

Themed Issue: Histamine Pharmacology Update

REVIEW

Twenty-first century mast cell stabilizers

D F Finn and J J Walsh

School of Pharmacy and Pharmaceutical Sciences, Trinity College Dublin, Dublin, Ireland

Correspondence

Dr John J Walsh, School of Pharmacy and Pharmaceutical Sciences, Trinity College Dublin, Dublin 2, Ireland. E-mail: jjwalsh@tcd.ie

Keywords

mast cell; sensitization; anti-IgE; mediator release; allergy; inhibitors; disodium cromoglycate; natural products

Received

3 December 2012

Revised

30 January 2013

Accepted

13 February 2013

Mast cell stabilizing drugs inhibit the release of allergic mediators from mast cells and are used clinically to prevent allergic reactions to common allergens. Despite the relative success of the most commonly prescribed mast cell stabilizer, disodium cromoglycate, in use for the preventative treatment of bronchial asthma, allergic conjunctivitis and vernal keratoconjunctivitis, there still remains an urgent need to design new substances that are less expensive and require less frequent dosing schedules. In this regard, recent developments towards the discovery of the next generation of mast cell stabilizing drugs has included studies on substances isolated from natural sources, biological, newly synthesized compounds and drugs licensed for other indications. The diversity of natural products evaluated range from simple phenols, alkaloids, terpenes to simple amino acids. While in some cases their precise mode of action remains unknown it has nevertheless sparked interest in the development of synthetic derivatives with improved pharmacological properties. Within the purely synthetic class of inhibitors, particular attention has been devoted to the inhibition of important signalling molecules including spleen TK and JAK3. The statin class of cholesterol-lowering drugs as well as nilotinib, a TK inhibitor, are just some examples of clinically used drugs that have been evaluated for their anti-allergic properties. Here, we examine each approach under investigation, summarize the test data generated and offer suggestions for further preclinical evaluation before their therapeutic potential can be realized.

LINKED ARTICLES

This article is part of a themed issue on Histamine Pharmacology Update. To view the other articles in this issue visit <http://dx.doi.org/10.1111/bph.2013.170.issue-1>

Abbreviations

AXE, *Amomum xanthiodes*; BEL, bromoenol lactone; BMDCs, bone marrow-derived cultured murine mast cells; CerK, ceramide kinase; CHMCs, cultured human mast cells; CMTs, chemically modified tetracyclines; DNP, dinitrophenyl; hCBMCs, human umbilical cord blood-derived cultured mast cells; iPLA₂β, calcium-independent phospholipase A₂; KL, *c-kit* ligand; PTL, parthenolide; RBL, rat basophilic leukaemia; SQTs, sesquiterpene lactones; Syk, spleen TK; TGA, C₇₀-tetraglycolic acid; TLCK, tosyl-L-lysine chloromethyl ketone; TPCK, *N-p*-tosyl-L-phenylalanine chloromethyl ketone

Introduction

Mast cells play a fundamental role in the occurrence of allergic diseases because of their hypersensitive response to otherwise innocuous substances that induces an allergic reaction. The allergic reaction begins with the interaction of allergen with polyvalent IgE-FcεRI complexes expressed on the surface of sensitized mast cells that causes receptor aggregation. A complex signalling cascade follows involving the activation of numerous signalling proteins such as spleen TK (Syk) and Lyn kinase, which in turn cause a series of downstream signal transduction events within the mast cell. Ultimately, this signal transduction process leads to calcium influx and release of preformed chemical mediators such as

histamine from mast cells as well as the synthesis of lipid mediators such as PGs and LTs and the production of cytokines and chemokines. The actions of these mediators on their receptors and surrounding tissue as well as their recruitment of other immune cells are responsible for the early and late effects of an IgE-mediated allergic reaction. Mast cells are therefore central players in both the development and maintenance of allergic diseases and are subsequently considered an attractive therapeutic target in the treatment of allergic diseases such as asthma, allergic rhinitis and allergic conjunctivitis.

Mast cells play a prominent role in the immunopathology of the immediate-hypersensitivity reaction, which occurs in response to contact with certain allergens. Human

mast cell (HMC) precursors originate from cluster of differentiation molecules CD34⁺, CD13⁺ and *c-kit*⁺ (Kirshenbaum *et al.*, 1999) pluripotent haematopoietic stem cells in the bone marrow (Kitamura *et al.*, 1978). These precursors circulate in the blood before they migrate to either connective or mucosal tissues where they mature to form connective or mucosal mast cells (Okayama and Kawakami, 2006). Mature mast cells are widely distributed throughout the body where they reside in vascularized tissue, in close proximity to blood vessels, nerves, smooth muscle cells, mucus-producing glands and hair follicles (Galli and Tsai, 2008). They are also resident in anatomical sites that are directly exposed to the environment including the skin, airways and gastrointestinal tract (Galli *et al.*, 2008).

Mature mast cells are classified into two subtypes depending on their location; connective tissue mast cells (CTMCs), which reside in tissues such as the skin, small bowel submucosa and peritoneal cavity, or mucosal mast cells (MMC_s), which mature in mucosal tissues such as the intestinal lamina propria and in the airways. CTMCs and MMC_s can be differentiated based on the protease content of their granules; MMC_s contain tryptase alone and are also known as MC_T while CTMCs contain tryptase and chymase and are also known as MC_{TC} (McEuen and Walls, 2008). However, mast cells even in the same tissue have been shown to respond differently to different stimuli due to their heterogeneity (Moon *et al.*, 2010).

Mast cell heterogeneity is a crucial point to consider when designing novel mast cell stabilizers. As typified by the gold standard mast cell stabilizer, disodium cromoglycate (DSCG), which has demonstrated effectiveness in both *in vitro* and *in vivo* rat models, it is largely ineffective in mouse models. A comparative view of these species shows that DSCG successfully inhibited IgE-dependent mast cell activation in rats *in vivo* (10 mg·kg⁻¹) and *in vitro* (10–100 μM) using peritoneal mast cells whereas mouse mast cells failed to inhibit mediator release under these conditions (Oka *et al.*, 2012). In another study, DSCG up to a concentration of 10 mM failed to inhibit histamine release from mucosal mast cells when stimulated by compound 48/80 and bee venom, but effectively inhibited histamine release from peritoneal mast cells under the same conditions (Pearce *et al.*, 1982). The influence of mast cell heterogeneity in the prevention of mediator release by DSCG and ketotifen is evident from the different responses observed between mast cell populations from lung, tonsillar and skin tissue. Mast cells from lung and tonsillar tissue are inhibited by ketotifen and to a lesser extent by DSCG when challenged with an IgE-dependent histamine release mechanism. However, both agents failed to inhibit mediator release from skin mast cells (Okayama and Church, 1992), while the mast cell stabilizer nedocromil sodium showed more effective inhibition of histamine release from mast cells isolated from the lung, tonsillar and adenoidal tissues at a high concentration (1 mM) than DSCG. However, in contrast, the effect of nedocromil sodium was weaker than DSCG in intestinal mast cells. Skin mast cells differ from mast cells of the other anatomical sites in being unresponsive to both mast cell stabilizers emphasizing the existence of difference in mast cells types (Okayama *et al.*, 1992).

Mast cells are key effector cells in the occurrence and maintenance of an allergic reaction. Sensitized mast cells respond to the exposure of a foreign substance by orchestrating a complex downstream signalling cascade within the mast cell resulting in the release a variety of chemical mediators. The effect of these mediators on surrounding cells and tissues are what cause the symptoms and severity of an allergic reaction.

An allergic reaction may be prevented or attenuated by interfering with certain signalling molecules within the signalling cascade of the mast cell. A primary target of intracellular signalling upon mast cell activation is PLCγ1, which is recruited to the membrane where it is tyrosine phosphorylated by Syk and Bruton's TK. PLCγ1 catalyses the breakdown of membrane phospholipid PIP2 to generate the second messengers inositol-1,4,5-triphosphate (IP3) and DAG (Suh *et al.*, 2008). These signalling molecules are responsible for the activation of PKC isoforms and the release of calcium (Ca²⁺) from intracellular stores, respectively (Cho *et al.*, 2004). This results in a transient rise in intracellular-free Ca²⁺, which triggers the entry of calcium from the extracellular environment. Degranulation and release of chemical mediators from the mast cell follows resulting in the onset of the allergic response. Agents that prevent mediator release from these cells are termed mast cell stabilizers with examples discovered over the last decade from natural, semi-synthetic and synthetic sources.

Sources of mast cell stabilizers

Nature has provided us with the basis of many medicines in clinical use today (Table 1). Indeed, the inspiration behind DSCG arises from the earlier data generated on Khellin, a plant-derived mast cell stabilizer from *Amni visnaga*. In the first section of this review, emphasis will be placed on recent studies with natural products with particular emphasis placed on flavonoids/phenolics, terpenoids, alkaloids and biologics. The discussion will then focus on work conducted with synthetic mast cell stabilizers whose structure was inspired from studies on plant-derived materials. The final section of the review will focus on recent developments on rationally designed specific inhibitors as well as studies with proprietary medications for other indications that also have been shown to stabilize mast cells (Figure 1).

Mast cell stabilizing agents from natural sources

Flavonoids. Flavonoids can be subdivided into many different classes including flavones, flavonols, flavonones, isoflavones and flavonol-3-ols and anthocyanidins. Regardless of the individual subdivisions, all contain the benzo-γ-pyrone architecture and are classified according to the presence of different substituents on the rings and to the degree of saturation of the benzo-γ-pyrone ring. Within the flavone class, the most active mast cell stabilizers are luteolin, disomitin and apigenin. Using anti-IgE to elicit degranulation, luteolin inhibited the release of histamine, LTs, PG₂ and GM-CSF from human cultured mast cells (HCMCs) in a concentration-dependent manner (1–100 μM) (Kimata *et al.*,

