

Basophils and Mast Cells in Chronic Idiopathic Urticaria

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Chronic idiopathic urticaria (CIU) is diagnosed in patients when urticarial eruptions recur for more than 6 weeks, and no specific cause is determined. Given that urticaria resembles the lesions induced by injection of histamine or allergen into the skin, a role for mast cells or basophils has been proposed in the generation of localized urticarial lesions. However, currently, the exact mechanisms governing regional mast cell or basophil activation are unknown. In the past decade, there has been mounting interest in viewing CIU as an autoimmune disease, given the presence of circulating autoantibodies to IgE or the alpha subunit of the high-affinity IgE receptor (FcεRI) in a subset of patients. In this review, we propose that in addition to autoantibodies, specific differences in the expression of FcεRI-signaling molecules in the basophils or mast cells of CIU patients may contribute to the generation of urticarial eruptions.

Introduction

Urticaria is a common disorder affecting up to 25% of the US population. Acute urticarial eruptions can be triggered by allergic reactions to medications, food, and insect venom from stings, or by stimulation of another immune system pathway, such as viral infections [1••]. In contrast, physical urticarias are triggered by specific stimuli, ranging from exposure to cold, heat, pressure, vibration, or wavelengths of light, and typically last less than 4 hours [2]. If urticarial eruptions recur for more than 6 weeks, the process is called chronic urticaria (CU), and in most cases, no specific cause is determined—hence, the label chronic idiopathic urticaria (CIU). There is a wide clinical spectrum of disease in CIU, including the distribution of urticaria, intensity of pruritus, and presence of angioedema or swelling (~40% of CIU), suggesting a heterogeneity in disease

origin and mediator release. Previous studies have not shown an increase of disease frequency in atopic patients, although there is increased incidence in women [2]. The typical duration for a single CIU lesion is between 4 and 36 hours [2]. Among the conditions that have been associated with CIU are thyroid autoantibodies (antimicrosomal or antithyroglobulin in 12% of cases [3••]) as well as chronic infections such as *Helicobacter pylori* [4,5]. Also, alternate diagnoses, such as chronic hepatitis-associated cryoglobulinemia or urticarial vasculitis, which can mimic CIU, must be excluded. Given that urticaria resembles the lesions induced by injection of histamine or allergen into the skin, a role for mast cells or basophils has been proposed in the generation of localized urticarial lesions. However, currently, the exact mechanisms governing regional mast cell or basophil activation are unknown.

Because the duration of CIU is unpredictable, patients are frequently frustrated by symptoms, which may spontaneously disappear within 12 months, but may also continue for years. A recent 5-year prospective study of 136 subjects states that at 6 months, 94% are still active, 75% at 12 months, 52% at 24 months, 43% at 36 months, and 14% at 5 years. Factors associated with longer CIU disease duration include the presence of angioedema, severe disease, and the presence of autoimmune features (*eg*, presence of autologous serum skin test [see later] or antithyroid antibodies) [3••].

Current therapies for CIU are solely symptomatic, and range from the use of only occasional H1-receptor antagonists in mild disease to an extensive list of add-on agents, including H2 receptor antagonists, leukotriene receptor antagonists (LTRA), and tricyclic compounds as well as short-term, systemic corticosteroids [1••]. In severe cases, several drug categories are used simultaneously, and in steroid-dependent cases, alternatives such as cyclosporine have been tried with success [6]. Although these agents are generally safe, it is clear that they offer only temporary relief of the symptoms of pruritus and discomfort. Due to a lack of understanding of the underlying disease process, none of these agents are currently curative. Given the emergence of specific immunomodulatory drugs for allergic disease, such as omalizumab, particularly in the arena of FcεRI-mediated activation of mast cells and basophils [7], there is interest in their application to a disease such as CIU.

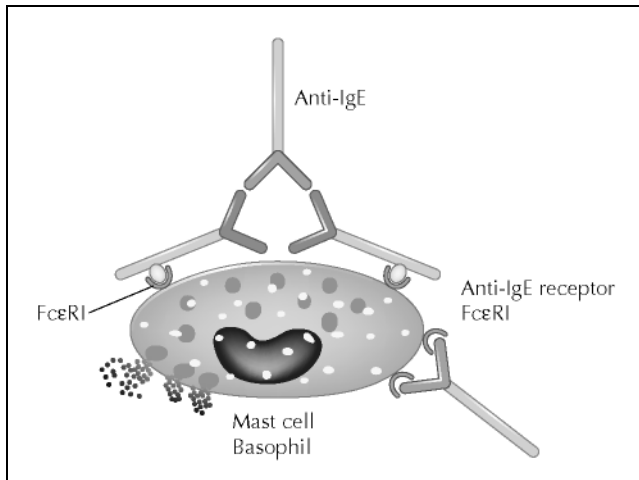


Figure 1. Types of autoantibodies. Autoantibodies to the alpha subunit of the FcεRI and to IgE have been reported to activate basophils and mast cells in vitro. FcεRI—high-affinity IgE receptor; IgE—immunoglobulin E.

