether glucocorticoids can modulate, *in vitro* as well as *in vivo*, the synthesis of these mediators in immunologically activated Fc ϵ RI⁺ cells. Glucocorticoids inhibit cytokine and chemokine production by mouse [58] and human mast cells [59,60]. Interestingly, the combination of glucocorticoids and the calcineurin inhibitor FK506 almost completely suppresses the induction of all chemokine genes [59]. This suggests that a combination of a glucocorticoid and a calcineurin inhibitor may be more effective than each single agent used alone in the modulation of IgE-mediated activation of human mast cells.

Non-steroidal anti-inflammatory drugs

Although aspirin and other non-steroidal anti-inflammatory drugs (NSAIDs) enhance IgE-mediated activation of basophils, but not human mast cells [61], nimesulide (4-nitro-2-phenoxy-methane sulfonanilide) and its active metabolite 4-hydroxy-nimesulide inhibit the release of preformed and *de novo* synthesized mediators from

human FcεRI⁺ cells challenged with IgE-mediated and non-IgE-mediated stimuli [22,61–63]. These data might explain why nimesulide is well tolerated in patients with acetylsalicylic acid idiosyncrasy [64].

Nimesulide, is a NSAID chemically unrelated to other compounds of the same class, its functional acid group being sulfonanilide, and it preferentially inhibits COX-2 [65]. It remains to be established whether the inhibitory effects of nimesulide on human mast cells are related to selective COX-2 inhibition or whether the effects of this drug are more complex than those of NSAIDs.

Sphingosine kinase 1 inhibitors

Sphingosine kinase (SphK) is a key enzyme in the sphingolipid metabolic pathway that phosphorylates sphingosine into sphingosine 1-phosphate (S1P) [66]. To date, two mammalian SphKs have been cloned, sequenced and characterized, *SPHK1* and *SPHK2*. Most of the effects exerted by S1P on the immune system are mediated by five specific S1P receptors, S1P_{1–5}, that belong to the family of G protein-coupled receptors.

Human mast cells express S1P₁ and S1P₂ [67]. FcεRI cross-linking on mast cells leads to the production and secretion of S1P through the activation of SphKs [68–73]. Although this process is involved in mast cell activation, the relative contribution of each SphK isoenzyme is still uncertain. Interestingly, it has been recently reported that the specific SphK1 inhibitor SK1-I reduces FcεRI-triggered degranulation and cytokine and chemokine secretion by human and mouse mast cells [74^{••}]. In a mouse model of allergic asthma that strictly depends on mast cells [75,76], SK1-I suppresses the development of airway inflammation, hyperresponsiveness, and goblet cell hyperplasia. Conversely, SphK2-deleted mice have exacerbated lung inflammatory responses. Thus, SphK1 may be a promising target for the treatment of allergic airway inflammation and mast cell-mediated disorders.

Tyrosine kinase inhibitors

Tyrosine phosphorylation of signaling proteins is a key step in mast cell activation and mediator release in response to FcεRI aggregation and SCF-induced dimerization of its receptor KIT [77]. KIT is endowed with intrinsic tyrosine kinase activity, while FcεRI is devoid of enzymatic activity and relies on cytosolic tyrosine kinases to transduce intracellular signals.

SCF is the principal growth factor for mast cells and is produced mainly by stromal cells (e.g. fibroblasts) [78–81]. It is the key cytokine that regulates the differentiation and maturation of mast cells, induces mast cell chemotaxis and potentiates antigen-mediated mast cell activation [6[•]]. Mastocytosis, a disorder characterized by abnormal clonal proliferation of mast cells, is frequently associated with a somatically acquired activating

mutation of KIT, most commonly the aspartic acid to valine substitution at residue 816 (KIT^{d816v}) [82,83]. Thus, inhibition of KIT activity may have a profound effect on the pathogenesis of mastocytosis. The tyrosine kinase inhibitors (TKIs) developed so far (e.g. imatinib and dasatinib) can suppress the activity of wild-type KIT but not its mutated form, KIT^{d816v} [84]. Thus, these drugs have only modest efficacy often accompanied by significant side effects, probably due to inhibition of other tyrosine kinases [85]. Novel TKIs as well as associations of TKIs are currently being evaluated for their *in vitro* and *in vivo* efficacy in modulating mast cell activation [86[•],87]. Since SCF potentiates FcεRI-mediated signaling in normal mast cells, studies are evaluating whether inhibition of KIT signaling is associated with the modulation of allergen-induced responses. The TKIs nilotinib and sunitinib prevent mast cell activation in *in vivo* models of anaphylaxis [88,89]. Hypothemycin, a compound that inhibits both KIT kinase activity and FcεRI-mediated signaling, efficiently suppresses mast cell activation *in vitro* and the development of antigen-induced passive cutaneous anaphylaxis [90].

FcεRI-mediated signaling is strictly dependent on the cytosolic spleen tyrosine kinase (Syk) in both mast cells [77,91] and basophils [92–94]. Several Syk inhibitors have been synthesized and have shown promise in such diseases as allergic rhinitis [95,96], asthma [97–101], rheumatoid arthritis [102] and B cell malignancies [103]. However, caution should be exercised in defining Syk inhibitors as ‘specific’ inhibitors of FcεRI⁺ cells. Syk is expressed widely in cells of the innate and adaptive immune system [104] as well as in non-hematopoietic cells under certain circumstances [105–107]. Thus, the relative contribution of mast cells to diseases in which Syk inhibitors have proved efficacious should be evaluated in the light of the above observations.

Immunophilin ligands and inhibitors of mTOR

Handschumacher *et al.* were the first to identify a family of proteins with high affinity for cyclosporin A (CsA) [108]. This protein, named ‘cyclophilin’ (CyP) displayed high affinity for CsA and low affinity for inactive analogs such as CsH. CyP belongs to a family of intracellular proteins, the immunophilins, that include the FK-binding proteins (FKBPs) [109]. Three natural macrolides, FK506 (tacrolimus), rapamycin (RAP), and ascomycin bind with high affinity to FKBP [110]. CsA-CyP complexes, as well as FK506-FKBP12 complexes, bind to and inhibit the phosphatase calcineurin [111], thus suppressing lymphocyte [110], basophil [112,113] and mast cell [114–116] activation. Of note, topical treatment with tacrolimus (or its related compound pimecrolimus) is effective for patients with atopic dermatitis [117,118]. Although skin mast cells are thought to play a role in atopic dermatitis [119^{••},120^{••},121^{••},122], the efficacy of tacrolimus is likely related also to the inhibition of T cell activity. Furthermore,

pimecrolimus induces apoptosis of mast cells in a murine model of cutaneous mastocytosis [123].

RAP interacts with FKBP12 and inhibits the mammalian target of rapamycin (mTOR), but does not inhibit calcineurin. mTOR is a serine/threonine kinase that regulates different pathways depending on its interaction with two regulatory proteins, raptor and rictor, that form mTOR complex 1 (mTORC1) and mTORC2, respectively [124]. RAP has no effect on FcεRI-mediated degranulation, but inhibits cytokine production and KIT mediated chemotaxis and cell survival by inhibiting mTORC1 [22,125]. On the other hand, mTORC2 regulates the proliferation of immature and neoplastic mast cells, but it hardly affects mature mast cells [126]. Thus, selective targeting of mTORCs may be used to modulate different aspects of mast cell biology.

Histamine receptor antagonists

H₁-antihistamines antagonize the effects of histamine at the H₁ receptor (H₁R) and are widely used in the treatment of several allergic conditions [127]. It has been known since 1975 that high concentrations of first-generation antihistamines inhibit antigen-induced histamine release from human basophils *in vitro* [128]. Oxatomide inhibits the release of both preformed and *de novo* synthesized mediators from mast cells [129,130]. Rupatadine, an antihistamine that also inhibits the effects of platelet-activating factor, suppresses mediator release from LAD2 cells and hCBMC [131,132]. Loratadine and desloratadine inhibit IgE-dependent and IgE-independent release of preformed and *de novo* synthesized mediators from HSMC [133,134]. Not all H₁R antagonists exert such activity *in vitro*. For instance, cetirizine prevents mediator release from HLMC but not from HSMC [135,136], whereas mizolastine inhibits only the *de novo* synthesis of LTC₄ from mast cells [137]. Taken together, these results indicate that the inhibitory effects of certain H₁R antagonists on human mast cells are not class-specific.

The most recently identified histamine receptor 4 (H₄R) is mainly expressed on hematopoietic cells, particularly eosinophils, neutrophils, mast cells, dendritic cells, T cells, and basophils [138]. H₄R activation mediates chemotaxis of mast cells [139]. Recently, it has been reported that an H₄R antagonist/inverse agonist suppresses FcεRI-mediated murine mast cell degranulation *in vitro* only if present during sensitization with IgE, at least in part by inhibiting IgE-induced FcεRI upregulation [140*].

Protein kinase C inhibitors

FcεRI-mediated signaling is coupled to phospholipase Cγ (PLCγ) activation. PLCγ cleaves phosphatidylinositol 4,5-bisphosphate (PIP₂) into inositol 1,4,5-trisphosphate (IP₃) and diacylglycerol (DAG). IP₃ induces the release of Ca²⁺ from its store, while DAG is an activator of the serine/threonine kinase protein kinase C (PKC). These

events are critical for mast cell degranulation and cytokine release [141]. The PKC family comprises a group of closely related protein kinases divided into three major groups: the calcium-dependent or conventional PKC (α, βI, βII and γ), the calcium-independent isoenzymes (δ, ε, η, θ and μ) and the atypical PKC (ζ and λ) [142]. Both calcium-dependent and calcium-independent PKC have been involved in FcεRI signaling in mast cells [143–149]; some of them have an inhibitory effect [150,151]. Interestingly, the PKC activators phorbol esters (e.g. PMA) and bryostatins induce histamine release from basophils [152–154] but inhibit FcεRI-mediated activation of HLMC and HSMC [152,155].