Table 1

Naturally occurring mast cell stabilizers

Source of MC stabilizers	Compound name	Mast cell population	Elicitor	In vivo evaluation	Reference	
Flavonoids	Luteolin	HCMCs BMMCs Basophils RBL-2H3	anti-IgE anti-IgE and IL-3 Antigen	Unspecified	Kimata <i>et al.</i> , 2000b Kimata <i>et al.</i> , 2000a Hirano <i>et al.</i> , 2006 Mastuda <i>et al.</i> , 2002	
	Diosmetin	RBL-2H3	Antigen	Unspecified	Mastuda <i>et al.</i> , 2002	
	Apigenin	Basophils	Anti-IgE and IL-3	Unspecified	Hirano <i>et al.</i> , 2006 Kimata <i>et al.</i> , 2000a	
	Quercetin	RBL-2H3 cells	Anti-IgE, phorbol-12-myristate-13-acetate, calcium ionophore A23187 and PMACI	Unspecified	Park <i>et al.</i> , 2008 Park <i>et al.</i> , 2007 Lee <i>et al.</i> , 2010 Kempuraj <i>et al.</i> , 2006	
	Fisetin	RBL-2H3 HMC-1	anti-IgE, PMACI PMACI	Unspecified	Park <i>et al.</i> , 2008 Park <i>et al.</i> , 2007	
	Kaempferol	RBL-2H3 cells	anti-IgE, phorbol-12-myristate-13-acetate, calcium ionophore A23187 and PMACI	Unspecified	Park <i>et al.</i> , 2008 Park <i>et al.</i> , 2007 Lee <i>et al.</i> , 2010	
	Ginkgetin	BMMCs	KL	Unspecified	Son <i>et al.</i> , 2005	
	EGCG	RPMCs RBL-2H3 cells	Compound 48/80	PCA reaction in rats	Li <i>et al.</i> , 2005 Inoue <i>et al.</i> , 2010	
	Coumarins	Silymarin	RPMCs	Compound 48/80	PCA reaction in mice	Choi and Yan, 2009c
		Scopletin	HMC-1	PMACI		Moon <i>et al.</i> , 2007
Scaporonone		RPMC	anti-DNP IgE	PCA reaction in rats	Choi and Yan, 2009a	
Artekeiskeanol A		RBL-2H3 cells	Calcium ionophore A23187	Unspecified	Hong <i>et al.</i> , 2009	
Selinidin		BMMC	Antigen	Unspecified	Kishiro <i>et al.</i> , 2008	
5-Methoxy-8-(2-hydroxy-3-butoxy-3-methylbutyloxy)-psoralen		BMMC	KL	Unspecified	Hua <i>et al.</i> , 2008	
Cinnamic acid		RBL-2H3 cells	Antigen	Unspecified	Ninomiya <i>et al.</i> , 2010	
Phenols	Ellagic acid	RPMCs	anti-DNP IgE	PCA reaction in rats	Choi and Yan, 2009b	
	Magnolol and honokiol	RBL-2H3 cells	IgE-antigen	PCA reaction in rats	Han <i>et al.</i> , 2007	
	Resveratrol	HMC-1 cells	PMACI		Kang <i>et al.</i> , 2009	
	Polydatin	RBL-2H3 cells	Anti-IgE	PCA reaction in mice	El-Agamy, 2012	
	Curcumin	RBL-2H3 cells and BMMCs	Antigen	PCA reaction in mice	Lee <i>et al.</i> , 2008	
	Mangostin- α , - β and - γ	RBL-2H3	Antigen		Itoh <i>et al.</i> , 2008	
	Parthenolide	RBL-2H3 cells and BMMCs	Antigen-IgE	PCA reaction in mice	Miyata <i>et al.</i> , 2008	
Terpenoids	Sesquiterpene lactones (SQLTs)	RBL-2H3	Antigen	PCA reaction in mice	Itoh <i>et al.</i> , 2009	
	Monoterpenes	RPMCs	compound 48/80	PCA reaction in mice	Kim <i>et al.</i> , 2007	
	Sinomenine	RBL-2H3	Antigen	Unspecified	Huang <i>et al.</i> , 2008	
	Indoline	BMMCs RPMCs	Antigen compound 48/80	Unspecified	Kiefer <i>et al.</i> , 2010 Ruster <i>et al.</i> , 2004	
	Xestospongic C	RBL-2H3	Antigen and thapsigargin	Unspecified	Oka <i>et al.</i> , 2002	
	Amino acids	Theanine	RMPCs and HMC-1 cells	Compound 48/80	PCA reaction in mice	Kim <i>et al.</i> , 2011

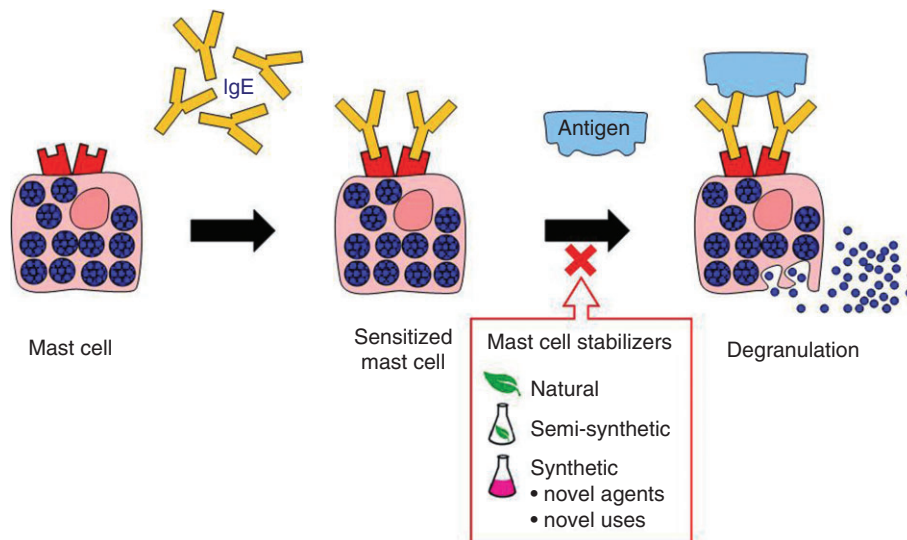


Figure 1

Sources of mast cell stabilizers. Significant data have been published on evaluating the mast cell stabilizing properties of natural products and their semi-synthetic derivatives, despite the fact that in many cases, their precise mechanism of action remains unknown. The data published on purely synthetic mast cell stabilizers have been more focussed on targeting specific events involved in the allergic cascade.

2000b). Luteolin also suppressed the production of proinflammatory cytokines TNF- α and IL-6 in bone marrow-derived cultured murine mast cells (BMDCs) (Kimata *et al.*, 2000a). Luteolin and apigenin were strong inhibitors of production of IL-4 by purified basophils following combined challenge with anti-IgE and IL-3 (Hirano *et al.*, 2006). Both luteolin and diosmetin showed potent inhibitory effects on the release of β -hexosaminidase from antigen-stimulated rat basophilic leukaemia (RBL)-2H3 cells with IC₅₀ values of 3.0 μ M and 2.1 μ M, respectively. Additionally, these flavones inhibited anti-IgE-mediated production of TNF- α and IL-4 from this cell line (Mastuda *et al.*, 2002). The structure of the flavonols differs from the flavones by the presence of an additional hydroxy-substituent at position 3. Examples of flavonols, which have demonstrated anti-allergic activity include kaempferol, fisetin, quercetin and morin. Kaempferol, fisetin and quercetin inhibited anti-IgE, phorbol-12-myristate-13-acetate, calcium ionophore A23187 and phorbol 12-myristate 13-acetate and calcium ionophore A23187 (PMACI)-induced histamine release in RBL-2H3 cells as well as the elevation of intracellular Ca²⁺ when evaluated at a relatively high concentration of 30 μ M. Additionally, fisetin and quercetin decreased gene expression and production of proinflammatory cytokines such as TNF- α , IL-1 β , IL-6 and IL-8 in PMACI-stimulated HMC-1 cells (Park *et al.*, 2008). Fisetin was also shown to decrease gene expression of IL-4, inhibit the phosphorylation of p38 MAPK, ERK and JNK and suppress the activation of NF- κ B (Park *et al.*, 2007). Quercetin and kaempferol inhibited the secretion of mediators at concentrations of 1 and 10 μ M from RBL-2H3 cells stimulated by anti-dinitrophenyl (DNP) and suppressed the mRNA expression of CD23 and p38 MAPK activation in Caco-2 cells stimulated by IL-4 (Lee *et al.*, 2010). These flavonols also significantly inhibited the release of histamine and cytokines; TNF- α , IL-6 and IL-8 from human umbilical cord blood-

derived cultured mast cells (hCBMCs) activated by anti-IgE and decreased the elevation of intracellular Ca²⁺ in this cell line (Kempuraj *et al.*, 2005). Quercetin has been shown to down-regulate the mRNA transcription of histidine decarboxylase in HMC-1 cells, an enzyme involved in the synthesis of histamine (Kempuraj *et al.*, 2006). Morin prevented the degranulation and the production of cytokines such as TNF- α and IL-4 in both RBL-2H3 cells and BMDCs stimulated by antigen at low concentrations (1–10 μ M). Morin demonstrated inhibition of activating phosphorylation of Syk. *In vivo*, this flavonol suppressed IgE-mediated passive cutaneous anaphylaxis (PCA) in mice almost completely at a dose of 100 mg·kg⁻¹ (Kim *et al.*, 2009). The isoflavone, genistein inhibited degranulation of HMCs challenged with anti-IgE in a dose-dependent manner with inhibition of histamine release by 92% at concentration of 100 μ g·mL⁻¹. Additionally, at this concentration, it inhibited phosphorylation of cellular proteins such as ERK-1 and ERK-2, which are involved in the downstream signalling cascade of activated mast cells (Suzuki *et al.*, 1997). Similarly, genistein also inhibited histamine release and protein TK activation in BMDCs stimulated with antigen (Kawakami *et al.*, 1992). The biflavone, ginkgetin, isolated from the leaves of *Ginkgo biloba*, demonstrated a dual COX-2/5-lipoxygenase inhibitory activity and was subsequently shown to inhibit the production of the *de novo* mediators, PGD₂ and LTC₄ in BMDCs stimulated with *c-kit* ligand (KL). Additionally, ginkgetin inhibited release of β -hexosaminidase from these cells stimulated with KL in a dose-dependent manner with IC₅₀ value of 6.52 μ M (Son *et al.*, 2005).

Epigallocatechin gallate (EGCG), a constituent found in green tea is related to the flavonoid family, but differs from the core flavone and flavonol structure by the absence of the double bond and carbonyl group at positions 2–3 and 4, respectively. EGCG demonstrated anti-allergic activity in

both *in vitro* and in *in vivo* models. It significantly inhibited the release of compound 48/80-induced degranulation of histamine in rat peritoneal mast cells (RPMCs). EGCG also suppressed compound 48/80-induced PCA reaction in rats (Li *et al.*, 2005). EGCG also inhibited antigen-induced degranulation and LTC₄ secretion in RBL-2H3 cells across a concentration range of 10–100 μM and has been shown to block store-operated Ca²⁺ entry of this cell line, which is the main route of calcium influx in mast cells that leads to the degranulation of allergic mediators (Inoue *et al.*, 2010).

Silymarin is a mixture of polyphenolic flavonoids isolated from milk thistle (*Silybum marianum*) and is used primarily for its treatment of liver diseases such as hepatitis, cirrhosis and jaundice. One of its primary constituents, silibinin was shown to inhibit the release of histamine from RPMCs stimulated by compound 48/80 and anti-DNP as well as the secretion of proinflammatory cytokines such as TNF-α and IL-6 from anti-DNP induced degranulation of RPMCs in a dose-dependent manner (10–100 μM). *In vivo*, silibinin inhibited compound 48/80-induced PCA reaction, in mice dose-dependently (10–100 mg·kg⁻¹) (Choi and Yan, 2009c).

Coumarins. Several reports exist that describe the mast cell stabilizing properties of coumarins. Scopletin (6-methoxy-7-hydroxycoumarin), which has been isolated from several plant species including *Erycibe obtusifolia* Benth inhibited the production of proinflammatory cytokines including TNF-α, IL-6 and IL-8 from the HMC-1 following challenge with PMACI. These cytokines play a role in triggering and sustaining allergic inflammation. However, scopletin did not affect the release of histamine induced by agents from HMC-1 cells (Moon *et al.*, 2007). Interestingly, scaplorone, the methylated analogues of scopletin dose-dependently decreased histamine release from rat peritoneal mast cells stimulated by anti-DNP IgE at concentrations ranging from 25 to 100 μM. It also inhibited PCA reaction in rats dose-dependently at concentrations of 10, 25 and 50 mg·kg⁻¹. Scaplorone also reduced the expression and secretion of proinflammatory cytokines such as TNF-α and IL-6 (Choi and Yan, 2009a).