Role of Serologic Factors: Evidence for and Against A Pathogenic Role of Autoantibodies

In the past decade, there has been mounting interest in viewing CIU as an autoimmune disease, given the absence of identifiable external triggers to explain the great majority (> 90%) of cases and high prevalence of thyroid autoantibodies in CIU subjects [1••]. Several groups have described the presence of circulating autoantibodies to immunoglobulin E (IgE) or the alpha subunit of the high-affinity IgE receptor (FcεRIα; Fig. 1). Specifically, approximately 40% of patients are reported to have serum autoantibodies, of which 90% are directed against FcεRIα, whereas 10% target IgE [2,8]. It has also been noted that another 40% of CIU patients possess serum histamine-releasing activity (HRA) in vitro against donor basophils [9]. Gruber, Hide, *et al.* [10,11] were the first to describe such CIU serum HRA in vitro. Additionally, in severe refractory CIU, case reports of plasmapheresis to remove such autoantibodies has been shown to be successful [12]. Methods to detect autoantibodies include skin injection with autologous serum to look for wheal formation (autologous skin test [AST]), as well as functional assays measuring serum HRA from normal blood basophils or skin mast cells [9]. In addition, Western blotting and plate-bound assays using purified FcεRIα protein have also been developed to detect autoantibodies to the FcεRI [9].

In terms of CIU pathogenesis, it has been suggested that these antibodies are functional in vivo and, at least in vitro, involve complement activation upon attachment to either mast cells or basophils and thereby stimulate mediator release [13–15]. A prospective analysis of the serum of 50 CIU subjects for HRA on basophils and for induction of mediator release from rat basophilic leukemia cells (RBLs) transfected with the human FcεRIα chain revealed 38 sera capable of releasing β-hexosaminidase from RBLs whereas

20 sera had positive basophil HRA [8]. In total, 30 subjects had IgG anti-FcεRIα antibodies, and six had IgG anti-IgE antibodies. In more recent studies, Kikuchi and Kaplan [13,14] have demonstrated that the IgG-purified fraction of serum bears serum HRA, which can be neutralized by preincubation of the IgG with FcεRIα protein. The addition of normal serum with intact complement factors to IgG fractions enhanced HRA, and HRA could be blocked by a specific antibody to the C5a receptor [13,14]. A role for complement-mediated activation via C5a has also been proposed to explain why only skin mast cells are triggered in CIU rather than mucosal mast cells such as those found in the lung, which lack responsiveness to the C5a receptor [16].

However, several unresolved issues exist with the autoantibody theory. Although it has been assumed by some investigators that serum HRA and serum immunoreactivity to FcεRIα detect the same entity, there is a significant discordance between the detection of autoantibodies using these two serum-based assays in CIU subjects [13,17•]. An explanation for this discrepancy among serum-based assays has not yet been elucidated, and the lack of a “gold-standard” assay to identify autoimmune subjects highlights one of the issues with the autoantibody hypothesis. Additionally, systemic complement depletion is not a feature of subjects with active CIU [3••] nor is complement deposition a feature noted on skin biopsies of active CIU lesions. Although certain CIU sera can induce significant mediator release from a healthy donor’s basophils in vitro, extensive basophil or mast cell activation and mediator release, such as that observed in anaphylaxis, is rarely reported in CIU in vivo. Furthermore, the reason that such autoantibodies may act only in distinct regions of the skin is also not clear.

Despite the attractive hypothesis regarding autoantibodies as a cause for CIU, several further issues remain. First, the pathogenic defect in most CIU subjects who lack autoantibodies or HRA remains unclear. Second, the published reports of autoantibody detection have used a recombinant unglycosylated FcεRIα protein of mass ~30 kD, which is roughly half the mass of the mature surface-expressed protein. In our previous human basophil IgE receptor studies, we demonstrated that mature human alpha chain, which is known to be heavily glycosylated, occurs as a heterogeneous surface protein of ~60 kD mass as well as an immature, homogenous, cytoplasmic form at 50 kD [18]. Therefore, the current assay would only be relevant if CIU basophils expressed an immature FcεRIα on their cell surface. Additionally, in antibody-negative subjects, there is also evidence for the presence of other histamine-releasing factors acting on both mast cells and basophils (such as cytokines, and complement components), suggesting great heterogeneity in serum HRA [17•].