A large number of putative specific inhibitors of different PKC isoforms have been developed [142,156] and some of them are being evaluated in clinical trials. However, thus far, the results of clinical trials of PKC modulators have been disappointing, largely owing to inadequate specificity and/or adverse effects. Many PKC inhibitors also affect other protein kinases and multiple PKC isoenzymes are expressed in most cell types. Thus, it is conceivable that PKC inhibitors may modulate cell types other than mast cells. Nevertheless, this family of kinases remains an enticing target for mast cell-mediated disorders.

Blockers of ion channels

FcεRI-mediated activation of mast cells is characterized by an influx of extracellular Ca²⁺ that is essential for the secretion of mediators. Mast cells express calcium channels as well as K⁺, Cl[−] and transient receptor potential (TRP) channels that, by modulating cell membrane potential, also play a role in cell activation [157]. HLMC express the Ca²⁺-activated K⁺ channel K_{Ca}3.1, and the K_{Ca}3.1 blocker TRAM-34 inhibits HLMC migration [158]. More recently, the CRACM/Orai Ca²⁺ channels were detected in HLMC. Selective blockers of these channels reduce FcεRI-dependent mediator release *in vitro* and allergen-induced bronchial smooth muscle contraction in isolated human bronchus [159*]. Although ion channels represent promising targets for a novel class of mast cell inhibitors, it is likely that the *in vivo* effects of ion channel blockers will be related also to the modulation of other cell types (e.g. lymphocytes) [160].

Omalizumab and new strategies to block the IgE-FcεRI interaction

Omalizumab is a humanized monoclonal antibody directed against IgE which can benefit patients with moderate to severe asthma [161]. The benefit of omalizumab treatment has been attributed to its ability to reduce the concentration of free IgE in the blood, which can in turn lower the expression of FcεRI on basophils, mast cells and perhaps other cell types [162–164]. Recent evidence indicates that omalizumab provides rapid clinical benefit in patients with chronic idiopathic urticaria, a disorder characterized by

recurrent episodes of HSMC activation [165,166**]. Indeed, omalizumab treatment in patients with allergic rhinitis reduces both the expression of FcεRI on HSMC and immediate skin test reactivity to allergens. This phenotype is reverted after cessation of omalizumab treatment [162,167].

Several strategies have been devised to interfere with FcεRI-mediated activation of mast cells. Small peptides have been used to inhibit the binding of IgE to FcεRI [168–170]. Some of these peptides prevent mast cell activation *in vitro* and *in vivo*. Another way of blocking IgE-FcεRI interaction is based on DNA oligonucleotides that bind to human IgE with high affinity and specificity [171]. These oligonucleotides inhibit the interaction of IgE with FcεRI and mediator release *in vitro*. Finally, macromolecular inhibitors named ‘designed ankyrin repeat proteins’ (referred to as DARPs) with specificity to FcεRIα and/or the constant region of IgE have been generated [172,173,174**]. Interestingly, these inhibitors not only prevent the interaction between IgE and FcεRI, but also induce the dissociation of preformed IgE-FcεRI complexes.

Mast cell inhibitory receptors

Inhibitory receptors comprise two main families of the Ig receptor super-family and the C-type (calcium dependent) lectin superfamily. Inhibitory receptors contain immunoreceptor tyrosine-based inhibition motifs (ITIMs) that downregulate the activation signals transmitted through immunoreceptor tyrosine-based inhibitory motifs (ITAMs). Upon activation of ITIM-containing receptors, tyrosine residues within the motifs become phosphorylated. This leads to the recruitment of protein phosphatases, Src homology 2 (SH) domain-containing protein tyrosine phosphatase-1/2 (SHP-1 and SHP-2) and lipid phosphatases SH2 domain-bearing inositol phosphatase (SHIP1). SHP-1/2 inhibits the action of tyrosine kinase, while SHIP1 terminates the PI3K-mediated pathway [175,176]. Several inhibitory receptors on mast cells have been characterized, among which, FcγRIIB, CD300, CD72, and sialic acid binding Ig-like lectin (Siglec) 8, Allergen-1 and mast cell function-associated antigen (MAFA).

FcγRIIB is a low affinity IgG receptor. Co-aggregation of FcγRIIB and FcεRI suppresses FcεRI-mediated activation [175]. Interestingly, a chimeric protein composed of key portions of the human γ1 and the human ε immunoglobulin heavy chains can inhibit FcεRI-mediated activation of human basophils *in vitro* and FcεRI-mediated degranulation of murine mast cell expressing the human FcεRI in an *in vivo* model of passive cutaneous anaphylaxis [177].

The CD300 molecules are a family of immunoglobulin receptors that includes activating (CD300b, CD300e) and inhibitory (CD300a, CD300f) members. CD300a and

CD300f, and their respective murine orthologs LMIR1 and LMIR3, are expressed by mast cells [178,179]. LMIR1/CD300a cross-linking inhibits both FcεRI-dependent and SCF-dependent signaling, thus impairing mast cell differentiation, survival and activation [180]. Interestingly, bispecific antibodies that coaggregate LMIR1/CD300a with either FcεRI or KIT inhibit allergic responses *in vivo* [181,182]. Recently, phosphatidylethanolamine and phosphatidylserine have been identified as the natural ligands for LMIR1/CD300a [183*,184]. Phosphatidylserine is expressed on the outer leaflet of the plasma membrane of apoptotic cells and suppresses LPS-induced mast cell activation via interaction with LMIR1/CD300a [185*]. Furthermore, LMIR3/CD300f binding to ceramide and sphingomyelin inhibits FcεRI-mediated activation of mast cells *in vitro* and *in vivo* [186**,187**]. Thus, the interaction between extracellular lipids and CD300 family members exerts an important role in modulating mast cell function.

Siglecs are a group of sialic acid-binding cell surface proteins predominantly expressed by immune cells. Among them, Siglec-8 is expressed on human eosinophils, mast cells, and to a lesser extent on basophils [188]. Although cross-linking of Siglec-8 (and of its mouse ortholog Siglec-F) induces eosinophil apoptosis, it does not affect mast cell viability. Instead, Siglec-8 cross-linking inhibits FcεRI-dependent histamine and PGD₂ release from human mast cells [189]. However, mouse mast cells do not express Siglec-F, which makes it difficult to understand its function on these cells *in vivo*.

CD72 is expressed on several immune cells, including mast cells [190,191]. CD72 cross-linking or activation with its ligand CD100 reduces KIT-mediated chemotaxis, MCP-1/CCL2 production of hPBMCs and suppresses the growth of the HMC-1.2 mast cell line that harbors the gain-of-function mutation in the KIT gene [190].

Allergen-1 is an immunoglobulin-like inhibitory receptor expressed mainly (but not exclusively) by human and mouse mast cells [192,193]. Allergen-1 suppresses IgE-mediated activation of mouse mast cells *in vitro* and the development of passive systemic and cutaneous anaphylaxis *in vivo* [192]. Furthermore, coligation of Allergen-1 and FcεRI reduces antigen-induced CD107a expression, a marker of human mast cell activation [194], on human culture-derived and primary mast cells [193].

MAFA, also named Killer cell lectin-like receptor subfamily G member 1 (KLRG1), is an inhibitory receptor belonging to the C-type lectin superfamily [195]. MAFA aggregation reduces degranulation, *de novo* synthesis and release of leukotrienes and secretion of some cytokines [196]. Interestingly, MAFA inhibition of FcεRI signaling does not strictly require their coligation. Although MAFA was identified on rat basophilic leukemia 2H3 (RBL-2H3)