The coumarin, artekeiskeanol A, isolated from *Artemisia keskeana* Miq. suppressed degranulation of RBL-2H3 cells induced by antigen and calcium ionophore A23187 in a concentration-dependent manner (10–100 μM). Artekeiskeanol A also suppressed the mRNA levels of proinflammatory cytokines TNF-α and IL-13 and phosphorylation of signalling kinases such as p38 MAPK and JNK, which are involved in downstream signalling events (Hong *et al.*, 2009). Selinidin, a coumarin derived from *Angelica keiskei* attenuated the release of β-hexosaminidase from bone marrow-derived mast cells (BMMCs) stimulated by antigen and the production of proinflammatory mediators such as LT C₄ and TNF-α. Selinidin also decreased phosphorylation of PLCγ-1 and p38 MAPK, enzymes involved in the signalling pathway of degranulation (Kishiro *et al.*, 2008). The furanocoumarin, 5-methoxy-8-(2-hydroxy-3-butoxy-3-methylbutyloxy)-psoralen, isolated from *Angelica dahurica* inhibited both COX-2 and 5-lipoxygenase activity and generation of the lipid mediators PGD₂ and LTC₄. This furanocoumarin also prevented degranulation of mice BMMCs activated with KL (Hua *et al.*, 2008). Interestingly, cinnamic acid, which might be considered as a precursor

to the coumarin structure, markedly suppressed antigen-stimulated degranulation of β-hexosaminidase from RBL-2H3 cells in a dose-dependent manner (10–100 μM) through inactivation of Syk and PLCγ pathways (Ninomiya *et al.*, 2010). Thunbergins A and B are isocoumarin derivatives isolated from the processed leaves of *Hydrangeae macrophylla* var. *thunbergii*, which have shown anti-allergic activity by inhibiting histamine release from RPMCs stimulated by calcium ionophore A23187 and antigen (Matsuda *et al.*, 1999). Thunberginol B demonstrated potent activity by completely inhibiting degranulation against both elicitors at a concentration of 30 μM. They both also inhibited degranulation of RBL-2H3 cells stimulated by calcium ionophore and antigen as well as the release of cytokines TNF-α and IL-4 (Wang *et al.*, 2007). Thunberginol B was also shown to inhibit mRNA expression of several cytokines including IL-2, IL-3, IL-4 and IL-13, TNF-α and granulocyte/macrophage colony-stimulating factor (GM-CSF) in RBL-2H3 cells stimulated by antigen (Matsuda *et al.*, 2008).

Ellagic acid (2,3,7,8-tetrahydroxy[1]benzopyrano [5,4,3-cde][1]benzopyran-5,10-dione) is a polyphenolic compound found in fruits and nuts such as raspberries, strawberries, walnuts, longan seeds, mango kernel and pomegranate, which has shown to attenuate anti-IgE-mediated allergic response *in vitro* and *in vivo*. Ellagic acid dose-dependently inhibited histamine release as well as the secretion of proinflammatory cytokines such as TNF-α and IL-6 from anti-DNP IgE induced degranulation of RPMCs across a concentration range of 50–200 μM. Ellagic acid attenuated anti-DNP IgE-mediated PCA in rats (Choi and Yan, 2009b).

Phenols. Magnolol and honokiol are phenolic structural isomers, isolated from the bark of *Magnolia obovata* that have shown to potently inhibit the degranulation of RBL-2H3 cells induced by IgE-antigen complex as well as the production of cytokines; IL-4 and TNF-α. Moreover, both compounds potently inhibited PCA reactions in mice induced by IgE-antigen complex dose-dependently at doses of 10 and 50 mg·kg⁻¹ (Han *et al.*, 2007). Resveratrol, is a phytoalexin, stilbene polyphenolic compound found in grapes, berries and peanuts. It suppressed the expression of inflammatory cytokines such as TNF-α, IL-6 and IL-8 in PMACI-induced-HMC-1 cells and decreased the levels of intracellular Ca²⁺ (Kang *et al.*, 2009). Likewise, the anti-allergic activity of polydatin, a resveratrol glucoside, significantly decreased FcεRI-mediated degranulation in IgE-sensitized RBL-2H3 cells in a dose-dependent manner (1–100 μM) and inhibited the IgE-dependent PCA reaction in mice (300 mg·kg⁻¹; El-Agamy, 2012). Curcumin is a polyphenolic compound found in *Curcuma longa* and related species. Curcumin has demonstrated anti-allergic activity in both *in vitro* and *in vivo* models. It significantly inhibited antigen-induced degranulation in a dose-dependent manner (1–10 μM) in both RBL-2H3 cells and BMMCs and moreover suppressed PCA reaction in mice at doses of 0.5–50 mg·kg⁻¹. Curcumin significantly inhibited the expression of mRNA for cytokines; IL-4 and TNF-α in a dose-dependent manner as well as their secretion in antigen-stimulated RBL-2H3 cells (Lee *et al.*, 2008). The xanthones; mangostin-α, -β and -γ isolated from the pericarp of *Garcinia mangostana* L. inhibited the release of histamine from IgE-sensitized RBL-2H3 cells in response to antigen

through suppression of the signalling transduction pathway involving Syk and PLC γ (Itoh *et al.*, 2008).

Within the flavonoid, coumarin and phenolic classes, it is evident from studies conducted to date that the precise mechanism by which these substances stabilize mast cells remains largely unknown. As with most planar molecules, as in these series, it is perhaps conceivable that many processes in the allergic cascade are targeted. This point is reinforced by the broad spectrum of biological actions exhibited by these classes of compounds and from the evidence presented above where they show effects against many different cell populations. The hypothesis that truly selective inhibitors of a given molecular target can only be generated from three-dimensional molecules is perhaps reinforced by findings within these studies. Nevertheless, as the allergic cascade may involve several inflammatory pathways, the idea of designing anti-allergic substances with multiple mechanisms of actions may have potential advantageous therapeutically.

Terpenoids. Some recent examples of mast cell stabilizers from the terpenoid class include parthenolide (PTL), a sesquiterpene lactone isolated from the herb feverfew (*Tanacetum parthenium*), extracts from which exhibits anti-inflammatory properties and are used for the treatment of migraine (Murphy *et al.*, 1988). PTL has also shown anti-allergic properties both *in vitro* and *in vivo* models. PTL inhibited antigen-IgE induced degranulation of both RBL-2H3 cells and BMMCs at low concentrations (0.6–5 μM) and strongly inhibited PCA reaction in mice by approximately 90% at a concentration of 10 mg·kg⁻¹. PTL was also shown to strongly suppress IgE-antigen-induced cytoskeletal rearrangement in RBL-2H3 cells, which is considered a critical step for the degranulation process in mast cells (Miyata *et al.*, 2008). Dehydroleucodine, a sesquiterpene lactone isolated from *Artemisia douglasiana* Besser and xanthatin, a xanthanolate lactone isolated from *Xanthium cavanillesii* Schouw inhibited the release of the mediator serotonin from RPMCs induced by compound 48/80. Both substances showed more potent inhibitory activity than the established mast cell stabilizers, disodium cromoglycate and ketotifen (Penissi *et al.*, 2009). Nine types of sesquiterpene lactones (SQLTs) isolated from *Eupatorium chinense* L. suppressed the degranulation from antigen-stimulated RBL-2H3 cells and furthermore were shown to suppress the elevation of intracellular Ca²⁺. *In vivo*, the sesquiterpene lactones-rich extract potently inhibited PCA reaction induced by antigen-IgE complex in mice in a dose-dependent manner (Itoh *et al.*, 2009). Monoterpenes including; borneol and camphene, terpene alcohol; linalool and sesquiterpene; nerolidol in the form of an extract of *Amomum xanthiodes* (AXE) showed good anti-allergic activity both in both *in vitro* and *in vivo* screens. AXE reduced histamine release from RPMCs stimulated by compound 48/80 in a dose-dependent manner and also reduced the level of intracellular Ca²⁺. AXE suppressed compound 48/80-induced PCA reaction in mice (Kim *et al.*, 2007).

Alkaloids. Within the alkaloid class, sinomenine (SIN) (7,8-didehydro-4-hydroxy-3,7-dimethoxy-17-methylmorphinan-6-one), an alkaloid isolated from *Sinomenium actum* inhibited antigen-induced mast cell degranulation in RBL-2H3 cells in a dose-dependent manner from 0.5 to 2 mM. Similarly, SIN

also inhibited the production of cytokines; IL-4 and TNF- α as well as the phosphorylation of various proteins involved in downstream signalling of activated mast cells such as Gab2 and p38 MAPK (Huang *et al.*, 2008). Indoline (*E,Z*)-3-(3',5'-dimethoxy-4-hydroxy-benzylidene)-2-indolinone, an alkaloid isolated from the medicinal plant *Isatis tinctoria*, demonstrated anti-allergic activity *in vitro*. Indoline-inhibited degranulation of BMMCs stimulated by antigen efficiently across a concentration range of 50–1000 nM. Indoline does not affect kinase activity directly downstream of Fc ϵ RI, but interrupts with granule exocytosis possibly by binding to proteins on the surface of granules such as soluble NSF (N-ethylmaleimide-sensitive factor) attachment protein receptors, which play an essential role in the exocytosis of mast cells (Kiefer *et al.*, 2010). Likewise, indoline inhibited compound 48/80-induced degranulation of RPMCs to a greater effect than that of the clinically used mast cell stabilizer, DSCG (Ruster *et al.*, 2004). Xestospongine C is an alkaloid isolated from the sponge *Xestospongia sp.*, which inhibited both antigen and thapsigargin induced degranulation of RBL-2H3 cells in a concentration-dependent manner (1–10 μM). It is suggested that xestospongine C exhibits its anti-allergic behaviour by crossing the mast cell membrane and blocking IP₃ receptors on the endoplasmic reticulum membrane. This action prevents Ca²⁺ store depletion and as a result inhibits the elevated levels of intracellular Ca²⁺ that is necessary for mast cell degranulation (Oka *et al.*, 2002). Theanine is the major amino acid present in green tea. Recently, the anti-allergic activity of theanine has been elucidated in both *in vitro* and *in vivo* models. The amino acid inhibited compound 48/80-induced histamine release from both RPMCs and HMC-1 cells in a dose-dependent manner and showed significant activity at low concentration (1 μM). Additionally, theanine significantly suppressed the secretion of proinflammatory cytokines such as TNF- α , IL-1 β , IL-6 and IL-8 by suppressing NF- κ B activation in PMACI-stimulated HMC-1 cells. This activity was translated to the *in vivo* setting as theanine inhibited PCA reaction in mice at 1 mg·kg⁻¹ concentration. It is suggested that it acts as a mast cell stabilizer by preventing perturbation of the lipid bilayer of mast cells (Kim *et al.*, 2011).