Selective expression of such autoantibodies in CIU versus other diseases is also an issue. Fiebiger *et al.* [19] examined serum from CIU subjects and from other autoimmune skin diseases for the presence of anti-FcεRIα

antibodies by an enzyme-linked immunosorbent assay (ELISA). They found anti-FcεRIα antibodies in the sera of 38% of CIU, 39% of pemphigus vulgaris, 36% of dermatomyositis, 20% of systemic lupus erythematosus (SLE), and 13% of bullous pemphigoid subjects. Although antibody titers were similar among the diseases, only CIU subjects had basophil HRA, and the antibodies were of IgG₁ or IgG₃ isotype, in support of a role for complement fixation. In the other non-CIU skin diseases, the predominant isotypes were IgG₂ or IgG₄. It has also been reported that anti-FcεRIα autoantibodies are part of the naturally occurring repertoire of the immune system [20].

A comparison of skin biopsies from CIU subjects with or without serum HRA did not detect significant differences in cellular infiltrates of wheals less than 4 hours old, whereas wheals older than 12 hours duration had increased eosinophil numbers in subjects lacking autoantibodies [21]. It has been reported that, clinically, subjects with HRA [22] have more severe disease and those with the dual presence of both HRA and autoantibodies have the greatest disease severity [17•].

Basophils in Chronic Idiopathic Urticaria: Evidence for A Cellular Defect

A role for basophils in CIU dates to the early descriptions of the disease in which basopenia was noted [23]. The reduction of blood basophils in CIU subjects was noted by Grattan *et al.* [24], who compared basophil numbers determined by traditional granule staining to the numbers of IgE-bearing cells detected by flow cytometry. The results showed that basophil numbers were comparable by both methods and especially reduced in subjects with significant serum HRA. Among the possible explanations for this basopenia are that basophils are recruited into tissue sites or are depleted due to serologic factors. However, as noted below, serologic factors such as serum HRA do not reveal impressive differences in basophil number on skin biopsies.

An early study comparing basophils from CIU subjects to those from nonatopic subjects revealed a reduction in anti-IgE-induced histamine release in CIU basophils but no difference was noted among the subjects in calcium ionophore-induced histamine release, indicating a specific defect in the FcεRI pathway [25]. In follow-up, CIU subjects were noted to have increased anti-IgE-induced histamine release in parallel with reductions in urticaria symptoms. Basophil numbers are also negatively related to urticarial activity scores [26]. Thus, there is evidence that basophil functional changes are transiently present and mirror CIU disease, suggesting that factors associated with active CIU disease impart these cellular changes [25]. A more recent study of basophils of CIU noted that subjects with serum HRA as a group had greater reductions in anti-IgE-induced histamine release and basophil number compared with HRA-negative subjects [27]. However, it is worth noting that a large number

of the HRA-negative (9/14) subjects also had significantly reduced anti-IgE basophil release. Again, non-receptor-mediated stimuli such as N-formyl-L-methionyl-L-leucyl-L-phenylalanine (fMLP) were not different in either CIU category from normals. Thus, there appears to be a specific acquired defect in histamine release via the anti-IgE pathway in the basophils of CIU subjects that may be independent of the presence of serum HRA.

In regard to CIU treatment, basophil-mediator release responses of all types (histamine, leukotrienes, and cytokines) are sensitive to inhibition by corticosteroids and, likewise, CIU disease activity is known to rapidly respond to systemic corticosteroid therapy. In contrast, skin mast cell degranulation is not generally steroid suppressed [28], supporting a role for basophils in CIU. Reduced blood basophil numbers have been reported to rise paradoxically in CIU subjects in response to systemic corticosteroids, whereas in normal subjects, the numbers fall [26]. It is known that steroids can inhibit basophil migration as well as recruitment to the skin in allergen-induced reactions [29]. This finding is especially notable given that steroids also induce basophil apoptosis, but this may be overcome in cells exposed to interleukin (IL)-3 [30].

Biopsy Studies: Cellular and Cytokine Patterns

More recently, skin biopsy data comparing CIU lesions to normal skin noted that CIU had increased CD3+, CD4+, CD8+, eosinophil, neutrophil, and basophil numbers. As in an earlier study, no cellular differences were seen in the biopsies of subjects with serum HRA, including basophil numbers [31•]. Skin biopsies in CIU show tissue edema, vascular dilatation, and a perivascular infiltrate of mononuclear cells, including CD4+ T cells. The leukocyte infiltrate of urticarial lesions is similar to that of allergen-induced, late-phase reactions, but CIU lesions express messenger RNA (mRNA) for both T_H2 (IL-4 and IL-5) and T_H1 (interferon [IFN]-γ) cytokines [31•,32]. Although there is some overlap in the histology of allergen-induced and CIU biopsies, the pathways to explain basophil arrival in skin tissues are not known. The specific molecules (chemokines and their receptors) involved in the recruitment of basophils to the skin in CIU are poorly characterized [32].