Table 1

Old and new therapeutic approaches for mast cell-mediated disorders

Class of modulators	Effects on mast cell activity	References
β 2-Adrenoreceptor agonists	β 2-agonists increase the intracellular concentration of cAMP and reduce mast cell mediator release. This effect is suppressed by long-term preincubation with β 2-agonists or SCF stimulation	[27–31,34]
PDE inhibitors	Non-selective PDE inhibitors negatively modulate the activation of HLMC and HSMC, while selective PDE4 inhibitors are ineffective	[39,40]
Cromones	Cromones inhibit mast cell mediator release, at least in part by releasing annexin A ₁	[41,42,43*]
Glucocorticoids	Glucocorticoids reduce the number of mast cells in human inflamed tissues and inhibit mast cell degranulation, cytokine and chemokine production	[49*,50–55,58–60]
NSAIDs	Among NSAIDs, only nimesulide and its active metabolite 4-hydroxy-nimesulide can inhibit mediator release from mast cells	[22,61–63]
Sphk1 inhibitors	The Sphk1 inhibitor SK1-I reduces mast cell activation <i>in vitro</i> and suppresses airway inflammation in a mast cell-dependent mouse model of allergic asthma	[74**]
TKIs	KIT inhibitors suppress the activity of wild-type but not mutated (D816V) KIT. Thus, these drugs are ineffective in mastocytosis. Novel TKIs and associations of TKIs are being evaluated for their efficacy in mastocytosis and allergen-induced responses. Syk inhibitors have shown promise in such diseases as allergic rhinitis, asthma and rheumatoid arthritis, although the relative contribution of mast cells to these disorders needs to be clarified	[84,85,86*, 87–90,95–103]
Calcineurin inhibitors	CsA and FK506 (tacrolimus) suppress mast cell activation <i>via</i> inhibition of the phosphatase calcineurin. Tacrolimus and its related compound pimecrolimus are effective in the treatment of atopic dermatitis. Pimecrolimus induces mast cells apoptosis in a murine model of cutaneous mastocytosis	[114–117,122]
mTOR inhibitors	Rapamycin does not affect Fc ϵ RI-mediated degranulation, but inhibits cytokine production and KIT-activated chemotaxis and cell survival	[22,124]
Histamine receptor antagonists	Some, but not all, H1R antagonists inhibit mediator release from mast cells. An H4R antagonist/inverse agonist suppresses Fc ϵ RI-mediated murine mast cell degranulation <i>in vitro</i> . Furthermore, H4R mediate chemotaxis of mast cells	[127–136,138,139]
PKC inhibitors	PKC play a prominent role in Fc ϵ RI-activated signaling in mast cells, and some of them exert an inhibitory effect. To date, there is no clear-cut evidence about the efficacy of PKC inhibitors in mast cell-mediated disorders	[140*,141–154]
Ion channel blockers	The K _{Ca} 3.1 blocker TRAM-34 inhibits HLMC migration. Selective blockers of CRACM/Orai Ca ²⁺ channels reduce Fc ϵ RI-dependent mediator release <i>in vitro</i> and allergen-induced bronchial smooth muscle contraction in isolated human bronchus	[157,158]
Omalizumab	Omalizumab reduces the concentration of free IgE in the blood, which can in turn lower the expression of Fc ϵ RI on basophils, mast cells and perhaps other cell types. Omalizumab is effective in patients with chronic idiopathic urticaria, a disorder characterized by recurrent episodes of HSMC activation	[161–165,166**]
Modulators of IgE-Fc ϵ RI interaction	Small peptides can inhibit the binding of IgE to Fc ϵ RI and suppress mast cell activation <i>in vitro</i> and <i>in vivo</i> . Appropriately designed DNA oligonucleotides bind to IgE thereby preventing its interaction with Fc ϵ RI and inhibit mediator release <i>in vitro</i> . Macromolecular inhibitors (DARPs) with specificity to Fc ϵ RI α and/or the constant region of IgE have been generated. Interestingly, DARPs can also dissociate preformed IgE-Fc ϵ RI complexes	[167–173]
Inhibitory receptors	Coaggregation of the low affinity IgG receptor Fc γ RIIB with Fc ϵ RI suppresses Fc ϵ RI-mediated activation. LMIR1/CD300a crosslinking, coaggregation with Fc ϵ RI or KIT, or binding to its ligand (phosphatidylserine) impairs mast cell responses <i>in vitro</i> and <i>in vivo</i> . Likewise, LMIR3/CD300f binding to ceramide and sphingomyelin inhibits Fc ϵ RI-mediated mast cell activation <i>in vitro</i> and <i>in vivo</i> . Siglec-8 crosslinking suppresses human mast cell mediator release. CD72 crosslinking or engagement by its ligand CD100 reduces KIT-mediated chemotaxis and chemokine production of human mast cells and impairs proliferation of neoplastic mast cells. Allergin-1 suppresses IgE-mediated activation of human and mouse mast cells. MAFA aggregation reduces degranulation, <i>de novo</i> synthesis and release of leukotrienes and secretion of some cytokines by RBL-2H3 cells.	[174**,175–182, 183*,184,185*, 186**,187**, 188–195]

Abbreviations: CsA, cyclosporine A; DARPs, designed ankyrin repeat proteins; PDE, phosphodiesterase; H1R, histamine 1 receptor; H4R, histamine 4 receptor; HLMC, human lung mast cells; HSMC, human skin mast cells; mTOR, mammalian target of rapamycin; NSAIDs, non steroidal anti-inflammatory drugs; PKC, protein kinase C; RBL-2H3, rat basophilic leukemia 2H3; SCF, stem cell factor; Sphk1, sphingosine kinase 1; Syk, spleen tyrosine kinase; TKIs, tyrosine kinase inhibitors.

cells, its mouse and human homologs are expressed mainly by NK and T cells. Thus, its *in vivo* relevance in modulating mast cell biology has yet to be identified.

In conclusion, mast cells express a wide spectrum of inhibitory receptors that can modulate the activation of FcεRI and KIT. Thus, inhibitory receptors may become targets for the modulation of mast cell mediated-immune disorders.

Conclusions

To date, several drugs that can modulate mast cell function *in vitro* and/or *in vivo* have been identified. Although their mechanism of action is not always completely understood and their *in vivo* efficacy probably involves other cell types, these compounds have uncovered several aspects of mast cell biology. Furthermore, the study of surface receptors and signaling pathways associated with mast cell activation has suggested novel therapeutic targets. Some of them have already been tested in preclinical models (Table 1).

Nevertheless, some considerations should be kept in mind in relation to pharmacologic targeting of mast cells. Mast cells isolated from humans and different animal models, as well as mast cells purified from different anatomic sites, differ biochemically, immunologically and pharmacologically. In addition, different stimuli can trigger distinct signaling events leading to the selective release of mediators. It is therefore not surprising that certain pharmacological agents can selectively modulate the release of mediators induced by some, but not all stimuli. Increasing evidence indicates that mast cells and their mediators play a beneficial role in host resistance against infections [21] and animal venoms [16,17[•],18,19[•],20]. Targeting mast cells with drugs or perhaps agents more specific than those available today might alter a delicate balance of physiological processes to which these cells contribute. Akin to all new therapeutic approaches, appropriately designed studies must be performed to evaluate whether the benefits of mast cell-targeted treatment outweigh any potential risks of adverse effects.

Conflict of interest statement

Nothing declared.

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References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as:

- of special interest
- of outstanding interest

1. Marone G, Lichtenstein LM, Galli S: *Mast Cells and Basophils*. Academic Press; 2000.

2. Galli SJ, Kalesnikoff J, Grimbaldeston MA, Piliponsky AM, Williams CM, Tsai M: **Mast cells as 'tunable' effector and immunoregulatory cells: recent advances**. *Annu Rev Immunol* 2005, **23**:749-786.
3. Kirshenbaum AS, Kessler SW, Goff JP, Metcalfe DD: **Demonstration of the origin of human mast cells from CD34+ bone marrow progenitor cells**. *J Immunol* 1991, **146**:1410-1415.
4. Rottem M, Okada T, Goff JP, Metcalfe DD: **Mast cells cultured from the peripheral blood of normal donors and patients with mastocytosis originate from a CD34+/Fc epsilon RI- cell population**. *Blood* 1994, **84**:2489-2496.
5. Valent P, Spanblochl E, Sperr WR, Sillaber C, Zsebo KM, Agis H, Strobl H, Geissler K, Bettelheim P, Lechner K: **Induction of differentiation of human mast cells from bone marrow and peripheral blood mononuclear cells by recombinant human stem cell factor/klt-ligand in long-term culture**. *Blood* 1992, **80**:2237-2245.
6. Galli SJ, Tsai M: **IgE and mast cells in allergic disease**. *Nat Med* 2012, **18**:693-704.
State of the art review on the biology of IgE and mast cells and their roles in the allergic response.
7. Metcalfe DD: **Mast cells and mastocytosis**. *Blood* 2008, **112**:946-956.
8. Reber LL, Marichal T, Galli SJ: **New models for analyzing mast cell functions in vivo**. *Trends Immunol* 2012, **33**:613-625.
The authors illustrate the results obtained using new mouse models that allow more selective mast targeting *in vivo* and compare them to the commonly used KIT-dependent mouse models of mast cell deficiency.
9. Dudeck A, Dudeck J, Scholten J, Petzold A, Surianarayanan S, Kohler A, Peschke K, Vohringer D, Waskow C, Krieg T et al.: **Mast cells are key promoters of contact allergy that mediate the adjuvant effects of haptens**. *Immunity* 2011, **34**:973-984.
10. Feyerabend TB, Weiser A, Tietz A, Stassen M, Harris N, Kopf M, Radermacher P, Moller P, Benoist C, Mathis D et al.: **Cre-mediated cell ablation contests mast cell contribution in models of antibody- and T cell-mediated autoimmunity**. *Immunity* 2011, **35**:832-844.
11. Lilla JN, Chen CC, Mukai K, BenBarak MJ, Franco CB, Kalesnikoff J, Yu M, Tsai M, Piliponsky AM, Galli SJ: **Reduced mast cell and basophil numbers and function in Cpa3-Cre; Mcl-1fl/fl mice**. *Blood* 2011, **118**:6930-6938.
12. Detoraki A, Staiano RI, Granata F, Giannattasio G, Prevete N, de Paulis A, Ribatti D, Genovese A, Triggiani M, Marone G: **Vascular endothelial growth factors synthesized by human lung mast cells exert angiogenic effects**. *J Allergy Clin Immunol* 2009, **123**:1142-1149 e1141-1145.
13. Melillo RM, Guarino V, Avilla E, Galdiero MR, Liotti F, Prevete N, Rossi FW, Basolo F, Ugolini C, de Paulis A et al.: **Mast cells have a protumorigenic role in human thyroid cancer**. *Oncogene* 2010, **29**:6203-6215.
14. Rabenhorst A, Schlaak M, Heukamp LC, Forster A, Theurich S, von Bergwelt-Baildon M, Buttner R, Kurschat P, Mauch C, Roers A et al.: **Mast cells play a protumorigenic role in primary cutaneous lymphoma**. *Blood* 2012, **120**:2042-2054.
15. Jensen-Jarolim E, Achatz G, Turner MC, Karagiannis S, Legrand F, Capron M, Penichet ML, Rodriguez JA, Siccardi AG, Vangelista L et al.: **AllergoOncology: the role of IgE-mediated allergy in cancer**. *Allergy* 2008, **63**:1255-1266.
16. Akahoshi M, Song CH, Piliponsky AM, Metz M, Guzzetta A, Abrink M, Schlenner SM, Feyerabend TB, Rodewald HR, Pejler G et al.: **Mast cell chymase reduces the toxicity of Gila monster venom, scorpion venom, and vasoactive intestinal polypeptide in mice**. *J Clin Invest* 2011, **121**:4180-4191.
17. Marichal T, Starkl P, Reber LL, Kalesnikoff J, Oettgen HC, Tsai M, •• Metz M, Galli SJ: **A beneficial role for immunoglobulin E in host defense against honeybee venom**. *Immunity* 2013, **39**:963-975.
Together with [19[•]], these landmark papers support the 'toxin hypothesis' conceived by Margie Profet in 1991 by demonstrating that the IgE response exerts a beneficial role (i.e. it protects the host against animal venom).