Biologics. Studies with biological inhibitors of mast cell degranulation have included the use of complement-derived peptide C3a. It was shown to inhibit degranulation of RBL-2H3 cells and BMMCs stimulated by antigen in a dose-dependent manner by interacting with the β -chain of Fc ϵ RI on mast cells. The binding of C3a to the mast cells caused a decrease in the proximity of IgE binding to Fc ϵ RI and as a result, suppressed the activating phosphorylation of TKs and the activity of PLC γ that are necessary for the signal transduction process involved in the degranulation of mast cells (Erdei *et al.*, 1999). A complement peptide derived from C3a, namely C3a9 inhibited the immediate phase response of antigen-stimulated-RBL-2H3 cells by causing dissociation of TKs, Lyn and Fyn with Fc ϵ RI and the inactivation of downstream MAPK, p38 and ERK. C3a9 also inhibited late phase responses of stimulated BMMCs by suppressing the secretion of proinflammatory cytokines such as IL-6 and TNF- α (Peterfy *et al.*, 2008). Likewise, several other anti-allergic peptides have been identified, namely LVA, LSY, RVS, ETI, TDG, RVV

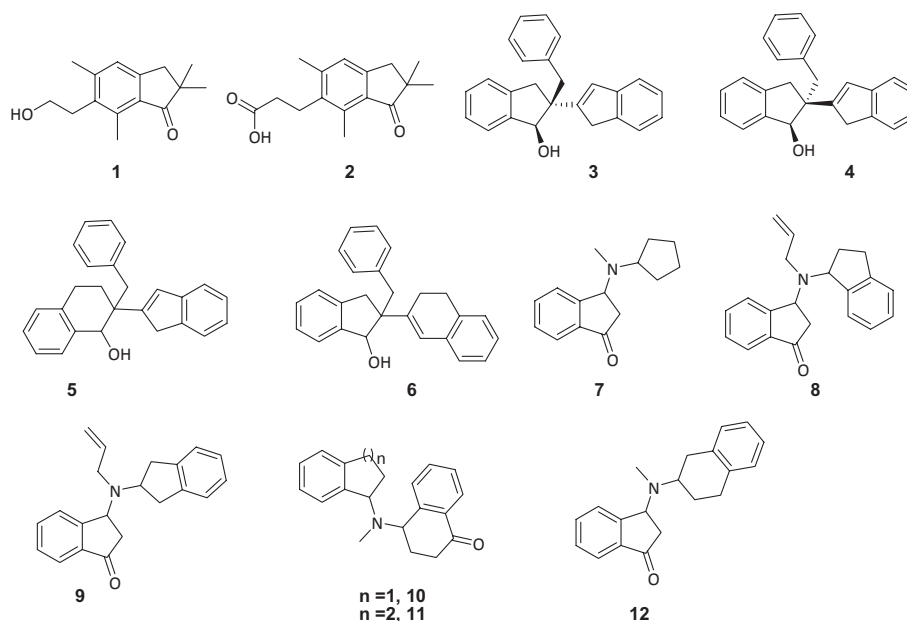


Figure 2

Benzocycloalkan-one (ol)-based mast cell stabilizers. Studies on semi-synthetic based mast cell stabilizers has primarily focussed on the preparation of 'dimer-like' compounds of the naturally occurring indanone, pterisin-Z.

and GFW, which inhibited antigen-stimulated release of β -hexosaminidase from RBL-2H3 cells. These peptides decreased the influx of Ca^{2+} and the phosphorylation of Lyn, ERK and PKC (Kim *et al.*, 2008). [Ala¹²] mast cell degranulating (MCD) peptide effectively competes with IgE in the binding affinity to Fc ϵ RI on mast cells and has consequently shown to inhibit antigen-stimulated mediator release in RBL-2H3 cells by 50% at 100 μM concentration (Buku *et al.*, 2005). Subsequently, Buku *et al.*, developed a range of modified peptide analogues of [Ala¹²] MCD, which conserved the alanine residue in position 12. Analogue [Val⁶, Ala¹²]MCD 7 is a potent inhibitor of IgE-mediated degranulation of RBL-2H3 cells causing almost complete inhibition at low concentrations (10 and 20 nM; Buku *et al.*, 2008).

Semi-synthetic inhibitors of mast cell degranulation

Natures' medicine cabinet has provided us and others with the inspiration to generate novel mast cell stabilizing compounds, stimulated by both simplicity of synthesis and observed biological effects in related systems (Figure 2). In this respect, initial studies focussed on analogues of the pterosins, which are indanone sesquiterpenes from *Pteridium aquilinum* Kuhn var. *latiusculum* (Hikino *et al.*, 1976). Of particular interest was the indanone, pterisin Z, **1**, (Farrell *et al.*, 1996), a compound that exhibited potent smooth muscle relaxant activity by inhibiting calcium-induced contraction of guinea pig ileum (Sheridan *et al.*, 1999). As calcium plays a central role in mast cell exocytosis, it was hypothesized that the pterosin indanone family of compounds may also act as inhibitors of mast cell mediator release. Earlier investigations focused on the synthesis of analogues of this compound (Frankish *et al.*, 2004). Within this series, the most active

compound was the propanoic acid analogue **2**, which demonstrated similar inhibition of compound 48/80-induced release from RPMC at 20 μM to DSCG. Encouraged by the somewhat fortuitous synthesis of and observation that dimer-type compounds containing the indane skeleton exhibited twofold superior activity to that of DSCG in mast cell assays, work commenced on building libraries of indane dimer-based compounds, which were not only investigated as mast cell stabilizers, but also as smooth muscle relaxants. Although preliminary findings indicated that no correlation existed between the structure and dual activity of these compounds, mast cell stabilizing compounds with superior activity to that of DSCG emerged from this study (Sheridan *et al.*, 2009b). In later studies, diastereomers of a benzylated indanol dimer compound as exemplified by **3** and **4**, which maintain the indanol and indenyl structural fragments were synthesized and shown to demonstrate dual mast cell stabilizing and anti-inflammatory effects in a range of *in vitro* and *in vivo* studies (Sheridan *et al.*, 2009a). While the data generated on the indane dimer series of compounds was encouraging, further studies were conducted to elucidate the exact features in these structural types that were necessary for activity. Initial studies involved replacement of one of the indane units by a tetralin skeleton in the overall molecular framework of the compounds. Replacement of either the indanol or indenyl moiety of these diastereoisomers by the corresponding tetralol or dihydronaphthalene groups to give the benzylated dimer alcohols **5** and **6** resulted in these compounds exhibiting potent mast cell stabilization activity against several elicitors including; compound 48/80 (**5**; IC₅₀ 4.1 μM , **6**; IC₅₀ 7.7 μM), concanavalin A (**5**; IC₅₀ 0.75 μM , **6**; IC₅₀ 3.6 μM) and calcium ionophore A23187 (**5**; IC₅₀ 10 μM , **6**; IC₅₀ 4.7 μM), in the RPMC assay (Barlow *et al.*, 2011a). Similarly, potent mast cell stabilizing activity was observed

against compound 48/80-induced release (IC_{50} value of $3.3 \mu\text{M}$) when the indanol core of **3** was deconstructed to give the naphthalenyl analogue **6** (Barlow *et al.*, 2011a). Earlier concurrent studies also identified a series of aminoindanones, including **7**, **8** and **9**. The cyclopentyl aminoindanone **7** and the substituted 1- and 2-N-indanyl aminoindanones **6** and **9**, respectively demonstrated substantial mast cell stabilizing activity in rodent models (Sheridan *et al.*, 2008). As a branched series of compounds, the effect of ring expansion of the core indanone skeleton(s) to tetralone on the pharmacological activity was investigated and shown in most cases to retain the activity of the indanone series *in vitro* and *in vivo* (Barlow and Walsh, 2008; 2010). Like the aminoindanone congeners as exemplified by **8**, both the allyl **9** and methyl **10**, **11** derivatives in this series demonstrated excellent mast cell stabilizing ability when evaluated in *in vitro* against compound 48/80-induced release from RPMC. Enlarging the ring further to give the benzosuberone series (7-membered B-ring) with the amino bearing a more bulky substituted benzyl group, either retained or enhanced the inhibition of mediator release from mast cells relative to **7** *in vitro* (Barlow *et al.*, 2011b), as indeed did the individual stereoisomer of **12** (Byrne *et al.*, 2011).

Synthetic inhibitors of mast cell degranulation

Syk inhibitors. Synthetic inhibitors of mast cell activation and degranulation include those that interfere with and inactivate signalling proteins and receptors required for the signal transduction of the allergic cascade (Table 2). As an example, Syk is an important mediator of immunoreceptor signalling in mast cells and other immune cells that cause inflammation. Activated Syk phosphorylates a variety of substrates including linker for activation of T cells (LAT), which orchestrates downstream signalling resulting in degranulation and cytokine gene transcription. Consequently, Syk is a potential target for the treatment of hypersensitivity reactions such as allergic rhinitis, asthma, urticaria and anaphylaxis. Studies by Mazuc *et al.* have shown that Compound 13 inhibited Fc ϵ RI-induced degranulation *in vitro* in RBL-2H3 cells and PCA reaction *in vivo*. It also impeded the interaction of Syk with other cellular signalling molecules by binding at the interface between two Src homology (SH2) domains and the inter domain A of Syk (Mazuc *et al.*, 2008). Other Syk kinase inhibitors include a series of 2, 4-diaminopyrimidines, of which the title compound, R112 demonstrated potent inhibition of Syk kinase in *in vitro* studies. R112 completely and rapidly inhibited histamine release in allergen-induced basophils in addition to lipid mediator and cytokine production of cultured HMCs (CHMCs) stimulated by allergen. The mechanism of action of R112 was confirmed to inactivate Syk, which consequently prevented the phosphorylation of LAT (Y191) and hence prevented its activation and the signalling cascade leading to degranulation (Rossi *et al.*, 2006). In clinical trials, R112 rapidly ameliorated the symptoms of allergic rhinitis in hypersensitive individuals (Masuda and Schmitz, 2008). ER-27317, an acridone-related compound, inhibited mast cell response by preventing the phosphorylation and activation of Syk kinase. *In vitro*, ER-27317 inhibited degranulation in a dose-dependent manner in RBL-2H3 cells, RPMC and

HMCs, all stimulated by antigen. Almost complete inhibition was demonstrated at a concentration of $30 \mu\text{M}$ in both rodent and human cell models. ER-27317 selectively interferes with Fc ϵ RI γ phospho-ITAM activation of Syk thus preventing the ensuing signalling cascade (Moriya *et al.*, 1997). More recently, 3-butyl-1-chloro-8-(2-methoxycarbonyl)phenyl-5H-imidazo[1,5-b]isoquinolin-10-one (U63A05) dose-dependently inhibited degranulation of RBL-2H3 cells and BMMCs stimulated by antigen across a concentration range of $1\text{--}10 \mu\text{M}$. This compound also suppressed the secretion of proinflammatory cytokines. U63A05 exerts its inhibitory effects on the activating phosphorylation of Syk, thereby preventing downstream activation of signalling molecules that lead to degranulation. *In vivo*, U63A05 suppressed antigen-stimulated PCA reaction in mice at doses ranging from 10 to $100 \text{ mg}\cdot\text{kg}^{-1}$ (Kim do *et al.*, 2011).