Studies comparing skin mast cell numbers in CIU have not shown significant differences from control subjects [32]. Furthermore, studies of mast cell numbers in lesional and nonlesional skin of CIU patients have not revealed an increase in mast cell number by tryptase and chymase staining [33]. However, the activation status of tissue resident mast cells or recruited basophils may be relevant to CIU disease expression and is just beginning to be explored. In a recent study, Caproni *et al.* [34] compared the tissue infiltrate in AST-induced wheals to the same CIU patient's uninvolved skin and the skin of healthy volunteers. A latent inflammatory state was noted in the uninvolved skin of CIU patients, consisting of lymphocytes

(CD3+, CD4+, CD8+) and basophils (BB1 positive), compared with the skin of healthy volunteers. AST-induced wheals demonstrated significant staining for IL-4, IL-5, and IFN- γ , again consistent with mixed T_H1/T_H2 response.

Mast Cells in Chronic Idiopathic Urticaria

Mast cell involvement in CIU is not generally disputed, but earlier studies reporting increased mast cell numbers in CIU-patient skin biopsies have not been reproduced [33]. Furthermore, there is no evidence of increased circulating alpha tryptase levels in CIU, an indicator of total body mast cell number [35], nor evidence of the increased presence of stem cell factor in the serum of CIU subjects [36]. In the case of physical urticarias, there is strong evidence of mast cell release coincident with the onset of the physical stimuli [37]. In humans, it is also recognized that different locations of the body yield different types of mast cells and that for skin, the predominant phenotype is a chymase/tryptase-positive cell [38]. However, the reason for regional mast cell activation in CIU is still not clear. Furthermore, there are limited data to support that increased mast cell releasability may occur in the skin [39].

In regard to CIU disease, it is well known that a variety of infections can stimulate exacerbations of the underlying condition [5], and recently the potential pathways for mast cell–pathogen interactions have become more clear. Given the diverse stimuli potentially available to activate mast cell engagement from viral, bacterial, or IgG sources, it is possible that the skin mast cells are activated via such pathways in the setting of CIU. Certainly, the role of *Staphylococcus aureus* in exacerbating atopic dermatitis is one example of bacterial modulation of a chronic skin disease [40]. In addition, given the expression of a T_H1 cytokine pattern on CIU skin biopsies, the presence of such alternate pathways for engagement seems plausible.

For example, there is increasing evidence that mast cells can respond to a variety of stimuli associated with innate immunity. Much of this knowledge has been gained via a rodent model of bacterial sepsis [41], but there are also emerging data in human cultured mast cells for this aspect of mast cell functions [42]. This mast cell activation pathway involves the toll-like receptors (TLR), which are highly conserved pattern-recognition receptors that play a role in host defense by engaging microbial products from a variety of pathogens. At least 10 receptors have been described, and all share similar intracellular domains homologous to IL-1 receptors. The best studied is the interaction of TLR4 with lipopolysaccharide (LPS) and the LPS-binding protein CD14. It has been noted that exposure of human mast cells to either IFN- γ or IL-4 induces a functional TLR4 [43]. It has been reported that mast cells also express functional high-affinity IgG receptors after IFN- γ exposure and can be activated by different stimuli in a manner similar to that via the IgE receptor, leading to both preformed and synthesized mediator release [44].

Cellular Releasability and Fc ϵ RI Signaling Pathways

In terms of basophil and mast cell Fc ϵ RI receptor signal transduction pathways, aggregation of receptors by a multivalent allergen or, potentially, an anti-Fc ϵ RI autoantibody, allows the tyrosine kinase Lyn to initiate signal transduction by transphosphorylation of specific residues in the receptor's β and γ subunits [45]. In addition to this positive role in signaling, Lyn both positively (*eg*, Syk kinase) and negatively (*eg*, Fyn kinase) regulates the activation of numerous additional signaling molecules. The aggregation of Fc ϵ RI ultimately results in the secretion of preformed (histamine, tryptase, chymase) and newly synthesized (cytokines, chemokines, prostaglandins, leukotrienes) mediators. In the context of CIU, activation of the Fc ϵ RI pathway on basophils or mast cells may contribute to both the clinical symptoms (histamine-induced pruritis) and the chronicity of the disease (chemokine recruitment of lymphocytes and eosinophils).

Conroy *et al.* [46] defined releasability several decades ago as the control of release of mediators from basophils in response to different stimuli. Independent of the surface expression of Fc ϵ RI or serum IgE, asthmatic donor basophils were found to release significantly more histamine when treated with D₂O than normal donor basophils [47]. Since then, specific changes in the expression of positive or negative signaling proteins have been associated with changes in basophil releasability (Fig. 2). As discussed in more detail subsequently, genetic or chemical ablation of these signaling proteins in rodent cells results in increased or decreased secretion of inflammatory mediators.

Syk tyrosine kinase is recruited to tyrosine phosphorylated Fc ϵ RI γ subunits and phosphorylates signaling molecules, including Shc and PLC γ 2 after Fc ϵ RI stimulation [45]. The absence of Syk in chemically treated RBL-2H3 cells prevents Fc ϵ RI secretion of granular contents, and cytokines. Furthermore, so-called non-releaser basophils are deficient in Syk or both Syk and Lyn protein and release less than 5% of total histamine content after anti-IgE treatment [48,49]. Likewise, a recent report described the characterization of nonreleasing human mast cells [50].