18. Metz M, Piliponsky AM, Chen CC, Lammell V, Abrink M, Pejler G, Tsai M, Galli SJ: **Mast cells can enhance resistance to snake and honeybee venoms.** *Science* 2006, **313**:526-530.
19. Palm NW, Rosenstein RK, Yu S, Schenten DD, Florsheim E, Medzhitov R: **Bee venom phospholipase A2 induces a primary type 2 response that is dependent on the receptor ST2 and confers protective immunity.** *Immunity* 2013, **39**:976-985.
- See annotation to Ref. [17**].
20. Schneider LA, Schlenner SM, Feyerabend TB, Wunderlin M, Rodewald HR: **Molecular mechanism of mast cell mediated innate defense against endothelin and snake venom sarafotoxin.** *J Exp Med* 2007, **204**:2629-2639.
21. Chan CY, St John AL, Abraham SN: **Plasticity in mast cell responses during bacterial infections.** *Curr Opin Microbiol* 2012, **15**:78-84.
22. de Paulis A, Ciccarelli A, Marino I, de Crescenzo G, Marino D, Marone G: **Human synovial mast cells II. Heterogeneity of the pharmacologic effects of antiinflammatory and immunosuppressive drugs.** *Arthritis Rheum* 1997, **40**:469-478.
23. Gurish MF, Austen KF: **Developmental origin and functional specialization of mast cell subsets.** *Immunity* 2012, **37**:25-33.
24. Halova I, Draberova L, Draber P: **Mast cell chemotaxis – chemoattractants and signaling pathways.** *Front Immunol* 2012, **3**:119.
25. Harvima IT, Levi-Schaffer F, Draber P, Friedman S, Polakovicova I, Gibbs BF, Blank U, Nilsson G, Maurer M: **Molecular targets on mast cells and basophils for novel therapies.** *J Allergy Clin Immunol* 2014 <http://dx.doi.org/10.1016/j.jaci.2014.03.0077>.
26. Barnes PJ: **Drugs for asthma.** *Br J Pharmacol* 2006, **147**(Suppl. 1):S297-S303.
27. Barnes PJ: **Effect of beta-agonists on inflammatory cells.** *J Allergy Clin Immunol* 1999, **104**:S10-S17.
28. Okayama Y, Church MK: **Comparison of the modulatory effect of ketotifen, sodium cromoglycate, procaterol and salbutamol in human skin, lung and tonsil mast cells.** *Int Arch Allergy Immunol* 1992, **97**:216-225.
29. Shichijo M, Inagaki N, Nakai N, Kimata M, Nakahata T, Serizawa I, Ikura Y, Saito H, Nagai H: **The effects of anti-asthma drugs on mediator release from cultured human mast cells.** *Clin Exp Allergy* 1998, **28**:1228-1236.
30. Wang XS, Lau HY: **Beta-adrenoceptor-mediated inhibition of mediator release from human peripheral blood-derived mast cells.** *Clin Exp Pharmacol Physiol* 2006, **33**:746-750.
31. Scola AM, Loxham M, Charlton SJ, Peachell PT: **The long-acting beta-adrenoceptor agonist, indacaterol, inhibits IgE-dependent responses of human lung mast cells.** *Br J Pharmacol* 2009, **158**:267-276.
32. Lukacs NW, Kunkel SL, Strieter RM, Evanoff HL, Kunkel RG, Key ML, Taub DD: **The role of stem cell factor (c-kit ligand) and inflammatory cytokines in pulmonary mast cell activation.** *Blood* 1996, **87**:2262-2268.
33. Takaishi T, Morita Y, Hirai K, Yamaguchi M, Ohta K, Noda E, Morita T, Ito K, Miyamoto T: **Effect of cytokines on mediator release from human dispersed lung mast cells.** *Allergy* 1994, **49**:837-842.
34. Cruse G, Yang W, Duffy SM, Chachi L, Leyland M, Amrani Y, Bradding P: **Counterregulation of beta(2)-adrenoceptor function in human mast cells by stem cell factor.** *J Allergy Clin Immunol* 2010, **125**:e251-e255 257-263.
35. de Paulis A, Minopoli G, Arbustini E, de Crescenzo G, Dal Piaz F, Pucci P, Russo T, Marone G: **Stem cell factor is localized in, released from, and cleaved by human mast cells.** *J Immunol* 1999, **163**:2799-2808.
36. Al-Muhsen SZ, Shablovsky G, Olivenstein R, Mazer B, Hamid Q: **The expression of stem cell factor and c-kit receptor in human asthmatic airways.** *Clin Exp Allergy* 2004, **34**:911-916.
37. Da Silva CA, Blay F, Israel-Biet D, Laval AM, Glasser N, Pauli G, Frossard N: **Effect of glucocorticoids on stem cell factor expression in human asthmatic bronchi.** *Clin Exp Allergy* 2006, **36**:317-324.
38. Page CP, Spina D: **Selective PDE inhibitors as novel treatments for respiratory diseases.** *Curr Opin Pharmacol* 2012, **12**:275-286.
39. Eskandari N, Bastan R, Peachell PT: **Regulation of human skin mast cell histamine release by PDE inhibitors.** *Allergol Immunopathol (Madr)* 2013 <http://dx.doi.org/10.1016/j.aller.2013.07.011>.
40. Weston MC, Anderson N, Peachell PT: **Effects of phosphodiesterase inhibitors on human lung mast cell and basophil function.** *Br J Pharmacol* 1997, **121**:287-295.
41. Foreman JC, Garland LG: **Cromoglycate and other antiallergic drugs: a possible mechanism of action.** *Br Med J* 1976, **1**:820-821.
42. Mazurek N, Bashkin P, Loyter A, Pecht I: **Restoration of Ca²⁺ influx and degranulation capacity of variant RBL-2H3 cells upon implantation of isolated cromolyn binding protein.** *Proc Natl Acad Sci U S A* 1983, **80**:6014-6018.
43. Yazid S, Sinniah A, Solito E, Calder V, Flower RJ: **Anti-allergic cromones inhibit histamine and eicosanoid release from activated human and murine mast cells by releasing Annexin A1.** *PLOS ONE* 2013, **8**:e58963.
- This article demonstrates that cromones induce PKC-dependent phosphorylation and release of Annexin A1. This event is associated with inhibition of IgE-mediated histamine and PGD2 release from human and murine mast cells.
44. Yazid S, Leoni G, Getting SJ, Cooper D, Solito E, Perretti M, Flower RJ: **Antiallergic cromones inhibit neutrophil recruitment onto vascular endothelium via annexin-A1 mobilization.** *Arterioscler Thromb Vasc Biol* 2010, **30**:1718-1724.
45. Yazid S, Solito E, Christian H, McArthur S, Goulding N, Flower R: **Cromoglycate drugs suppress eicosanoid generation in U937 cells by promoting the release of Anx-A1.** *Biochem Pharmacol* 2009, **77**:1814-1826.
46. Oka T, Kalesnikoff J, Starkl P, Tsai M, Galli SJ: **Evidence questioning cromolyn's effectiveness and selectivity as a 'mast cell stabilizer' in mice.** *Lab Invest* 2012, **92**:1472-1482.
47. Velema WA, van der Toorn M, Szymanski W, Feringa BL: **Design, synthesis, and inhibitory activity of potent, photoswitchable mast cell activation inhibitors.** *J Med Chem* 2013, **56**:4456-4464.
- This article describes two novel mast cell inhibitors 100 times more potent than DSCG. These inhibitors can be used to develop novel antiallergic drugs as well as photoswitchable compounds for the study of allergic reactions.
48. Stellato C: **Glucocorticoid actions on airway epithelial responses in immunity: functional outcomes and molecular targets.** *J Allergy Clin Immunol* 2007, **120**:1245-1264 1247-1263; quiz.
49. Oppong E, Flink N, Cato AC: **Molecular mechanisms of glucocorticoid action in mast cells.** *Mol Cell Endocrinol* 2013, **380**:119-126.
- This is a comprehensive review that illustrates how glucocorticoids modulate mast cell activity at different levels.
50. Benhamou M, Ninio E, Salem P, Hieblot C, Bessou G, Pitton C, Liu FT, Mencia-Huerta JM: **Decrease in IgE Fc receptor expression on mouse bone marrow-derived mast cells and inhibition of paf-acether formation and of beta-hexosaminidase release by dexamethasone.** *J Immunol* 1986, **136**:1385-1392.
51. Robin JL, Seldin DC, Austen KF, Lewis RA: **Regulation of mediator release from mouse bone marrow-derived mast cells by glucocorticoids.** *J Immunol* 1985, **135**:2719-2726.
52. Yamaguchi M, Hirai K, Komiya A, Miyamasu M, Furumoto Y, Teshima R, Ohta K, Morita Y, Galli SJ, Ra C *et al.*: **Regulation of mouse mast cell surface Fc epsilon RI expression by dexamethasone.** *Int Immunol* 2001, **13**:843-851.
53. Zhou J, Liu DF, Liu C, Kang ZM, Shen XH, Chen YZ, Xu T, Jiang CL: **Glucocorticoids inhibit degranulation of mast cells in allergic asthma via nongenomic mechanism.** *Allergy* 2008, **63**:1177-1185.