JAK3 inhibitors. JAK3 is a protein TK expressed in mast cells and plays a pivotal role in the Fc ϵ RI-mediated mast cell inflammatory response. JAK3 is activated by cytokines such as IL-2, IL-4, IL-7 and IL-9 upon mast cell activation. This causes phosphorylation and dimerization of STAT 5A for transcription of target genes involved in inflammation (D'Cruz and Uckun, 2007). A selective inhibitor of JAK3, 4-(4'-hydroxyphenyl)-amino-6,7-dimethoxyquinazoline (WHI-131) inhibited calcium ionophore A23187 induced- and IgE/antigen induced degranulation of RBL-2H3 cells in a concentration-dependent fashion from 1 to $30 \mu\text{M}$. Additionally, WHI-131 prevented the release of the lipid mediator LTC $_4$ and the proinflammatory cytokine TNF- α . WHI-131 also prevented PCA reaction in mice by blocking degranulation *in vivo* (Malaviya *et al.*, 1999). However, later studies indicated that degranulation of JAK3 deficient BMMCs from mice were inhibited by WHI-131 to the same extent as wild-type mice which implies that WHI-131 has other underlying mechanisms of mast cell stabilization (Linwong *et al.*, 2005).

Astaurosporine-based compound entitled compound 32 was shown to potently inhibit JAK3-type signalling in different cell types. It inhibited the JAK3 enzyme in Jurkat cells as well as the activating phosphorylation of STAT5 in T cells. Additionally, it demonstrated potent activity in mast cells by inhibiting IgE/antigen-induced hexosaminidase release (IC_{50} value of 55 nM) as well as the release of the proinflammatory cytokine TNF- α . *In vivo*, this compound reduced ovalbumin-induced IgE production by 70% ($30 \text{ mg}\cdot\text{kg}^{-1}$) in mice.

Kit TK inhibitors. The Kit ligand (stem cell factor) is essential for mast growth, differentiation, survival and enhances antigen-mediated mast cell degranulation. Therefore, inhibition of Kit is an attractive approach to prevent Fc ϵ RI-mediated allergic reactions. Hypothemycin is a resorcylic acid lactone, which blocked Kit activation, inhibited degranulation of both HMCs and BMMCs as well as cytokine production at $10 \mu\text{M}$. *In vivo*, hypothemycin reduced PCA reaction in mice ($500 \mu\text{g}\cdot 30 \text{ g}^{-1}$) (Jensen *et al.*, 2008).

Midostaurin (PKC412) is a TK inhibitor that interacts with Kit on mast cells and is used in clinical trials to counteract the growth of neoplastic mast cells in mastocytosis. Midostaurin inhibited calcium ionophore A23187-induced degranulation of both the basophil and mast cell lines, KU812 and HMC-1

Table 2
Synthetic mast cell stabilizers

Compound name	Molecular target	Mast cell population	Elicitor	In vivo evaluation	Reference
Compound 13	Syk kinase	RBL-2H3	Antigen	Unspecified	Mazuc <i>et al.</i> , 2008
R112	Syk kinase	Basophils	Allergen	Unspecified	Rossi <i>et al.</i> , 2006
ER-27317	Syk kinase	RBL-2H3 RPMC HCMC	Antigen	Unspecified	Moriya <i>et al.</i> , 1997
U63A05	Syk kinase	Unspecified	Unspecified	10–100 mg·kg ⁻¹	Kim do <i>et al.</i> , 2011
WHI-131	JAK3	RBL-2H3	Calcium ionophore	Prevent PCA reaction	Malaviya <i>et al.</i> , 1999
Hypothemycin	Kit Ligand	HuMCs BMMC	Calcium ionophore	Reduced PCA reaction in mice (500 µg·30 kg ⁻¹)	Jensen <i>et al.</i> , 2008
Midostaurin (PKC412)	Kit Ligand	Basophils KU812, HMC-1	Calcium ionophore		Krauth <i>et al.</i> , 2009
CP99994	Substance P	Unspecified	Unspecified	Unspecified	Erin <i>et al.</i> , 2004
K1	Ceramide kinase	BMMC	Calcium ionophore	Unspecified	Kumada <i>et al.</i> , 2007
Ro 20–1724, rolipram and Siguazodan	Phosphodiesterase 4	RPMC	anti-IgE		Lau and Kam, 2005
Fullerenes	Unspecified	Unspecified	Unspecified	Unspecified	Norton <i>et al.</i> , 2010
Vacuolin-1	Unspecified	BMMC	Unspecified	Unspecified	Shaik <i>et al.</i> , 2009
CMT-3	Unspecified	RPMC HuMCs	Compound 48/80	Unspecified	Sandler <i>et al.</i> , 2005
OR-1384, OR-1958	Unspecified	RPMC	Compound 48/80	Unspecified	Vendelin <i>et al.</i> , 2005
TLCK, TPCK	Unspecified	Lung mast cells	Calcium ionophore anti-IgE	Reduced PCA reaction in mice (30 mg·kg ⁻¹)	Nunomura <i>et al.</i> , 2008
Bromoenoil lactone (BEL)	iPLA ₂ β	RBL-2H3 cells BMMCs	Antigen Calcium ionophore A23187 or thapsigargin	Unspecified	Fensome-Green <i>et al.</i> , 2007
Cerivastatin, atorvastatin and fluvastatin,	Unspecified	Mature lung mast cells RBL-3H2	Anti-IgE	Unspecified	Krauth <i>et al.</i> , 2006
Nilotinib	TK	RPMCs	Compound 48/80	Inhibited allergic paw oedema in rats at concentrations of 25 and 50 mg·kg ⁻¹	Yuan <i>et al.</i> , 2012

in a dose-dependent manner at low concentrations (1–1000 nM) (Krauth *et al.*, 2009).

Mast cells are associated with stress-induced inflammatory skin disease such as psoriasis through involvement of substance P present in skin mast cells, which is an initiator of the stress response. CP99994 is a substance P NK-1 antagonist which prevented stress-induced response mast cell mediator release when Sprague-Dawley rats were treated peripherally at (1 and 2 mg·kg⁻¹) or i.c.v. (5, 10 and 20 µg). It is suggested that NK-1 antagonists may be used therapeutically to treat stress-induced inflammatory skin diseases (Erin *et al.*, 2004).

Ceramide kinase (CerK) inhibition. CerK is an enzyme involved in the phosphorylation of ceramide, a precursor of sphingolipids found in the plasma membrane of cells, which is involved in differentiation and apoptosis of the cell. CerK was found to be involved in the activation of RBL-2H3 cells (Mitsutake *et al.*, 2004). The CerK inhibitor, K1 dose-dependently (10–50 µM) suppressed degranulation of calcium ionophore A23187 induced BMMCs mediator release (Kumada *et al.*, 2007).

Phosphodiesterases (PDEs) inhibition. PDEs are a family of 11 isoenzymes that hydrolyse cyclic nucleotides such as cAMP and cGMP to form inactive metabolites 5'-AMP and 5'-GMP, respectively. These are important secondary messengers involved in many biological processes such as the activation of PKA (a cAMP-dependent Ser/Thr kinase) and PKG (a cGMP-dependent Ser/Thr kinase). Inhibition of cAMP- or cGMP-dependent PDE has the effect of raising the intracellular levels of these nucleotides (Boswell-Smith *et al.*, 2006). Inhibitors of PDEs have shown mast cell stabilizing properties (Weston *et al.*, 1997). The PDE inhibitors of PDE 4, Ro 20-1724 and rolipram, dose-dependently (1–100 µM) inhibited histamine release from anti-IgE induced RPMCs and this inhibitory activity was enhanced when each of these PDE 4 inhibitors were combined with the PDE 3 inhibitor, siguazodan. The level of inhibition of rolipram at 10 µM was 29.4 ± 7.1% which, was elevated in the presence of 1 µM siguazodan to 49.5 ± 7.1% (Lau and Kam, 2005).

Miscellaneous inhibitors. Fullerenes present a carbon sphere structure with delocalised π molecular orbital electrons, which show unusual activity in electron transfer systems. By virtue of their unique properties, fullerene derivatives have been used to treat a range of diseases including types of cancer (Mroz *et al.*, 2007) and neurodegenerative disease (Dugan *et al.*, 1997). Recently, the water-soluble fullerene, C₇₀-tetraglycolic acid (TGA) inhibited anti-IgE stimulated degranulation from human skin mast cells (10 µg·mL⁻¹; % inhibition = 39.3 ± 9.2%) and peripheral blood basophils (5 µg·mL⁻¹; % inhibition = 15.8 ± 4.2%). TGA-inhibited GM-CSF cytokine production as well as the phosphorylation of mast cell signalling proteins such as ERK 1/2, p38 MAPK, LAT and PI3K, which are involved in the release of chemical mediators. This activity was translated *in vivo* where TGA suppressed PCA reaction in mice at a concentration of 100 ng. (Norton *et al.*, 2010).

Vacuolin-1 is an inducer of large vacuole formation in various cell types. At a concentration of 10 µM, vacuolin-1 inhibited exocytosis of BMMCs stimulated with antigen, but

did not inhibit exocytosis of RBL-2H3 cells under the same conditions. The early stages of mast cell activation such as phosphorylation of LAT, MAPK and ERK were not affected in stimulated BMMCs nor was F-actin polymerization, which is necessary for translocation of secretory granules in the later stages of exocytosis (Shaik *et al.*, 2009).

Chemically modified tetracyclines (CMTs) have shown anti-inflammatory activity such as the inhibition of COX-2-mediated PGE₂ production (Patel *et al.*, 1999). Previously, CMT-3, also known as COL-3 had demonstrated promising anti-tumour activity (Lokeshwar *et al.*, 2002) CMT-3 inhibited compound 48/80 stimulated-RPMCs and HuMCs dose-dependently. In RPMCs, the level of degranulation was reduced from 71.7 ± 4.2% to 28.7 ± 3.4% in the presence of 25 µM CMT-3. Similarly, CMT-3 also inhibited the secretion of cytokines TNF-α and IL-8 from HMC-1 cells and reduced the expression of TNF-α mRNA in these cells. Additionally, CMT-3 inhibited PKC activity with an IC₅₀ value of 31 µM (Sandler *et al.*, 2005).

Orazipone (OR-1384) and its derivative OR-1958 are novel sulphhydryl reactive anti-inflammatory compounds. In *in vitro* studies, these compounds affect mast cell functions, of which OR-1958 demonstrated the most effective anti-allergic activity. OR-1958 dose-dependently inhibited compound 48/80-induced histamine release from RPMCs with an inhibition value of 56 ± 10% at 20 µM and inhibited the expression of TNF-α mRNA in HMC-1 cells. Both OR-1958 and OR-1384 dose-dependently inhibited the production of TNF-α in HMC-1 cells stimulated by PMACI with IC₅₀ values of 10 and 20 µM, respectively. It is suggested that these sulphhydryl compounds exert their effects by inactivating thiol-containing molecules involved in the signal transduction process of activated mast cells (Vendelin *et al.*, 2005).