A novel Src homology 2 (SH2)-containing inositol phosphatase (SHIP-1) has been gaining prominence as a gatekeeper of the IgE-mediated response and as a negative regulator of hematopoietic cell activation, survival, and proliferation [51,52•]. SHIP-1 is a recently cloned hematopoietic signaling molecule with multiple forms (145, 135, 125, 110 Kd) that hydrolyzes phosphatidylinositol-3, 4,5 triphosphate (PIP3), thereby limiting a crucial step between the release of calcium from intracellular stores and the influx of extracellular calcium. Mast cells of SHIP-1 knockout mice more readily degranulated after IgE receptor activation or even IgE sensitization alone. Similarly, in hyperreleasable human basophils, a reduction in SHIP-1 protein levels was associated with heightened response to human recombinant histamine-releasing factor (HrHRF) [53,54]. A related molecule, SHIP-2,

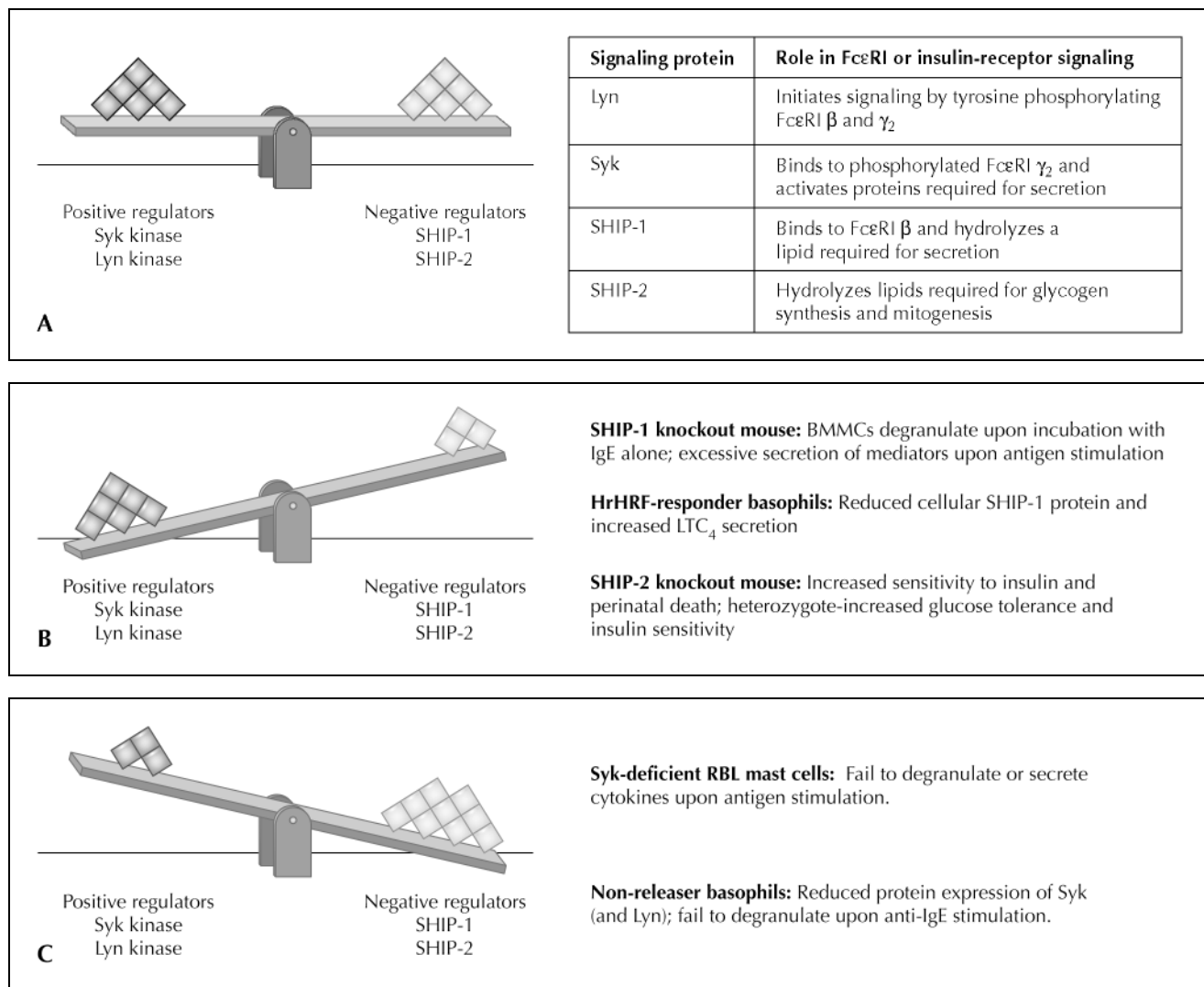


Figure 2. Basophil releasability and FcεRI signaling. Specific changes in the expression of positive or negative signaling proteins are associated with changes in basophil releasability. Genetic or chemical ablation of these signaling proteins in rodent mast cells results in increased or decreased secretion of inflammatory mediators. **A**, homeostasis-normal basophils; **B**, hyperreleasable basophils; **C**, hyporeleasable basophils. BMMC—bone marrow-derived mast cells; FcεRI—high-affinity immunoglobulin E receptor; HrHRF—human recombinant histamine releasing factor; LTC—leukotriene C.