54. Goldsmith P, McGarity B, Walls AF, Church MK, Millward-Sadler GH, Robertson DA: **Corticosteroid treatment reduces mast cell numbers in inflammatory bowel disease.** *Dig Dis Sci* 1990, **35**:1409-1413.
 55. Lavker RM, Schechter NM: **Cutaneous mast cell depletion results from topical corticosteroid usage.** *J Immunol* 1985, **135**:2368-2373.
 56. Finotto S, Mekori YA, Metcalfe DD: **Glucocorticoids decrease tissue mast cell number by reducing the production of the c-kit ligand, stem cell factor, by resident cells: *in vitro* and *in vivo* evidence in murine systems.** *J Clin Invest* 1997, **99**:1721-1728.
 57. Kassel O, Schmidlin F, Duvernelle C, de Blay F, Frossard N: **Up- and down-regulation by glucocorticoids of the constitutive expression of the mast cell growth factor stem cell factor by human lung fibroblasts in culture.** *Mol Pharmacol* 1998, **54**:1073-1079.
 58. Wershil BK, Furuta GT, Lavigne JA, Choudhury AR, Wang ZS, Galli SJ: **Dexamethasone or cyclosporin A suppress mast cell-leukocyte cytokine cascades. Multiple mechanisms of inhibition of IgE- and mast cell-dependent cutaneous inflammation in the mouse.** *J Immunol* 1995, **154**:1391-1398.
 59. Kato A, Chustz RT, Ogasawara T, Kulka M, Saito H, Schleimer RP, Matsumoto K: **Dexamethasone FK506 inhibit expression of distinct subsets of chemokines in human mast cells.** *J Immunol* 2009, **182**:7233-7243.
 60. Matsuda K, Piliponsky AM, Iikura M, Nakae S, Wang EW, Dutta SM, Kawakami T, Tsai M, Galli SJ: **Monomeric IgE enhances human mast cell chemokine production: IL-4 augments and dexamethasone suppresses the response.** *J Allergy Clin Immunol* 2005, **116**:1357-1363.
 61. Casolaro V, Melioli S, Marino O, Patella V, de Paulis A, Guidi G, Marone G: **Nimesulide, a sulfonanilide nonsteroidal anti-inflammatory drug, inhibits mediator release from human basophils and mast cells.** *J Pharmacol Exp Ther* 1993, **267**:1375-1385.
 62. Patella V, de Crescenzo G, Marino I, Genovese A, Adt M, Gleich GJ, Marone G: **Eosinophil granule proteins activate human heart mast cells.** *J Immunol* 1996, **157**:1219-1225.
 63. Ward A, Brogden RN: **Nimesulide. A preliminary review of its pharmacological properties and therapeutic efficacy in inflammation and pain states.** *Drugs* 1988, **36**:732-753.
 64. Andri L, Senna G, Betteli C, Giovanni S, Scaricabarozzi I, Mezzelani P, Andri G: **Tolerability of nimesulide in aspirin-sensitive patients.** *Ann Allergy* 1994, **72**:29-32.
 65. Bennett A, Villa G: **Nimesulide: an NSAID that preferentially inhibits COX-2, and has various unique pharmacological activities.** *Expert Opin Pharmacother* 2000, **1**:277-286.
 66. Spiegel S, Milstien S: **The outs and the ins of sphingosine-1-phosphate in immunity.** *Nat Rev Immunol* 2011, **11**:403-415.
 67. Oskeritzian CA, Alvarez SE, Hait NC, Price MM, Milstien S, Spiegel S: **Distinct roles of sphingosine kinases 1 and 2 in human mast-cell functions.** *Blood* 2008, **111**:4193-4200.
 68. Choi OH, Kim JH, Kinet JP: **Calcium mobilization via sphingosine kinase in signalling by the Fc epsilon RI antigen receptor.** *Nature* 1996, **380**:634-636.
 69. Jolly PS, Bektas M, Olivera A, Gonzalez-Espinosa C, Proia RL, Rivera J, Milstien S, Spiegel S: **Transactivation of sphingosine-1-phosphate receptors by Fc epsilon RI triggering is required for normal mast cell degranulation and chemotaxis.** *J Exp Med* 2004, **199**:959-970.
 70. Melendez AJ, Khaw AK: **Dichotomy of Ca²⁺ signals triggered by different phospholipid pathways in antigen stimulation of human mast cells.** *J Biol Chem* 2002, **277**:17255-17262.
 71. Olivera A, Mizugishi K, Tikhonova A, Ciaccia L, Odom S, Proia RL, Rivera J: **The sphingosine kinase-sphingosine-1-phosphate axis is a determinant of mast cell function and anaphylaxis.** *Immunity* 2007, **26**:287-297.
 72. Olivera A, Urtz N, Mizugishi K, Yamashita Y, Gilfillan AM, Furumoto Y, Gu H, Proia RL, Baumruker T, Rivera J: **IgE-dependent activation of sphingosine kinases 1 and 2 and secretion of sphingosine 1-phosphate requires Fyn kinase and contributes to mast cell responses.** *J Biol Chem* 2006, **281**:2515-2525.
 73. Pushparaj PN, Manikandan J, Tay HK, H'Ng SC, Kumar SD, Pfeilschifter J, Huwiler A, Melendez AJ: **Sphingosine kinase 1 is pivotal for Fc epsilon RI-mediated mast cell signaling and functional responses *in vitro* and *in vivo*.** *J Immunol* 2009, **183**:221-227.
 74. Price MM, Oskeritzian CA, Falanga YT, Harikumar KB, Allegood JC, Alvarez SE, Conrad D, Ryan JJ, Milstien S, Spiegel S: **A specific sphingosine kinase 1 inhibitor attenuates airway hyperresponsiveness and inflammation in a mast cell-dependent murine model of allergic asthma.** *J Allergy Clin Immunol* 2013, **131**:501-511.e501.
- This article demonstrates that a specific sphingosine kinase 1 inhibitor suppresses human and mouse mast cell activation *in vitro* and reduces inflammation in a murine model of mast cell-dependent asthma
75. Nakae S, Lunderius C, Ho LH, Schafer B, Tsai M, Galli SJ: **TNF can contribute to multiple features of ovalbumin-induced allergic inflammation of the airways in mice.** *J Allergy Clin Immunol* 2007, **119**:680-686.
 76. Williams CM, Galli SJ: **Mast cells can amplify airway reactivity and features of chronic inflammation in an asthma model in mice.** *J Exp Med* 2000, **192**:455-462.
 77. Gilfillan AM, Rivera J: **The tyrosine kinase network regulating mast cell activation.** *Immunol Rev* 2009, **228**:149-169.
 78. Huang E, Nocka K, Beier DR, Chu TY, Buck J, Lahm HW, Wellner D, Leder P, Besmer P: **The hematopoietic growth factor KL is encoded by the Sl locus and is the ligand of the c-kit receptor, the gene product of the W locus.** *Cell* 1990, **63**:225-233.
 79. Zsebo KM, Williams DA, Geissler EN, Broudy VC, Martin FH, Atkins HL, Hsu RY, Birkett NC, Okino KH, Murdock DC et al.: **Stem cell factor is encoded at the Sl locus of the mouse and is the ligand for the c-kit tyrosine kinase receptor.** *Cell* 1990, **63**:213-224.
 80. Zsebo KM, Wypych J, McNiece IK, Lu HS, Smith KA, Karkare SB, Sachdev RK, Yuschenko VN, Birkett NC, Williams LR et al.: **Identification, purification, and biological characterization of hematopoietic stem cell factor from buffalo rat liver — conditioned medium.** *Cell* 1990, **63**:195-201.
 81. Anderson DM, Lyman SD, Baird A, Wignall JM, Eisenman J, Rauch C, March CJ, Boswell HS, Gimpel SD, Cosman D et al.: **Molecular cloning of mast cell growth factor, a hematopoietin that is active in both membrane bound and soluble forms.** *Cell* 1990, **63**:235-243.
 82. Garcia-Montero AC, Jara-Acevedo M, Teodosio C, Sanchez ML, Nunez R, Prados A, Aldanondo I, Sanchez L, Dominguez M, Botana LM et al.: **KIT mutation in mast cells and other bone marrow hematopoietic cell lineages in systemic mast cell disorders: a prospective study of the Spanish Network on Mastocytosis (REMA) in a series of 113 patients.** *Blood* 2006, **108**:2366-2372.
 83. Nagata H, Worobec AS, Oh CK, Chowdhury BA, Tannenbaum S, Suzuki Y, Metcalfe DD: **Identification of a point mutation in the catalytic domain of the protooncogene c-kit in peripheral blood mononuclear cells of patients who have mastocytosis with an associated hematologic disorder.** *Proc Natl Acad Sci U S A* 1995, **92**:10560-10564.
 84. Ma Y, Zeng S, Metcalfe DD, Akin C, Dimitrijevic S, Butterfield JH, McMahon G, Longley BJ: **The c-KIT mutation causing human mastocytosis is resistant to STI571 and other KIT kinase inhibitors; kinases with enzymatic site mutations show different inhibitor sensitivity profiles than wild-type kinases and those with regulatory-type mutations.** *Blood* 2002, **99**:1741-1744.
 85. Ustun C, DeRemer DL, Akin C: **Tyrosine kinase inhibitors in the treatment of systemic mastocytosis.** *Leuk Res* 2011, **35**:1143-1152.
 86. Bai Y, Bandara G, Ching Chan E, Maric I, Simakova O, Bandara SN, Lu WP, Wise SC, Flynn DL, Metcalfe DD et al.:

Targeting the KIT activating switch control pocket: a novel mechanism to inhibit neoplastic mast cell proliferation and mast cell activation. *Leukemia* 2013, **27**:278-285.

The authors propose a novel approach to inhibit KIT tyrosine kinase activity. Interestingly, they have designed compounds that inhibit the activity of both wild-type and mutated (D816V) KIT.