In addition to inhibiting the release of the protease, trypsin, the protease inhibitors, *N*-α-tosyl-L-lysine chloromethyl ketone (TLCK) and *N*-*p*-tosyl-L-phenylalanine chloromethyl ketone (TPCK) have been shown to inhibit the release of histamine from both anti-IgE and calcium ionophore A23187-stimulated lung mast cells in a concentration-dependent manner. The maximum inhibition of histamine release induced by anti-IgE was approximately 40.7% with 100 µg·mL⁻¹ TLCK and 40.2% with 80 µg·mL⁻¹ TPCK (He and Xie, 2004). Later studies showed that TPCK (50 µM) almost completely suppressed degranulation of BMMCs co-stimulated with antigen and adenosine by inhibiting granule movement as well as the secretion of cytokine IL-13. *In vivo*, TPCK (30 mg·kg⁻¹) reduced PCA reaction in mice (Nunomura *et al.*, 2008).

Bromo-enol lactone (BEL) is a suicide-based irreversible inhibitor of calcium-independent PLA₂ (iPLA₂β) (Hazen *et al.*, 1991). BEL demonstrated inhibition of exocytosis of both RBL-2H3 cells and BMMCs stimulated with either antigen, calcium ionophore A23187 or thapsigargin, with maximal inhibition of exocytosis at a concentration of 25 µM of BEL. The original hypothesis was that BEL prevented the calcium influx mediated by iPLA₂ through SOCCS in these cells. However, BEL also inhibited exocytosis from permeabilized mast cells where Ca²⁺ entry mechanism was no longer relevant, which suggested that BEL interferes with events downstream of Ca²⁺ signalling, which are required for exocytosis (Fensome-Green *et al.*, 2007).

Old drugs with new uses as mast cell stabilizers. Statins are a class of hydroxymethylglutaryl-CoA (HMG-CoA) reductase inhibitors. This enzyme is essential for the biosynthesis of mevalonic acid, an essential precursor to isoprenoid compounds including cholesterol. Compounds from this series including cerivastatin, atorvastatin and fluvastatin have demonstrated anti-allergic activity. Cerivastatin and atorvastatin inhibited anti-IgE-induced histamine release from mature lung mast cells in a dose-dependent manner. The statin-induced changes in histamine release are expressed as a % of the histamine release induced by anti-IgE. The release of histamine was reduced from 100% to $40.2 \pm 14.8\%$ in the presence of 50 μM cerivastatin and reduced to $26.3 \pm 13.5\%$ in the presence of 50 μM atorvastatin. Additionally, these statins suppressed cytokine-dependent growth of normal mast cell progenitors in HMC-1 cells, which suggested that these statins are inhibitors of mast cell growth and function (Krauth *et al.*, 2006). Fluvastatin inhibited degranulation of antigen-induced RBL-3H2 cells in a concentration-dependent manner (0.5–10 μM) without affecting cytosolic calcium levels or the granule content of these cells. It is suggested that the inhibitory action of fluvastatin may be mediated by the suppression of geranylgeranyl transferase *via* the depletion of intracellular mevalonic acid. This leads to the inactivation of small GTP-binding proteins involved in microtubule formation, which are important in the translocation of the granules in a calcium-independent manner (Fujimoto *et al.*, 2009).

Nilotinib is a second-generation TK inhibitor that has been used for the treatment of BCR-ABL-positive chronic myelogenous leukemia. Recently, the anti-allergic effects of nilotinib have been reported. Administration of nilotinib prevented systematic anaphylaxis in mice mediated by compound 48/80 in a dose-dependent manner (5–50 $\text{mg}\cdot\text{kg}^{-1}$) and significantly inhibited allergic paw edema in rats at concentrations of 25 and 50 $\text{mg}\cdot\text{kg}^{-1}$. In addition, it dose-dependently (5–20 μM) reduced histamine release from RPMCs activated by either compound 48/80 or ovalbumin and attenuated the secretion of pro-inflammatory cytokines as well as TNF- α expression in the RPMCs (Yuan *et al.*, 2012).

The mucolytic agent, ambroxol was shown to inhibit histamine release by more than 50% from human adenoidal mast cells (1000 μM) stimulated by concanavalin A and from skin mast cells (100 μM) stimulated by compound 48/80. Additionally ambroxol inhibited anti-IgE-induced release of histamine, LTC₄, IL-4 and IL-13 from basophils at a concentration of 100 μM (Gibbs *et al.*, 1999).

The loop diuretic, frusemide, used in the treatment of congestive heart failure has been reported to prevent exercise-induced asthma. Frusemide dose-dependently inhibited histamine release from RPMCs within a concentration of 10^{-3} – 10^{-5} M stimulated by a various secretagogues known to increase the concentration of intracellular calcium. Frusemide was shown to protect mast cells in a similar manner to DSCG (Stenton and Lau, 1996).

Conclusions

Treatment of allergic diseases relies on clinically prescribed drug classes such as mast cell stabilizers and H1 antagonists,

which control the symptoms associated with allergic diseases. Mast cell stabilisers act by stabilizing the mast cell upon allergen exposure to inhibit the release of chemical mediators while H1 antagonists antagonise histamine at the H1 receptor to eliminate the effects mediated by this biogenic amine released during an allergic reaction. Although the first generation mast cell stabilisers such as DSCG and nedocromil sodium effectively inhibit mast cell degranulation, the second-generation mast cell stabilisers typified by olopatadine and ketotifen additionally possess anti-histaminic properties which present anti-allergic agents with dual activity.

However, the number of anti-allergic agents is not limited to these drugs in clinical use. It is evident from the numerous reports outlined in this review that a broad range of compounds have been isolated from natural sources that demonstrate substantial anti-allergic activity in a panel of *in vitro* and *in vivo* screens. In some situations, their mechanism of action has been elucidated, while in many situations their activity is not solely limited to their effect on mast cells. Indeed, in many situations they, also target many inflammatory events, which may ultimately complement their effect on mast cell degranulation. Moreover, many of these anti-allergic agents are sourced from foodstuffs such as the flavonoid family which enter the body on a daily basis where they could potentially target activated mast cells and attenuate the mast cell response to allergens. The significance of these natural products may be further emphasized by the history of the natural mast cell stabilizer, Khellin, which subsequently led to the synthesis of the first established mast cell stabilizer, DSCG. Additionally, various biological agents such as natural and synthetic peptides have shown promising anti-allergic behaviour by blocking IgE/Fc ϵ R1 binding or by interfering with the signalling pathway of the allergic response.

Also discussed here are several synthetic compounds that potentially inhibit mast cell degranulation. A comprehensive understanding of the pathway involved in the allergic cascade is invaluable towards the design of novel, selective inhibitors of mast cell activation. Inactivation of signalling proteins such as Syk and JAK3 kinases, which play important roles in the signalling cascade of an allergic reaction, are just some of the molecular targets identified in the design of novel mast cell stabilizers. The development of new anti-allergic agents within our own research group and others has identified numerous, indane-, tetralin- and benzosuberone-based compounds that have demonstrated good inhibition of mast cell degranulation, although, in many cases their precise mode of action remains unanswered.

A diverse range of mast cell stabilizing compounds have been identified in the last decade from; natural, biological and synthetic sources to drugs already in clinical uses for other indications. Although in many cases, the precise mode of action of these molecules is unclear, all of these substances have demonstrated mast cell stabilization activity and therefore may have potential therapeutic use in the treatment of allergic and related diseases where mast cells are intrinsically involved. However, owing to the heterogeneity of the mast cell, and their molecular targets, the real potential of any new mast cell stabilizer can only be realized once its properties are evaluated in an extended range of preclinical *in vitro*, *ex vivo* and *in vivo* models of efficacy and toxicity.

Acknowledgements

DF is grateful to Enterprise Ireland for financial support CFTD/2004/115. We wish to thank Sheila Ryder for her assistance in producing Figure 1.

Conflict of interest

None.

References

- Barlow JW, Walsh JJ (2008). Synthesis and evaluation of 4-amino-3,4-dihydro-2H-naphthalen-1-one derivatives as mast cell stabilising and anti-inflammatory compounds. *Eur J Med Chem* 43: 2891–2900.
- Barlow JW, Walsh JJ (2010). Synthesis and evaluation of dimeric 1,2,3,4-tetrahydro-naphthalenylamine and indan-1-ylamine derivatives with mast cell-stabilising and anti-allergic activity. *Eur J Med Chem* 45: 25–37.
- Barlow JW, McHugh AP, Woods O, Walsh JJ (2011a). Synthesis of novel mast cell-stabilising and anti-allergic 1,2,3,4-tetrahydro-1-naphthalenes and related compounds. *Eur J Med Chem* 46: 1545–1554.
- Barlow JW, Zhang T, Woods O, Byrne AJ, Walsh JJ (2011b). Novel Mast Cell-Stabilising Amine Derivatives of 3,4-Dihydronaphthalen-1(2H)-one and 6,7,8,9-Tetrahydro-5H-benzo[7]annulen-5-one. *Med Chem* 7: 213–223.
- Boswell-Smith V, Spina D, Page CP (2006). Phosphodiesterase inhibitors. *Br J Pharmacol* 147 (Suppl. 1): S252–S257.
- Buku A, Condie BA, Price JA, Mezei M (2005). [Ala12]MCD peptide: a lead peptide to inhibitors of immunoglobulin E binding to mast cell receptors. *J Pept Res* 66: 132–137.
- Buku A, Keselman I, Lupyan D, Mezei M, Price JA (2008). Effective mast cell degranulating peptide inhibitors of the IgE/Fc epsilonRI receptor interaction. *Chem Biol Drug Des* 72: 133–139.
- Byrne AJ, Barlow JW, Walsh JJ (2011). Synthesis and pharmacological evaluation of the individual stereoisomers of 3-[methyl(1,2,3,4-tetrahydro-2-naphthalenyl)amino]-1-indanone, a potent mast cell stabilising agent. *Bioorg Med Chem Lett* 21: 1191–1194.
- Cho SH, Woo CH, Yoon SB, Kim JH (2004). Protein kinase Cdelta functions downstream of Ca²⁺ mobilization in FcepsilonRI signaling to degranulation in mast cells. *J Allergy Clin Immunol* 114: 1085–1092.
- Choi YH, Yan GH (2009a). Anti-allergic effects of scoparone on mast cell-mediated allergic model. *Phytomedicine* 16: 1089–1094.
- Choi YH, Yan GH (2009b). Ellagic Acid attenuates immunoglobulin E-mediated allergic response in mast cells. *Biol Pharm Bull* 32: 1118–1121.
- Choi YH, Yan GH (2009c). Silibinin attenuates mast cell-mediated anaphylaxis-like reactions. *Biol Pharm Bull* 32: 868–875.
- D'cruz OJ, Uckun FM (2007). Targeting mast cells in endometriosis with janus kinase 3 inhibitor, JANEX-1. *Am J Reprod Immunol* 58: 75–97.
- Dugan LL, Turetsky DM, Du C, Lobner D, Wheeler M, Almlı CR *et al.* (1997). Carboxyfullerenes as neuroprotective agents. *Proc Natl Acad Sci U S A* 94: 9434–9439.
- El-Agamy DS (2012). Anti-allergic effects of nilotinib on mast cell-mediated anaphylaxis like reactions. *Eur J Pharmacol* 680: 115–121.
- Erdei A, Toth GK, Andrasfalvy M, Matko J, Bene L, Bajtaj Z *et al.* (1999). Inhibition of IgE-mediated triggering of mast cells by complement-derived peptides interacting with the Fc epsilon RI. *Immunol Lett* 68: 79–82.
- Erin N, Ersoy Y, Ercan F, Akici A, Oktay S (2004). NK-1 antagonist CP99994 inhibits stress-induced mast cell degranulation in rats. *Clin Exp Dermatol* 29: 644–648.
- Farrell R, Kelleher F, Sheridan H (1996). Synthesis of fern sesquiterpenes pterisin Z via a novel palladium-catalyzed route. *J Nat Prod* 59: 446–447.
- Fensome-Green A, Stannard N, Li M, Bolsover S, Cockcroft S (2007). Bromoenol lactone, an inhibitor of Group V1A calcium-independent phospholipase A2 inhibits antigen-stimulated mast cell exocytosis without blocking Ca²⁺ influx. *Cell Calcium* 41: 145–153.
- Frankish N, Farrell R, Sheridan H (2004). Investigation into the mast cell stabilizing activity of nature-identical and synthetic indanones. *J Pharm Pharmacol* 56: 1423–1427.
- Fujimoto M, Oka T, Murata T, Hori M, Ozaki H (2009). Fluvastatin inhibits mast cell degranulation without changing the cytoplasmic Ca²⁺ level. *Eur J Pharmacol* 602: 432–438.
- Galli SJ, Tsai M (2008). Mast cells: versatile regulators of inflammation, tissue remodeling, host defense and homeostasis. *J Dermatol Sci* 49: 7–19.
- Galli SJ, Grimaldeston M, Tsai M (2008). Immunomodulatory mast cells: negative, as well as positive, regulators of immunity. *Nat Rev Immunol* 8: 478–486.
- Gibbs BF, Schmutzler W, Vollrath IB, Brosthardt P, Braam U, Wolff HH *et al.* (1999). Ambroxol inhibits the release of histamine, leukotrienes and cytokines from human leukocytes and mast cells. *Inflamm Res* 48: 86–93.
- Han SJ, Bae EA, Trinh HT, Yang JH, Youn UJ, Bae KH *et al.* (2007). Magnolol and honokiol: inhibitors against mouse passive cutaneous anaphylaxis reaction and scratching behaviors. *Biol Pharm Bull* 30: 2201–2203.
- Hazen SL, Zupan LA, Weiss RH, Getman DP, Gross RW (1991). Suicide inhibition of canine myocardial cytosolic calcium-independent phospholipase A2. Mechanism-based discrimination between calcium-dependent and -independent phospholipases A2. *J Biol Chem* 266: 7227–7232.
- He S, Xie H (2004). Modulation of tryptase and histamine release from human lung mast cells by protease inhibitors. *Asian Pac J Allergy Immunol* 22: 205–212.
- Hikino H, Miyase T, Takemoto T (1976). Biosynthesis of pteroside B in *Pteridium aquilinum* var. *Latiusculum*, proof of the sesquiterpenoid origin of the pterosides. *Phytochemistry* 15: 121–123.
- Hirano T, Higa S, Arimitsu J, Naka T, Ogata A, Shima Y *et al.* (2006). Luteolin, a flavonoid, inhibits AP-1 activation by basophils. *Biochem Biophys Res Commun* 340: 1–7.
- Hong J, Sasaki H, Hirasawa N, Ishihara K, Kwak JH, Zee O *et al.* (2009). Suppression of the antigen-stimulated RBL-2H3 mast cell activation by Artekeiskeanol A. *Planta Med* 75: 1494–1498.