also hydrolyzes PIP3 and, in mice, is a potent, negative regulator of insulin signaling and sensitivity [55]. Mutations in the *SHIP-2* gene contribute to the genetic susceptibility to type 2 diabetes in rats and humans [56].

As represented in Figure 2, a decrease in the expression of a negative regulator (SHIP-1) may contribute to the basophil hyperreleasable state characteristic of many allergic and asthmatic donors [53], whereas the opposite imbalance—the hyporeleasable state noted in CIU basophils—may result from increased expression of negative regulators such as SHIP-1 and SHIP-2 [25,57].

Conclusions

Current biopsy data indicate that both mast cells and basophils are resident in the skin lesions of CIU patients.

Autoantibodies directed to FcεRI or IgE are observed in the sera of a subset of CIU patients that possess either HRA or immunoreactivity to components of the FcεRI. However, the presence of similar autoantibodies in other diseases, as well as the lack of specificity in the assays used to detect them, raise questions about the pathogenic role of these autoantibodies in CIU. We propose that, alternatively, specific differences in the expression of signaling molecules in the basophils or mast cells of CIU patients may contribute to the generation of hives. When present, an anti-FcεRI autoantibody or bacterial infection may exacerbate the disease by stimulating mast cells and basophils to release histamine and cytokines. Examination of circulating basophils as well as tissue-resident mast cells and basophils for changes in expression of FcεRI-specific signaling molecules may provide new targets for the therapy of CIU.

References and Recommended Reading

Papers of particular interest, published recently, have been highlighted as:

- Of importance
- Of major importance

- 1.●● Kaplan AP: Chronic urticaria: pathogenesis and treatment. *J Allergy Clin Immunol* 2004, 114:465–474; quiz 75.
- Excellent review of current clinical treatments and autoantibody theory in CIU.
2. Kaplan AP: Clinical practice: chronic urticaria and angioedema. *N Engl J Med* 2002, 346:175–179.
- 3.●● Toubi E, Kessel A, Avshovich N, et al.: Clinical and laboratory parameters in predicting chronic urticaria duration: a prospective study of 139 patients. *Allergy* 2004, 59:869–873.
- Prospective study of clinical features in CIU related to disease duration.
4. Federman DG, Kirsner RS, Moriarty JP, Concato J: The effect of antibiotic therapy for patients infected with *Helicobacter pylori* who have chronic urticaria. *J Am Acad Dermatol* 2003, 49:861–864.
5. Wedi B, Raap U, Kapp A: Chronic urticaria and infections. *Curr Opin Allergy Clin Immunol* 2004, 4:387–396.
6. Grattan CE, O'Donnell BF, Francis DM, et al.: Randomized double-blind study of cyclosporin in chronic "idiopathic" urticaria. *Br J Dermatol* 2000, 143:365–372.
7. Beck L, Marcotte G, MacGlashan J, et al.: Omalizumab induced reductions in mast cell FcεRI expression and function. *J Allergy Clin Immunol* 2004, 114:527–530.
8. Tong LJ, Balakrishnan G, Kochan JP, et al.: Assessment of autoimmunity in patients with chronic urticaria. *J Allergy Clin Immunol* 1997, 99:461–465.
9. Ferrer M, Kinet JP, Kaplan AP: Comparative studies of functional and binding assays for IgG anti-Fc(εpsilon)RIα (α-subunit) in chronic urticaria. *J Allergy Clin Immunol* 1998, 101:672–676.
10. Gruber BL, Baeza ML, Marchese MJ, et al.: Prevalence and functional role of anti-IgE autoantibodies in urticarial syndromes. *J Invest Dermatol* 1988, 90:213–217.
11. Hide M, Francis DM, Grattan CE, et al.: Autoantibodies against the high-affinity IgE receptor as a cause of histamine release in chronic urticaria. *N Engl J Med* 1993, 328:1599–1604.
12. Grattan CE, Francis DM, Slater NG, et al.: Plasmapheresis for severe, unremitting, chronic urticaria. *Lancet* 1992, 339:1078–1080.
13. Kikuchi Y, Kaplan AP: Mechanisms of autoimmune activation of basophils in chronic urticaria. *J Allergy Clin Immunol* 2001, 107:1056–1062.
14. Kikuchi Y, Kaplan AP: A role for C5a in augmenting IgG-dependent histamine release from basophils in chronic urticaria. *J Allergy Clin Immunol* 2002, 109:114–118.
15. Niimi N, Francis DM, Keremani F, et al.: Dermal mast cell activation by autoantibodies against the high affinity IgE receptor in chronic urticaria. *J Invest Dermatol* 1996, 106:1001–1006.
16. Schulman ES, Kagey-Sobotka A, MacGlashan DW Jr, et al.: Heterogeneity of human mast cells. *J Immunol* 1983, 131:1936–1941.
- 17.● Sabroe RA, Fiebiger E, Francis DM, et al.: Classification of anti-FcεpsilonRI and anti-IgE autoantibodies in chronic idiopathic urticaria and correlation with disease severity. *J Allergy Clin Immunol* 2002, 110:492–499.
- Insight into relationship between serologic factors and CIU disease severity.
18. Saini SS, Richardson JJ, Wofsy C, et al.: Expression and modulation of FcεpsilonRIα and FcεpsilonRIβ in human blood basophils. *J Allergy Clin Immunol* 2001, 107:832–841.
19. Fiebiger E, Hammerschmidt F, Stingl G, Maurer D: Anti-FcεpsilonRIα autoantibodies in autoimmune-mediated disorders. Identification of a structure-function relationship. *J Clin Invest* 1998, 101:243–251.
20. Miescher SM, Horn MP, Pachlopnik JM, et al.: Natural anti-FcεpsilonRIα autoantibodies isolated from healthy donors and chronic idiopathic urticaria patients reveal a restricted repertoire and autoreactivity on human basophils. *Hum Antibodies* 2001, 10:119–126.
21. Sabroe RA, Poon E, Orchard GE, et al.: Cutaneous inflammatory cell infiltrate in chronic idiopathic urticaria: comparison of patients with and without anti-FcεpsilonRI or anti-IgE autoantibodies. *J Allergy Clin Immunol* 1999, 103:484–493.
22. Sabroe RA, Seed PT, Francis DM, et al.: Chronic idiopathic urticaria: comparison of the clinical features of patients with and without anti-FcεpsilonRI or anti-IgE autoantibodies. *J Am Acad Dermatol* 1999, 40:443–450.
23. Grattan CE: Basophils in chronic urticaria. *J Invest Dermatol Symp Proc* 2001, 6:139–140.
24. Grattan CE, Walpole D, Francis DM, et al.: Flow cytometric analysis of basophil numbers in chronic urticaria: basopenia is related to serum histamine releasing activity. *Clin Exp Allergy* 1997, 27:1417–1424.
25. Kern F, Lichtenstein LM: Defective histamine release in chronic urticaria. *J Clin Invest* 1976, 57:1369–1377.
26. Grattan CE, Dawn G, Gibbs S, Francis DM: Blood basophil numbers in chronic ordinary urticaria and healthy controls: diurnal variation, influence of loratadine and prednisolone and relationship to disease activity. *Clin Exp Allergy* 2003, 33:337–341.
27. Sabroe RA, Francis DM, Barr RM, et al.: Anti-Fc(εpsilon)RI auto antibodies and basophil histamine releasability in chronic idiopathic urticaria. *J Allergy Clin Immunol* 1998, 102:651–658.
28. Schleimer RP: Glucocorticosteroids: their mechanism of action and use in allergic diseases. In *Allergy Principles and Practice*, edn 4. Edited by Middleton E, Reed CE, Ellis EF, et al. St. Louis, MO: Mosby; 1993:893–925.
29. Charlesworth EN, Kagey-Sobotka A, Schleimer RP, et al.: Prednisone inhibits the appearance of inflammatory mediators and the influx of eosinophils and basophils associated with the cutaneous late-phase response to allergen. *J Immunol* 1991, 146:671–676.
30. Yoshimura C, Miyamasu M, Nagase H, et al.: Glucocorticoids induce basophil apoptosis. *J Allergy Clin Immunol* 2001, 108:215–220.
- 31.● Ying S, Kikuchi Y, Meng Q, et al.: TH1/TH2 cytokines and inflammatory cells in skin biopsy specimens from patients with chronic idiopathic urticaria: comparison with the allergen-induced late-phase cutaneous reaction. *J Allergy Clin Immunol* 2002, 109:694–700.
- Recent study defining characteristics of cellular infiltrate in CIU.
32. Caproni M, Volpi W, Macchia D, et al.: Infiltrating cells and related cytokines in lesional skin of patients with chronic idiopathic urticaria and positive autologous serum skin test. *Exp Dermatol* 2003, 12:621–628.
33. Smith CH, Kepley C, Schwartz LB, Lee TH: Mast cell number and phenotype in chronic idiopathic urticaria. *J Allergy Clin Immunol* 1995, 96:360–364.
34. Caproni M, Giomi B, Volpi W, et al.: Chronic idiopathic urticaria: infiltrating cells and related cytokines in autologous serum-induced wheals. *Clin Immunol* 2005, 114:284–292.
35. Schwartz LB, Sakai K, Bradford TR, et al.: The alpha form of human tryptase is the predominant type present in blood at baseline in normal subjects and is elevated in those with systemic mastocytosis. *J Clin Invest* 1995, 96:2702–2710.
36. Asero R, Tedeschi A, Lorini M, et al.: Circulating stem cell factor in patients with chronic idiopathic urticaria. *Ann Allergy Asthma Immunol* 2003, 91:79–81.
37. Kaplan AP, Garofalo J, Sigler R, Hauber T: Idiopathic cold urticaria: in vitro demonstration of histamine release upon challenge of skin biopsies. *N Engl J Med* 1981, 305:1074–1077.
38. Irani AA, Schechter NM, Craig SS, et al.: Two types of human mast cells that have distinct neutral protease compositions. *Proc Natl Acad Sci U S A* 1986, 83:4464–4468.
39. Jacques P, Lavoie A, Bedard PM, et al.: Chronic idiopathic urticaria: profiles of skin mast cell histamine release during active disease and remission. *J Allergy Clin Immunol* 1992, 89:1139–1143.
40. Leung DY: Infection in atopic dermatitis. *Curr Opin Pediatr* 2003, 15:399–404.