87. Gleixner KV, Peter B, Blatt K, Suppan V, Reiter A, Radia D, Hadzijušević E, Valent P: **Synergistic growth-inhibitory effects of ponatinib and midostaurin (PKC412) on neoplastic mast cells carrying KIT D816V.** *Haematologica* 2013, **98**:1450-1457.
88. El-Agamy DS: **Anti-allergic effects of nilotinib on mast cell-mediated anaphylaxis like reactions.** *Eur J Pharmacol* 2012, **680**:115-121.
89. Yamaki K, Yoshino S: **Tyrosine kinase inhibitor sunitinib relieves systemic and oral antigen-induced anaphylaxes in mice.** *Allergy* 2012, **67**:114-122.
90. Jensen BM, Beaven MA, Iwaki S, Metcalfe DD, Gilfillan AM: **Concurrent inhibition of kit- and FcεRI-mediated signaling: coordinated suppression of mast cell activation.** *J Pharmacol Exp Ther* 2008, **324**:128-138.
91. Havard S, Scola AM, Kay LJ, Ishmael SS, MacGlashan DW Jr, Peachell PT: **Characterization of syk expression in human lung mast cells: relationship with function.** *Clin Exp Allergy* 2011, **41**:378-388.
92. Kepley CL, Youssef L, Andrews RP, Wilson BS, Oliver JM: **Syk deficiency in nonreleaser basophils.** *J Allergy Clin Immunol* 1999, **104**:279-284.
93. Lavens-Phillips SE, MacGlashan DW Jr: **The tyrosine kinases p53/56lyn and p72syk are differentially expressed at the protein level but not at the messenger RNA level in nonreleasing human basophils.** *Am J Respir Cell Mol Biol* 2000, **23**:566-571.
94. MacGlashan DW Jr: **Relationship between spleen tyrosine kinase and phosphatidylinositol 5' phosphatase expression and secretion from human basophils in the general population.** *J Allergy Clin Immunol* 2007, **119**:626-633.
95. Masuda ES, Schmitz J: **Syk inhibitors as treatment for allergic rhinitis.** *Pulm Pharmacol Ther* 2008, **21**:461-467.
96. Meltzer EO, Berkowitz RB, Grossbard EB: **An intranasal Syk-kinase inhibitor (R112) improves the symptoms of seasonal allergic rhinitis in a park environment.** *J Allergy Clin Immunol* 2005, **115**:791-796.
97. Matsubara S, Koya T, Takeda K, Joetham A, Miyahara N, Pine P, Masuda ES, Swasey CH, Gelfand EW: **Syk activation in dendritic cells is essential for airway hyperresponsiveness and inflammation.** *Am J Respir Cell Mol Biol* 2006, **34**:426-433.
98. Matsubara S, Li G, Takeda K, Loader JE, Pine P, Masuda ES, Miyahara N, Miyahara S, Lucas JJ, Dakhama A *et al.*: **Inhibition of spleen tyrosine kinase prevents mast cell activation and airway hyperresponsiveness.** *Am J Respir Crit Care Med* 2006, **173**:56-63.
99. Penton PC, Wang X, Amatullah H, Cooper J, Godri K, North ML, Khanna N, Scott JA, Chow CW: **Spleen tyrosine kinase inhibition attenuates airway hyperresponsiveness and pollution-induced enhanced airway response in a chronic mouse model of asthma.** *J Allergy Clin Immunol* 2013, **131**:510-511 512-520.
100. Stenton GR, Kim MK, Nohara O, Chen CF, Hirji N, Wills FL, Gilchrist M, Hwang PH, Park JG, Finlay W *et al.*: **Aerosolized Syk antisense suppresses Syk expression, mediator release from macrophages, and pulmonary inflammation.** *J Immunol* 2000, **164**:3790-3797.
101. Stenton GR, Ulanova M, Dery RE, Merani S, Kim MK, Gilchrist M, Puttagunta L, Musat-Marcu S, James D, Schreiber AD *et al.*: **Inhibition of allergic inflammation in the airways using aerosolized antisense to Syk kinase.** *J Immunol* 2002, **169**:1028-1036.
102. Weinblatt ME, Kavanaugh A, Genovese MC, Musser TK, Grossbard EB, Magilavy DB: **An oral spleen tyrosine kinase (Syk) inhibitor for rheumatoid arthritis.** *N Engl J Med* 2010, **363**:1303-1312.
103. Friedberg JW, Sharman J, Sweetenham J, Johnston PB, Vose JM, Lacasce A, Schaefer-Cuttillo J, De Vos S, Sinha R, Leonard JP *et al.*: **Inhibition of Syk with fostamatinib disodium has significant clinical activity in non-Hodgkin lymphoma and chronic lymphocytic leukemia.** *Blood* 2010, **115**:2578-2585.
104. Mocsai A, Ruland J, Tybulewicz VL: **The SYK tyrosine kinase: a crucial player in diverse biological functions.** *Nat Rev Immunol* 2010, **10**:387-402.
105. Lau C, Castellanos P, Ranev D, Wang X, Chow CW: **HRV signaling in airway epithelial cells is regulated by ITAM-mediated recruitment and activation of Syk.** *Protein Pept Lett* 2011, **18**:518-529.
106. Lau C, Wang X, Song L, North M, Wiehler S, Proud D, Chow CW: **Syk associates with clathrin and mediates phosphatidylinositol 3-kinase activation during human rhinovirus internalization.** *J Immunol* 2008, **180**:870-880.
107. Wang X, Lau C, Wiehler S, Pow A, Mazzulli T, Gutierrez C, Proud D, Chow CW: **Syk is downstream of intercellular adhesion molecule-1 and mediates human rhinovirus activation of p38 MAPK in airway epithelial cells.** *J Immunol* 2006, **177**:6859-6870.
108. Hait WN, Harding MW, Handschumacher RE: **Calmodulin, cyclophilin, and cyclosporin A.** *Science* 1986, **233**:987-989.
109. Standaert RF, Galat A, Verdine GL, Schreiber SL: **Molecular cloning and overexpression of the human FK506-binding protein FKBP.** *Nature* 1990, **346**:671-674.
110. O'Keefe SJ, Tamura J, Kincaid RL, Tocci MJ, O'Neill EA: **FK-506- and CsA-sensitive activation of the interleukin-2 promoter by calcineurin.** *Nature* 1992, **357**:692-694.
111. Liu J, Farmer JD Jr, Lane WS, Friedman J, Weissman I, Schreiber SL: **Calcineurin is a common target of cyclophilin-cyclosporin A and FKBP-FK506 complexes.** *Cell* 1991, **66**:807-815.
112. Cirillo R, Triggiani M, Siri L, Ciccarelli A, Pettit GR, Condorelli M, Marone G: **Cyclosporin A rapidly inhibits mediator release from human basophils presumably by interacting with cyclophilin.** *J Immunol* 1990, **144**:3891-3897.
113. de Paulis A, Cirillo R, Ciccarelli A, Condorelli M, Marone G: **FK-506 a potent novel inhibitor of the release of proinflammatory mediators from human Fc epsilon RI+ cells.** *J Immunol* 1991, **146**:2374-2381.
114. de Paulis A, Cirillo R, Ciccarelli A, de Crescenzo G, Oriente A, Marone G: **Characterization of the anti-inflammatory effect of FK-506 on human mast cells.** *J Immunol* 1991, **147**:4278-4285.
115. de Paulis A, Stellato C, Cirillo R, Ciccarelli A, Oriente A, Marone G: **Anti-inflammatory effect of FK-506 on human skin mast cells.** *J Invest Dermatol* 1992, **99**:723-728.
116. Stellato C, de Paulis A, Ciccarelli A, Cirillo R, Patella V, Casolaro V, Marone G: **Anti-inflammatory effect of cyclosporin A on human skin mast cells.** *J Invest Dermatol* 1992, **98**:800-804.
117. Williams HC: **Clinical practice. Atopic dermatitis.** *N Engl J Med* 2005, **352**:2314-2324.
118. Marone G: **Tacrolimus ointment for atopic dermatitis.** *N Engl J Med* 1998, **339**:1788-1789 1788; author reply.
119. Ando T, Matsumoto K, Namiranian S, Yamashita H, Glatthorn H, Kimura M, Dolan BR, Lee JJ, Galli SJ, Kawakami Y *et al.*: **Mast cells are required for full expression of allergen/SEB-induced skin inflammation.** *J Invest Dermatol* 2013, **133**:2695-2705.

The authors identify an essential role for mast cells and T cells in the development of a model of allergen-induced skin inflammation.

120. Ando T, Xiao W, Gao P, Namiranian S, Matsumoto K, Tomimori Y, Hong H, Yamashita H, Kimura M, Kashiwakura J-I *et al.*: **Critical role for mast cell Stat5 activity in skin inflammation.** *Cell Rep* 2014, **6**:366-376.

The authors describe a model of TSLP-driven skin inflammation in which STAT5 activation in mast cells play a critical role.

121. Nakamura Y, Oscherwitz J, Cease KB, Chan SM, Munoz-Planillo R, Hasegawa M, Villaruz AE, Cheung GY, McGavin MJ, Travers JB *et al.*: **Staphylococcus delta-toxin induces allergic**