- Hua JM, Moon TC, Hong TG, Park KM, Son JK, Chang HW (2008). 5-Methoxy-8-(2-hydroxy-3-butoxy-3-methylbutyloxy)-psoralen isolated from *Angelica dahurica* inhibits cyclooxygenase-2 and 5-lipoxygenase in mouse bone marrow-derived mast cells. *Arch Pharm Res* 31: 617–621.
- Huang F, Yamaki K, Tong X, Fu L, Zhang R, Cai Y *et al.* (2008). Inhibition of the antigen-induced activation of RBL-2H3 cells by sinomenine. *Int Immunopharmacol* 8: 502–507.
- Inoue T, Suzuki Y, Ra C (2010). Epigallocatechin-3-gallate inhibits mast cell degranulation, leukotriene C4 secretion, and calcium influx via mitochondrial calcium dysfunction. *Free Radic Biol Med* 49: 632–640.
- Itoh T, Ohguchi K, Iinuma M, Nozawa Y, Akao Y (2008). Inhibitory effect of xanthenes isolated from the pericarp of *Garcinia mangostana* L. on rat basophilic leukemia RBL-2H3 cell degranulation. *Bioorg Med Chem* 16: 4500–4508.
- Itoh T, Oyama M, Takimoto N, Kato C, Nozawa Y, Akao Y *et al.* (2009). Inhibitory effects of sesquiterpene lactones isolated from *Eupatorium chinense* L. on IgE-mediated degranulation in rat basophilic leukemia RBL-2H3 cells and passive cutaneous anaphylaxis reaction in mice. *Bioorg Med Chem* 17: 3189–3197.
- Jensen BM, Beaven MA, Iwaki S, Metcalfe DD, Gilfillan AM (2008). Concurrent inhibition of kit- and FcεRI-mediated signaling: coordinated suppression of mast cell activation. *J Pharmacol Exp Ther* 324: 128–138.
- Kang OH, Jang HJ, Chae HS, Oh YC, Choi JG, Lee YS *et al.* (2009). Anti-inflammatory mechanisms of resveratrol in activated HMC-1 cells: pivotal roles of NF-κB and MAPK. *Pharmacol Res* 59: 330–337.
- Kawakami T, Inagaki N, Takei M, Fukamachi H, Coggeshall KM, Ishizaka K *et al.* (1992). Tyrosine phosphorylation is required for mast cell activation by Fc εRI cross-linking. *J Immunol* 148: 3513–3519.
- Kempuraj D, Madhappan B, Christodoulou S, Boucher W, Cao J, Papadopoulou N *et al.* (2005). Flavonols inhibit proinflammatory mediator release, intracellular calcium ion levels and protein kinase C theta phosphorylation in human mast cells. *Br J Pharmacol* 145: 934–944.
- Kempuraj D, Castellani ML, Petrarca C, Frydas S, Conti P, Theoharides TC *et al.* (2006). Inhibitory effect of quercetin on tryptase and interleukin-6 release, and histidine decarboxylase mRNA transcription by human mast cell-1 cell line. *Clin Exp Med* 6: 150–156.
- Kiefer S, Mertz AC, Koryakina A, Hamburger M, Kuenzi P (2010). (E,Z)-3-(3',5'-Dimethoxy-4'-hydroxy-benzylidene)-2-indolinone blocks mast cell degranulation. *Eur J Pharm Sci* 40: 143–147.
- Kim JW, Lee JH, Hwang BY, Mun SH, Ko NY, Kim Do K *et al.* (2009). Morin inhibits Fyn kinase in mast cells and IgE-mediated type I hypersensitivity response in vivo. *Biochem Pharmacol* 77: 1506–1512.
- Kim K, Kim Y, Kim HY, Ro JY, Jeoung D (2008). Inhibitory mechanism of anti-allergic peptides in RBL2H3 cells. *Eur J Pharmacol* 581: 191–203.
- Kim NH, Jeong HJ, Kim HM (2011). Theanine is a candidate amino acid for pharmacological stabilization of mast cells. *Amino Acids* 42: 1609–1618.
- Kim SH, Lee S, Kim IK, Kwon TK, Moon JY, Park WH *et al.* (2007). Suppression of mast cell-mediated allergic reaction by *Amomum xanthiodes*. *Food Chem Toxicol* 45: 2138–2144.
- Kim Do K, Lee JH, Kim JW, Kim HS, Kim AR, Kim BK *et al.* (2011). A novel imidazo[1,5-b]isoquinolinone derivative, U63A05, inhibits syk activation in mast cells to suppress IgE-mediated anaphylaxis in mice. *J Pharmacol Sci* 115: 500–508.
- Kimata M, Inagaki N, Nagai H (2000a). Effects of luteolin and other flavonoids on IgE-mediated allergic reactions. *Planta Med* 66: 25–29.
- Kimata M, Shichijo M, Miura T, Serizawa I, Inagaki N, Nagai H (2000b). Effects of luteolin, quercetin and baicalein on immunoglobulin E-mediated mediator release from human cultured mast cells. *Clin Exp Allergy* 30: 501–508.
- Kirshenbaum AS, Goff JP, Semere T, Foster B, Scott LM, Metcalfe DD (1999). Demonstration that human mast cells arise from a progenitor cell population that is CD34(+), c-kit(+), and expresses aminopeptidase N (CD13). *Blood* 94: 2333–2342.
- Kishiro S, Nunomura S, Nagai H, Akihisa T, Ra C (2008). Selinidin suppresses IgE-mediated mast cell activation by inhibiting multiple steps of Fc εRI signaling. *Biol Pharm Bull* 31: 442–448.
- Kitamura Y, Go S, Hatanaka K (1978). Decrease of mast cells in W/W^v mice and their increase by bone marrow transplantation. *Blood* 52: 447–452.
- Krauth MT, Majlesi Y, Sonneck K, Samorapoompichit P, Ghannadan M, Hauswirth AW *et al.* (2006). Effects of various statins on cytokine-dependent growth and IgE-dependent release of histamine in human mast cells. *Allergy* 61: 281–288.
- Krauth MT, Mirkina I, Herrmann H, Baumgartner C, Kneidinger M, Valent P (2009). Midostaurin (PKC412) inhibits immunoglobulin E-dependent activation and mediator release in human blood basophils and mast cells. *Clin Exp Allergy* 39: 1711–1720.
- Kumada H, Mitsutake S, Inagaki Y, Mitsunaga S, Tsuchikawa H, Katsumura S *et al.* (2007). Kinetics of the ceramide kinase inhibitor K1, a suppressor of mast-cell activation. *Biosci Biotechnol Biochem* 71: 2581–2584.
- Lau HY, Kam MF (2005). Inhibition of mast cell histamine release by specific phosphodiesterase inhibitors. *Inflamm Res* 54 (Suppl. 1): S05–S06.
- Lee EJ, Ji GE, Sung MK (2010). Quercetin and kaempferol suppress immunoglobulin E-mediated allergic inflammation in RBL-2H3 and Caco-2 cells. *Inflamm Res* 59: 847–854.
- Lee JH, Kim JW, Ko NY, Mun SH, Her E, Kim BK *et al.* (2008). Curcumin, a constituent of curry, suppresses IgE-mediated allergic response and mast cell activation at the level of Syk. *J Allergy Clin Immunol* 121: 1225–1231.
- Li GZ, Chai OH, Song CH (2005). Inhibitory effects of epigallocatechin gallate on compound 48/80-induced mast cell activation and passive cutaneous anaphylaxis. *Exp Mol Med* 37: 290–296.
- Linwong W, Hirasawa N, Aoyama S, Hamada H, Saito T, Ohuchi K (2005). Inhibition of the antigen-induced activation of rodent mast cells by putative Janus kinase 3 inhibitors WHI-P131 and WHI-P154 in a Janus kinase 3-independent manner. *Br J Pharmacol* 145: 818–828.
- Lokeshwar BL, Selzer MG, Zhu BQ, Block NL, Golub LM (2002). Inhibition of cell proliferation, invasion, tumor growth and metastasis by an oral non-antimicrobial tetracycline analog (COL-3) in a metastatic prostate cancer model. *Int J Cancer* 98: 297–309.
- Malaviya R, Zhu D, Dibirdik I, Uckun FM (1999). Targeting Janus kinase 3 in mast cells prevents immediate hypersensitivity reactions and anaphylaxis. *J Biol Chem* 274: 27028–27038.