41. Supajatura V, Ushio H, Nakao A, *et al.*: Differential responses of mast cell Toll-like receptors 2 and 4 in allergy and innate immunity. *J Clin Invest* 2002, 109:1351–1359.
 42. Marshall JS, McCurdy JD, Olynch T: Toll-like receptor-mediated activation of mast cells: implications for allergic disease? *Int Arch Allergy Immunol* 2003, 132:87–97.
 43. Varadaradjalou S, Feger F, Thieblemont N, *et al.*: Toll-like receptor 2 (TLR2) and TLR4 differentially activate human mast cells. *Eur J Immunol* 2003, 33:899–906.
 44. Tkaczyk C, Okayama Y, Metcalfe DD, Gilfillan AM: Fcγ receptors on mast cells: activatory and inhibitory regulation of mediator release. *Int Arch Allergy Immunol* 2004, 133:305–315.
 45. Blank U, Rivera J: The ins and outs of IgE-dependent mast-cell exocytosis. *Trends Immunol* 2004, 25:266–273.
 46. Conroy MC, Adkinson NF Jr, Lichtenstein LM: Measurement of IgE on human basophils: relation to serum IgE and anti-IgE-induced histamine release. *J Immunol* 1977, 118:1317–1321.
 47. Findlay SR, Lichtenstein LM: Basophil releasability in patients with asthma. *Am Rev Respir Dis* 1980, 122:53–59.
 48. Kepley CL, Youssef L, Andrews RP, *et al.*: Syk deficiency in non-releaser basophils. *J Allergy Clin Immunol* 1999, 104:279–284.
 49. Lavens-Phillip S, MacGlashan D: The tyrosine kinases, p53 lyn, p72 syk are differentially expressed at the protein level, not the mRNA level in non-releasing human basophils. *Am J Cell Respir Mol Med* 2000, 23:566–571.
 50. Kepley CL, Cohen N: Evidence for human mast cell nonreleaser phenotype. *J Allergy Clin Immunol* 2003, 112:457–459.
 51. Huber M, Helgason CD, Damen JE, *et al.*: The src homology 2-containing inositol phosphatase (SHIP) is the gatekeeper of mast cell degranulation. *Proc Natl Acad Sci U S A* 1998, 95:11330–11335.
 52. Rauh MJ, Krystal G: Of mice and men: elucidating the role of SH2-containing inositol 5-phosphatase (SHIP) in human disease. *Clin Invest Med* 2002, 25:68–70.
- Excellent review of the clinical implications of SHIP phosphatase activity in human disease.
53. MacDonald SM, Rafnar T, Langdon J, Lichtenstein LM: Molecular identification of an IgE-dependent histamine-releasing factor. *Science* 1995, 269:688–690.
 54. Vonakis BM, Gibbons S Jr, Sora R, *et al.*: Src homology 2 domain-containing inositol 5' phosphatase is negatively associated with histamine release to human recombinant histamine-releasing factor in human basophils. *J Allergy Clin Immunol* 2001, 108:822–831.
 55. Clement S, Krause U, Desmedt F, *et al.*: The lipid phosphatase SHIP2 controls insulin sensitivity. *Nature* 2001, 409:92–97.
 56. Marion E, Kaisaki PJ, Pouillon V, *et al.*: The gene INPPL1, encoding the lipid phosphatase SHIP2, is a candidate for type 2 diabetes in rat and man. *Diabetes* 2002, 51:2012–2017.
 57. Saini SS, Vasagar K, Huang F, *et al.*: Signaling defects in basophils in chronic urticaria. *J Allergy Clin Immunol* 2003, 111:S178.