- skin disease by activating mast cells.** *Nature* 2013, **503**:397-401.
- The authors propose an important pathogenetic link between *S. aureus* skin colonization and atopic dermatitis via *S. aureus* delta-toxin-dependent mast cell activation.
122. Toshiaki K, Tomoaki A, Miho K, Bridget SW, Yuko K: **Mast cells in atopic dermatitis.** *Curr Opin Immunol* 2009, **21**:666-678.
 123. Ma Z, Tovar JP, Kwong KY, Paek D: **Pimecrolimus induces apoptosis of mast cells in a murine model of cutaneous mastocytosis.** *Int Arch Allergy Immunol* 2010, **153**:413-418.
 124. Foster KG, Fingar DC: **Mammalian target of rapamycin (mTOR): conducting the cellular signaling symphony.** *J Biol Chem* 2010, **285**:14071-14077.
 125. Kim MS, Kuehn HS, Metcalfe DD, Gilfillan AM: **Activation and function of the mTORC1 pathway in mast cells.** *J Immunol* 2008, **180**:4586-4595.
 126. Smrz D, Kim MS, Zhang S, Mock BA, Smrzova S, DuBois W, Simakova O, Maric I, Wilson TM, Metcalfe DD et al.: **mTORC1 and mTORC2 differentially regulate homeostasis of neoplastic and non-neoplastic human mast cells.** *Blood* 2011, **118**:6803-6813.
 127. Simons FE, Simons KJ: **Histamine and H1-antihistamines: celebrating a century of progress.** *J Allergy Clin Immunol* 2011, **128**:1139-1150.e1134.
 128. Lichtenstein LM, Gillespie E: **The effects of the H1 and H2 antihistamines on 'allergic' histamine release and its inhibition by histamine.** *J Pharmacol Exp Ther* 1975, **192**:441-450.
 129. Marone G, Granata F, Spadaro G, Onorati AM, Triggiani M: **Antiinflammatory effects of oxatomide.** *J Invest Allergol Clin Immunol* 1999, **9**:207-214.
 130. Patella V, de Crescenzo G, Marino O, Spadaro G, Genovese A, Marone G: **Oxatomide inhibits the release of proinflammatory mediators from human basophils and mast cells.** *Int Arch Allergy Immunol* 1996, **111**:23-29.
 131. Alevizos M, Karagkouni A, Vasiadi M, Sismanopoulos N, Makris M, Kalogeromitros D, Theoharides TC: **Rupatadine inhibits inflammatory mediator release from human laboratory of allergic diseases 2 cultured mast cells stimulated by platelet-activating factor.** *Ann Allergy Asthma Immunol* 2013, **111**:542-547.
 132. Vasiadi M, Kalogeromitros D, Kempuraj D, Clemons A, Zhang B, Chliwa C, Makris M, Wolfberg A, House M, Theoharides TC: **Rupatadine inhibits inflammatory mediator secretion from human mast cells triggered by different stimuli.** *Int Arch Allergy Immunol* 2010, **151**:38-45.
 133. Genovese A, Patella V, De Crescenzo G, De Paulis A, Spadaro G, Marone G: **Loratadine and desethoxycarbonyl-loratadine inhibit the immunological release of mediators from human Fc epsilon RI+ cells.** *Clin Exp Allergy* 1997, **27**:559-567.
 134. Weller K, Maurer M: **Desloratadine inhibits human skin mast cell activation and histamine release.** *J Invest Dermatol* 2009, **129**:2723-2726.
 135. Nielsen PN, Skov PS, Poulsen LK, Schmelz M, Petersen LJ: **Cetirizine inhibits skin reactions but not mediator release in immediate and developing late-phase allergic cutaneous reactions. A double-blind, placebo-controlled study.** *Clin Exp Allergy* 2001, **31**:1378-1384.
 136. Okayama Y, Benyon RC, Lowman MA, Church MK: **In vitro effects of H1-antihistamines on histamine and PGD2 release from mast cells of human lung, tonsil, and skin.** *Allergy* 1994, **49**:246-253.
 137. Triggiani M, Giannattasio G, Balestrieri B, Granata F, Gelb MH, de Paulis A, Marone G: **Differential modulation of mediator release from human basophils and mast cells by mizolastine.** *Clin Exp Allergy* 2004, **34**:241-249.
 138. O'Mahony L, Akdis M, Akdis CA: **Regulation of the immune response and inflammation by histamine and histamine receptors.** *J Allergy Clin Immunol* 2011, **128**:1153-1162.
 139. Hofstra CL, Desai PJ, Thurmond RL, Fung-Leung WP: **Histamine H4 receptor mediates chemotaxis and calcium mobilization of mast cells.** *J Pharmacol Exp Ther* 2003, **305**:1212-1221.
 140. Mirzahasosini A, Dalmadi B, Csutora P: **Histamine receptor H4 regulates mast cell degranulation and IgE induced Fc epsilon RI upregulation in murine bone marrow-derived mast cells.** *Cell Immunol* 2013, **283**:38-44.
- This article demonstrates that antagonism of H4R negatively modulates murine mast cell degranulation *in vitro* only if present during sensitization with IgE.
141. MacGlashan DW Jr: **IgE-dependent signaling as a therapeutic target for allergies.** *Trends Pharmacol Sci* 2012, **33**:502-509.
 142. Mochly-Rosen D, Das K, Grimes KV: **Protein kinase C, an elusive therapeutic target?** *Nat Rev Drug Discov* 2012, **11**:937-957.
 143. Abdel-Raheem IT, Hide I, Yanase Y, Shigemoto-Mogami Y, Sakai N, Shirai Y, Saito N, Hamada FM, El-Mahdy NA, Elsisy Ael D et al.: **Protein kinase C-alpha mediates TNF release process in RBL-2H3 mast cells.** *Br J Pharmacol* 2005, **145**:415-423.
 144. Chang EY, Szallasi Z, Acs P, Raizada V, Wolfe PC, Fewtrell C, Blumberg PM, Rivera J: **Functional effects of overexpression of protein kinase C-alpha, -beta, -delta, -epsilon, and -eta in the mast cell line RBL-2H3.** *J Immunol* 1997, **159**:2624-2632.
 145. Li G, Lucas JJ, Gelfand EW: **Protein kinase C alpha, beta, and betaII isozymes regulate cytokine production in mast cells through MEKK2/ERK5-dependent and -independent pathways.** *Cell Immunol* 2005, **238**:10-18.
 146. Liu Y, Graham C, Parravicini V, Brown MJ, Rivera J, Shaw S: **Protein kinase C theta is expressed in mast cells and is functionally involved in Fc epsilon receptor I signaling.** *J Leukoc Biol* 2001, **69**:831-840.
 147. Nechushtan H, Leitges M, Cohen C, Kay G, Razin E: **Inhibition of degranulation and interleukin-6 production in mast cells derived from mice deficient in protein kinase Cbeta.** *Blood* 2000, **95**:1752-1757.
 148. Ozawa K, Szallasi Z, Kazanietz MG, Blumberg PM, Mischak H, Mushinski JF, Beaven MA: **Ca(2+)-dependent and Ca(2+)-independent isozymes of protein kinase C mediate exocytosis in antigen-stimulated rat basophilic RBL-2H3 cells. Reconstitution of secretory responses with Ca2+ and purified isozymes in washed permeabilized cells.** *J Biol Chem* 1993, **268**:1749-1756.
 149. Sagi-Eisenberg R, Lieman H, Pecht I: **Protein kinase C regulation of the receptor-coupled calcium signal in histamine-secreting rat basophilic leukaemia cells.** *Nature* 1985, **313**:59-60.
 150. Leitges M, Gimborn K, Elis W, Kalesnikoff J, Hughes MR, Krystal G, Huber M: **Protein kinase C-delta is a negative regulator of antigen-induced mast cell degranulation.** *Mol Cell Biol* 2002, **22**:3970-3980.
 151. Ozawa K, Yamada K, Kazanietz MG, Blumberg PM, Beaven MA: **Different isozymes of protein kinase C mediate feedback inhibition of phospholipase C and stimulatory signals for exocytosis in rat RBL-2H3 cells.** *J Biol Chem* 1993, **268**:2280-2283.
 152. Patella V, Casolaro V, Ciccirelli A, Pettit GR, Columbo M, Marone G: **The antineoplastic bryostatins affect human basophils and mast cells differently.** *Blood* 1995, **85**:1272-1281.
 153. Prevete N, Staiano RI, Granata F, Detoraki A, Necchi V, Ricci V, Triggiani M, De Paulis A, Marone G, Genovese A: **Expression and function of angiopoietins and their tie receptors in human basophils and mast cells.** *J Biol Regul Homeost Agents* 2013, **27**:827-839.
 154. Schleimer RP, Gillespie E, Lichtenstein LM: **Release of histamine from human leukocytes stimulated with the tumor-promoting phorbol diesters. I. Characterization of the response.** *J Immunol* 1981, **126**:570-574.
 155. Massey WA, Cohan VL, MacGlashan DW Jr, Gittlen SW, Kagey-Sobotka A, Lichtenstein LM, Warner JA: **Protein kinase C modulates immunoglobulin E-mediated activation of human mast cells from lung and skin. I. Pharmacologic inhibition.** *J Pharmacol Exp Ther* 1991, **258**:824-829.