- Mastuda H, Morikawa T, Ueda K, Managi H, Yoshikawa M (2002). Structural requirements of flavonoids for inhibition of antigen-Induced degranulation, TNF-alpha and IL-4 production from RBL-2H3 cells. *Bioorg Med Chem* 10: 3123–3128.
- Masuda ES, Schmitz J (2008). Syk inhibitors as treatment for allergic rhinitis. *Pulm Pharmacol Ther* 21: 461–467.
- Matsuda H, Shimoda H, Yoshikawa M (1999). Structure-requirements of isocoumarins, phthalides, and stilbenes from *Hydrangeae Dulcis Folium* for inhibitory activity on histamine release from rat peritoneal mast cells. *Bioorg Med Chem* 7: 1445–1450.
- Matsuda H, Wang Q, Matsuhira K, Nakamura S, Yuan D, Yoshikawa M (2008). Inhibitory effects of thunberginols A and B isolated from *hydrangeae dulcis folium* on mRNA expression of cytokines and on activation of activator protein-1 in RBL-2H3 cells. *Phytomedicine* 15: 177–184.
- Mazuc E, Villoutreix BO, Malbec O, Roumier T, Fleury S, Leonetti JP *et al.* (2008). A novel druglike spleen tyrosine kinase binder prevents anaphylactic shock when administered orally. *J Allergy Clin Immunol* 122: 188–194.
- Mceuen AR, Walls AF (2008). Purification and characterization of mast cell tryptase and chymase from human tissues. *Methods Mol Med* 138: 299–317.
- Mitsutake S, Kim TJ, Inagaki Y, Kato M, Yamashita T, Igarashi Y (2004). Ceramide kinase is a mediator of calcium-dependent degranulation in mast cells. *J Biol Chem* 279: 17570–17577.
- Miyata N, Gon Y, Nunomura S, Endo D, Yamashita K, Matsumoto K *et al.* (2008). Inhibitory effects of parthenolide on antigen-induced microtubule formation and degranulation in mast cells. *Int Immunopharmacol* 8: 874–880.
- Moon PD, Lee BH, Jeong HJ, An HJ, Park SJ, Kim HR *et al.* (2007). Use of scopoletin to inhibit the production of inflammatory cytokines through inhibition of the IkappaB/NF-kappaB signal cascade in the human mast cell line HMC-1. *Eur J Pharmacol* 555: 218–225.
- Moon TC, St Laurent CD, Morris KE, Marcet C, Yoshimura T, Sekar Y *et al.* (2010). Advances in mast cell biology: new understanding of heterogeneity and function. *Mucosal Immunol* 3: 111–128.
- Moriya K, Rivera J, Odom S, Sakuma Y, Muramoto K, Yoshiuchi T *et al.* (1997). ER-27319, an acridone-related compound, inhibits release of antigen-induced allergic mediators from mast cells by selective inhibition of fcepsilon receptor I-mediated activation of Syk. *Proc Natl Acad Sci U S A* 94: 12539–12544.
- Mroz P, Tegos GP, Gali H, Wharton T, Sarna T, Hamblin MR (2007). Photodynamic therapy with fullerenes. *Photochem Photobiol Sci* 6: 1139–1149.
- Murphy JJ, Heptinstall S, Mitchell JR (1988). Randomised double-blind placebo-controlled trial of feverfew in migraine prevention. *Lancet* 2: 189–192.
- Ninomiya M, Itoh T, Ishikawa S, Saiki M, Narumiya K, Yasuda M *et al.* (2010). Phenolic constituents isolated from *Fragaria ananassa* Duch. inhibit antigen-stimulated degranulation through direct inhibition of spleen tyrosine kinase activation. *Bioorg Med Chem* 18: 5932–5937.
- Norton SK, Dellinger A, Zhou Z, Lenk R, Macfarland D, Vonakis B *et al.* (2010). A new class of human mast cell and peripheral blood basophil stabilizers that differentially control allergic mediator release. *Clin Transl Sci* 3: 158–169.
- Nunomura S, Yoshimaru T, Ra C (2008). Na-Tosyl-Phe chloromethyl ketone prevents granule movement and mast cell synergistic degranulation elicited by costimulation of antigen and adenosine. *Life Sci* 83: 242–249.
- Oka T, Sato K, Hori M, Ozaki H, Karaki H (2002). Xestospongins C, a novel blocker of IP3 receptor, attenuates the increase in cytosolic calcium level and degranulation that is induced by antigen in RBL-2H3 mast cells. *Br J Pharmacol* 135: 1959–1966.
- Oka T, Kalesnikoff J, Starkl P, Tsai M, Galli SJ (2012). Evidence questioning cromolyn's effectiveness and selectivity as a 'mast cell stabilizer' in mice. *Lab Invest* 92: 1472–1482.
- Okayama Y, Church MK (1992). Comparison of the modulatory effect of ketotifen, sodium cromoglycate, procaterol and salbutamol in human skin, lung and tonsil mast cells. *Int Arch Allergy Immunol* 97: 216–225.
- Okayama Y, Kawakami T (2006). Development, migration, and survival of mast cells. *Immunol Res* 34: 97–115.
- Okayama Y, Benyon RC, Rees PH, Lowman MA, Hillier K, Church MK (1992). Inhibition profiles of sodium cromoglycate and nedocromil sodium on mediator release from mast cells of human skin, lung, tonsil, adenoid and intestine. *Clin Exp Allergy* 22: 401–409.
- Park HH, Lee S, Oh JM, Lee MS, Yoon KH, Park BH *et al.* (2007). Anti-inflammatory activity of fisetin in human mast cells (HMC-1). *Pharmacol Res* 55: 31–37.
- Park HH, Lee S, Son HY, Park SB, Kim MS, Choi EJ *et al.* (2008). Flavonoids inhibit histamine release and expression of proinflammatory cytokines in mast cells. *Arch Pharm Res* 31: 1303–1311.
- Patel RN, Attur MG, Dave MN, Patel IV, Stuchin SA, Abramson SB *et al.* (1999). A novel mechanism of action of chemically modified tetracyclines: inhibition of COX-2-mediated prostaglandin E2 production. *J Immunol* 163: 3459–3467.
- Pearce FL, Befus AD, Gaudie J, Bienenstock J (1982). Mucosal mast cells. II. Effects of anti-allergic compounds on histamine secretion by isolated intestinal mast cells. *J Immunol* 128: 2481–2486.
- Penissi AB, Vera ME, Mariani ML, Rudolph MI, Cenal JP, De Rosas JC *et al.* (2009). Novel anti-ulcer alpha,beta-unsaturated lactones inhibit compound 48/80-induced mast cell degranulation. *Eur J Pharmacol* 612: 122–130.
- Peterfy H, Toth G, Pecht I, Erdei A (2008). C3a-derived peptide binds to the type I fcepsilonR and inhibits proximal-coupling signal processes and cytokine secretion by mast cells. *Int Immunol* 20: 1239–1245.
- Rossi AB, Herlaar E, Braselmann S, Huynh S, Taylor V, Frances R *et al.* (2006). Identification of the Syk kinase inhibitor R112 by a human mast cell screen. *J Allergy Clin Immunol* 118: 749–755.
- Ruster GU, Hoffmann B, Hamburger M (2004). Inhibitory activity of indolin-2-one derivatives on compound 48/80-induced histamine release from mast cells. *Pharmazie* 59: 236–237.
- Sandler C, Ekokoski E, Lindstedt KA, Vainio PJ, Finel M, Sorsa T *et al.* (2005). Chemically modified tetracycline (CMT)-3 inhibits histamine release and cytokine production in mast cells: possible involvement of protein kinase C. *Inflamm Res* 54: 304–312.
- Shaik GM, Draberova L, Heneberg P, Draber P (2009). Vacuolin-1-modulated exocytosis and cell resealing in mast cells. *Cell Signal* 21: 1337–1345.

- Sheridan H, Frankish N, Farrell R (1999). Synthesis and antispasmodic activity of analogues of natural pterosins. *Eur J Med Chem* 34: 953–966.
- Sheridan H, Butterly S, Walsh JJ, Cogan C, Jordan M, Nolan O *et al.* (2008). Synthesis and pharmacological activity of aminoindanone dimers and related compounds. *Bioorg Med Chem* 16: 248–254.
- Sheridan H, Walsh JJ, Cogan C, Jordan M, McCabe T, Passante E *et al.* (2009a). Diastereoisomers of 2-benzyl-2,3-dihydro-2-(1H-inden-2-yl)-1H-inden-1-ol: potential anti-inflammatory agents. *Bioorg Med Chem Lett* 19: 5927–5930.
- Sheridan H, Walsh JJ, Jordan M, Cogan C, Frankish N (2009b). A series of 1, 2-coupled indane dimers with mast cell stabilisation and smooth muscle relaxation properties. *Eur J Med Chem* 44: 5018–5022.
- Son JK, Son MJ, Lee E, Moon TC, Son KH, Kim CH *et al.* (2005). Ginkgetin, a Biflavone from Ginkgo biloba leaves, inhibits cyclooxygenases-2 and 5-lipoxygenase in mouse bone marrow-derived mast cells. *Biol Pharm Bull* 28: 2181–2184.
- Stenton GR, Lau HY (1996). Inhibition of rat peritoneal mast cell exocytosis by frusemide: a study with different secretagogues. *Inflamm Res* 45: 508–512.
- Suh PG, Park JI, Manzoli L, Cocco L, Peak JC, Katan M *et al.* (2008). Multiple roles of phosphoinositide-specific phospholipase C isozymes. *BMB Rep* 41: 415–434.
- Suzuki H, Takei M, Yanagida M, Nakahata T, Kawakami T, Fukamachi H (1997). Early and late events in Fc epsilon RI signal transduction in human cultured mast cells. *J Immunol* 159: 5881–5888.
- Vendelin J, Laitinen C, Vainio PJ, Nissinen E, Maki T, Eklund KK (2005). Novel sulfhydryl-reactive compounds orazipone and OR-1958 inhibit cytokine production and histamine release in rat and human mast cells. *Int Immunopharmacol* 5: 177–184.
- Wang Q, Matsuda H, Matsuhira K, Nakamura S, Yuan D, Yoshikawa M (2007). Inhibitory effects of thunberginols A, B, and F on degranulations and releases of TNF-alpha and IL-4 in RBL-2H3 cells. *Biol Pharm Bull* 30: 388–392.
- Weston MC, Anderson N, Peachell PT (1997). Effects of phosphodiesterase inhibitors on human lung mast cell and basophil function. *Br J Pharmacol* 121: 287–295.
- Yuan M, Li J, Lv J, Mo X, Yang C, Chen X *et al.* (2012). Polydatin (PD) inhibits IgE-mediated passive cutaneous anaphylaxis in mice by stabilizing mast cells through modulating Ca(2+) mobilization. *Toxicol Appl Pharmacol* 264: 462–469.