156. Wu-Zhang AX, Newton AC: **Protein kinase C pharmacology: refining the toolbox.** *Biochem J* 2013, **452**:195-209.
 157. Ashmole I, Bradding P: **Ion channels regulating mast cell biology.** *Clin Exp Allergy* 2013, **43**:491-502.
 158. Cruse G, Duffy SM, Brightling CE, Bradding P: **Functional KCa3.1 K⁺ channels are required for human lung mast cell migration.** *Thorax* 2006, **61**:880-885.
 159. Ashmole I, Duffy SM, Leyland ML, Morrison VS, Begg M, Bradding P: **CRACM/Orai ion channel expression and function in human lung mast cells.** *J Allergy Clin Immunol* 2012, **129**:1628-1635.e1622.
- This article suggests that CRACM/Orai Ca²⁺ channels mediates Ca²⁺ influx in HLMC after IgE- mediated activation. It also shows that CRACM/Orai channel blockers inhibit HLMC activation *in vitro* and allergen-dependent bronchial smooth muscle contraction *ex vivo*
160. Feske S: **Calcium signalling in lymphocyte activation and disease.** *Nat Rev Immunol* 2007, **7**:690-702.
 161. Humbert M, Beasley R, Ayres J, Slavin R, Hebert J, Bousquet J, Beeh KM, Ramos S, Canonica GW, Hedgecock S *et al.*: **Benefits of omalizumab as add-on therapy in patients with severe persistent asthma who are inadequately controlled despite best available therapy (GINA 2002 step 4 treatment): INNOVATE.** *Allergy* 2005, **60**:309-316.
 162. Beck LA, Marcotte GV, MacGlashan D, Togias A, Saini S: **Omalizumab-induced reductions in mast cell Fcεpsilon RI expression and function.** *J Allergy Clin Immunol* 2004, **114**:527-530.
 163. MacGlashan DW Jr, Bochner BS, Adelman DC, Jardieu PM, Togias A, McKenzie-White J, Sterbinsky SA, Hamilton RG, Lichtenstein LM: **Down-regulation of Fc(epsilon)RI expression on human basophils during *in vivo* treatment of atopic patients with anti-IgE antibody.** *J Immunol* 1997, **158**:1438-1445.
 164. Prussin C, Griffith DT, Boesel KM, Lin H, Foster B, Casale TB: **Omalizumab treatment downregulates dendritic cell FcεpsilonRI expression.** *J Allergy Clin Immunol* 2003, **112**:1147-1154.
 165. Kaplan AP, Joseph K, Maykut RJ, Geba GP, Zeldin RK: **Treatment of chronic autoimmune urticaria with omalizumab.** *J Allergy Clin Immunol* 2008, **122**:569-573.
 166. Maurer M, Rosen K, Hsieh HJ, Saini S, Grattan C, Gimenez-Arnau A, Agarwal S, Doyle R, Canvin J, Kaplan A *et al.*: **Omalizumab for the treatment of chronic idiopathic or spontaneous urticaria.** *N Engl J Med* 2013, **368**:924-935.
- This is an important phase 3, multicenter, randomized, double-blind study showing that omalizumab treatment reduces clinical symptoms and signs of chronic idiopathic urticaria in patients resistant to H1-antihistamine treatment.
167. Corren J, Shapiro G, Reimann J, Deniz Y, Wong D, Adelman D, Togias A: **Allergen skin tests and free IgE levels during reduction and cessation of omalizumab therapy.** *J Allergy Clin Immunol* 2008, **121**:506-511.
 168. McDonnell JM, Beavil AJ, Mackay GA, Jameson BA, Korngold R, Gould HJ, Sutton BJ: **Structure based design and characterization of peptides that inhibit IgE binding to its high-affinity receptor.** *Nat Struct Biol* 1996, **3**:419-426.
 169. Rossi M, Ruvo M, Marasco D, Colombo M, Cassani G, Verdoliva A: **Anti-allergic properties of a new all-D synthetic immunoglobulin-binding peptide.** *Mol Immunol* 2008, **45**:226-234.
 170. Sandomenico A, Monti SM, Marasco D, Dathan N, Palumbo R, Saviano M, Ruvo M: **IgE-binding properties and selectivity of peptide mimics of the FcεvarepsilonRI binding site.** *Mol Immunol* 2009, **46**:3300-3309.
 171. Wiegand TW, Williams PB, Dreskin SC, Jouvin MH, Kinet JP, Tasset D: **High-affinity oligonucleotide ligands to human IgE inhibit binding to Fc epsilon receptor I.** *J Immunol* 1996, **157**:221-230.
 172. Baumann MJ, Eggel A, Amstutz P, Stadler BM, Vogel M: **DARPinS against a functional IgE epitope.** *Immunol Lett* 2010, **133**:78-84.
 173. Eggel A, Baumann MJ, Amstutz P, Stadler BM, Vogel M: **DARPinS as bispecific receptor antagonists analyzed for immunoglobulin E receptor blockade.** *J Mol Biol* 2009, **393**:598-607.
 174. Kim B, Eggel A, Tarchevskaya SS, Vogel M, Prinz H, Jardetzky TS: **Accelerated disassembly of IgE-receptor complexes by a disruptive macromolecular inhibitor.** *Nature* 2012, **491**:613-617.
- This article demonstrates that a macromolecular inhibitor of the DARPIn family not only blocks IgE-FcεRI interaction, but also dissociates previously assembled IgE-FcεRI complexes.
175. Daeron M, Jaeger S, Du Pasquier L, Vivier E: **Immunoreceptor tyrosine-based inhibition motifs: a quest in the past and future.** *Immunol Rev* 2008, **224**:11-43.
 176. Karra L, Levi-Schaffer F: **Down-regulation of mast cell responses through ITIM containing inhibitory receptors.** *Adv Exp Med Biol* 2011, **716**:143-159.
 177. Zhu D, Kepley CL, Zhang M, Zhang K, Saxon A: **A novel human immunoglobulin Fc gamma Fc epsilon bifunctional fusion protein inhibits Fc epsilon RI-mediated degranulation.** *Nat Med* 2002, **8**:518-521.
 178. Clark GJ, Ju X, Tate C, Hart DN: **The CD300 family of molecules are evolutionarily significant regulators of leukocyte functions.** *Trends Immunol* 2009, **30**:209-217.
 179. Kumagai H, Oki T, Tamitsu K, Feng SZ, Ono M, Nakajima H, Bao YC, Kawakami Y, Nagayoshi K, Copeland NG *et al.*: **Identification and characterization of a new pair of immunoglobulin-like receptors LMIR1 and 2 derived from murine bone marrow-derived mast cells.** *Biochem Biophys Res Commun* 2003, **307**:719-729.
 180. Bachelet I, Munitz A, Moretta A, Moretta L, Levi-Schaffer F: **The inhibitory receptor IRp60 (CD300a) is expressed and functional on human mast cells.** *J Immunol* 2005, **175**:7989-7995.
 181. Bachelet I, Munitz A, Berent-Maoz B, Mankuta D, Levi-Schaffer F: **Suppression of normal and malignant kit signaling by a bispecific antibody linking kit with CD300a.** *J Immunol* 2008, **180**:6064-6069.
 182. Bachelet I, Munitz A, Levi-Schaffer F: **Abrogation of allergic reactions by a bispecific antibody fragment linking IgE to CD300a.** *J Allergy Clin Immunol* 2006, **117**:1314-1320.
 183. Simhadri VR, Andersen JF, Calvo E, Choi SC, Coligan JE, Borrego F: **Human CD300a binds to phosphatidylethanolamine and phosphatidylserine, and modulates the phagocytosis of dead cells.** *Blood* 2012, **119**:2799-2809.
- The authors show that phosphatidylethanolamine and phosphatidylserine are the ligands for the inhibitory receptor LMIR1/CD300a.
184. Takahashi M, Izawa K, Kashiwakura J, Yamanishi Y, Enomoto Y, Kaitani A, Maehara A, Isobe M, Ito S, Matsukawa T *et al.*: **Human CD300C delivers an Fc receptor-gamma-dependent activating signal in mast cells and monocytes and differs from CD300A in ligand recognition.** *J Biol Chem* 2013, **288**:7662-7675.
 185. Nakahashi-Oda C, Tahara-Hanaoka S, Shoji M, Okoshi Y, Nakano-Yokomizo T, Ohkohchi N, Yasui T, Kikutani H, Honda S, Shibuya K *et al.*: **Apoptotic cells suppress mast cell inflammatory responses via the CD300a immunoreceptor.** *J Exp Med* 2012, **209**:1493-1503.
- This article demonstrates that LMIR1/CD300a binds phosphatidylserine and its activation reduces mast cell mediator release in response to microbial stimuli.
186. Izawa K, Isobe M, Matsukawa T, Ito S, Maehara A, Takahashi M, Yamanishi Y, Kaitani A, Oki T, Okumura K *et al.*: **Sphingomyelin and ceramide are physiological ligands for human LMIR3/CD300f, inhibiting FcεpsilonRI-mediated mast cell activation.** *J Allergy Clin Immunol* 2014, **133**:270-273 e271-277.
- The authors show that, unlike murine LMIR3/CD300f, human LMIR3/CD300f binds both ceramide and sphingomyelin, thereby reducing IgE-dependent mast cell activation
187. Izawa K, Yamanishi Y, Maehara A, Takahashi M, Isobe M, Ito S, Kaitani A, Matsukawa T, Matsukawa T, Nakahara F *et al.*: **The receptor LMIR3 negatively regulates mast cell activation and allergic responses by binding to extracellular ceramide.** *Immunity* 2012, **37**:827-839.
- The authors show that murine LMIR3/CD300f binds to ceramide and negatively modulates mast cell activation *in vitro* and *in vivo*.

188. Kiwamoto T, Kawasaki N, Paulson JC, Bochner BS: **Siglec-8 as a drugable target to treat eosinophil and mast cell-associated conditions.** *Pharmacol Ther* 2012, **135**:327-336.
189. Yokoi H, Choi OH, Hubbard W, Lee HS, Canning BJ, Lee HH, Ryu SD, von Gunten S, Bickel CA, Hudson SA *et al.*: **Inhibition of FcepsilonRI-dependent mediator release and calcium flux from human mast cells by sialic acid-binding immunoglobulin-like lectin 8 engagement.** *J Allergy Clin Immunol* 2008, **121**:499-505.e491.
190. Kataoka TR, Kumanogoh A, Bandara G, Metcalfe DD, Gilfillan AM: **CD72 negatively regulates KIT-mediated responses in human mast cells.** *J Immunol* 2010, **184**:2468-2475.
191. Wu HJ, Bondada S: **CD72 a coreceptor with both positive and negative effects on B lymphocyte development and function.** *J Clin Immunol* 2009, **29**:12-21.
192. Hitomi K, Tahara-Hanaoka S, Someya S, Fujiki A, Tada H, Sugiyama T, Shibayama S, Shibuya K, Shibuya A: **An immunoglobulin-like receptor Allergin-1, inhibits immunoglobulin E-mediated immediate hypersensitivity reactions.** *Nat Immunol* 2010, **11**:601-607.
193. Nagai K, Tahara-Hanaoka S, Morishima Y, Tokunaga T, Imoto Y, Noguchi E, Kanemaru K, Imai M, Shibayama S, Hizawa N *et al.*: **Expression and function of Allergin-1 on human primary mast cells.** *PLOS ONE* 2013, **8**:e76160.
194. Andreas Gt, Alina S, Undine L, Loreen K, Metin A, Beate MH: **LAMP-1 and LAMP-2, but not LAMP-3, are reliable markers for activation-induced secretion of human mast cells.** *Cytometry* 2004, **61**:62-68.
195. Abramson J, Xu R, Pecht I: **An unusual inhibitory receptor — the mast cell function-associated antigen (MAFA).** *Mol Immunol* 2002, **38**:1307-1313.
196. Abramson J, Licht A, Pecht I: **Selective inhibition of the Fc epsilon RI-induced de novo synthesis of mediators by an inhibitory receptor.** *EMBO J* 2006, **25**:323-334.