

Differential Diagnosis of Chronic Urticaria and Angioedema Based on Molecular Biology, Pharmacology, and Proteomics



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KEYWORDS

- Autoantigen • Proteomics • Virokine • HERV • Herpes • Metagenome
- Microbiome omalizumab • Autoimmune

KEY POINTS

- Differential diagnosis and classification of chronic urticaria and angioedema should reflect advances in molecular biology, pharmacology, and proteomics.
- Advances in molecular biology and genetics illustrate the importance of the acquired immune system, autoreactive autoantibodies, pattern recognition molecules, and complement proteins.
- Chronic inflammation in chronic urticaria and angioedema may be related to complex interactions between the host immune system and the metagenome.

INTRODUCTION

Several recent practice guidelines have produced consensus differential diagnosis schemes for urticaria and angioedema with input from both US and European investigators.^{1–5} The reader is referred to these comprehensive summaries for a traditional perspective on the differential diagnosis and therapy of urticaria and angioedema. Although historical classification schemes are useful, many patients with chronic urticaria do not neatly fit into current categories. Novel agents such as omalizumab (Xolair) approved for therapy of chronic urticaria, also often defy current classification schemes because some forms of chronic urticaria thought to be distinct in mechanism have similar responses to omalizumab therapy.

For the purposes of this article, urticaria and angioedema that persist for more than 6 to 8 weeks are termed “chronic urticaria,” whatever the disease phenotype. However,

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many forms of urticaria and angioedema that seem to be phenotypically similar differ in mechanisms and response to therapy.^{6–9} Familial or hereditary forms of urticaria, although rare, have recently also been extensively characterized at a molecular level, and some patients even with no family history of urticaria may in the future be found to harbor some variants of these novel genes. With the decreasing costs of whole exome and whole genome sequencing and it is likely in the future that many families will have questions regarding genetic data available regarding specific gene mutations either in germ line cells or somatic tissues that were not available previously.^{10–12}

For example, patients with angioedema owing to hereditary defects in the C1 inhibitor protein require different therapy than patients with angioedema owing to medications or autoreactive autoantibodies. Depending on the underlying mechanism recurrent episodes of angioedema, which can occur both with and without urticaria, some cases respond to omalizumab, some cases to discontinuation of medications such as angiotensin-converting enzyme inhibitors, whereas other cases respond to a variety of new therapies targeting the C1 inhibitor protein.^{1,13} A diagnosis of hereditary angioedema, which is covered in more detail elsewhere in this issue, is currently divided into at least 3 categories (types 1, 2, and 3) based on distinct molecular mechanisms affecting the C1 inhibitor protein function or levels, but other proteins in the complement pathway may also be involved, leading to altered requirements and response to therapy.¹ The acquired form of angioedema associated with deficiency of C1 inhibitor proteins is usually associated with either malignant (type 1) or autoimmune disease (type 2) also recently characterized both by molecular mechanisms and response to therapy.¹³

Autoreactive antibodies are also present in many cases of chronic urticaria as are associations with other autoimmune syndromes such as autoimmune thyroid disease, although the mechanism underlying these associations is debated.^{14–17} Improved methods of immune assays such as peptide arrays and proteomics can provide detailed molecular information about the targets of autoreactive antibodies in some forms of chronic urticaria, as will be discussed in more detail elsewhere in this review. Contributing still more complexity, increasing understanding of the microbiome and the metagenome (viruses, bacteriophage, and endogenous retroviruses) is likely to provide additional insight into urticaria and angioedema diagnosis as the prevalence of both atopy and autoimmune disease increases.¹⁸

In this review, the author provides a brief summary of conventional classification of urticaria and angioedema based on disease phenotype, because characterization of disease based on phenotype is a sufficient for clinicians in many cases. However, the author also proposes that new information with regard to disease mechanisms should be incorporated into future classification schemes (**Fig. 1**). The author then provides some suggestions regarding how a new understanding of disease mechanisms should ideally be incorporated and combined with new molecular information and response to therapy into any new classification schemes of urticaria and angioedema. The intention of the author is to augment the existing schemes of classification as outlined by incorporating specific molecular features of the innate immune system versus acquired immunity as well as response to conventional antihistamines, corticosteroids, and novel therapy such as cyclosporine and omalizumab into the diagnostic framework.

TRADITIONAL SCHEMES OF CLASSIFICATION OF CHRONIC URTICARIA BASED ON DISEASE PHENOTYPE AND THEIR LIMITATIONS IN DIFFERENTIAL DIAGNOSIS

As shown in **Fig. 1**, chronic urticaria and angioedema can be classified on the phenotypic presentation without any reference to underlying mechanism. In the author's own

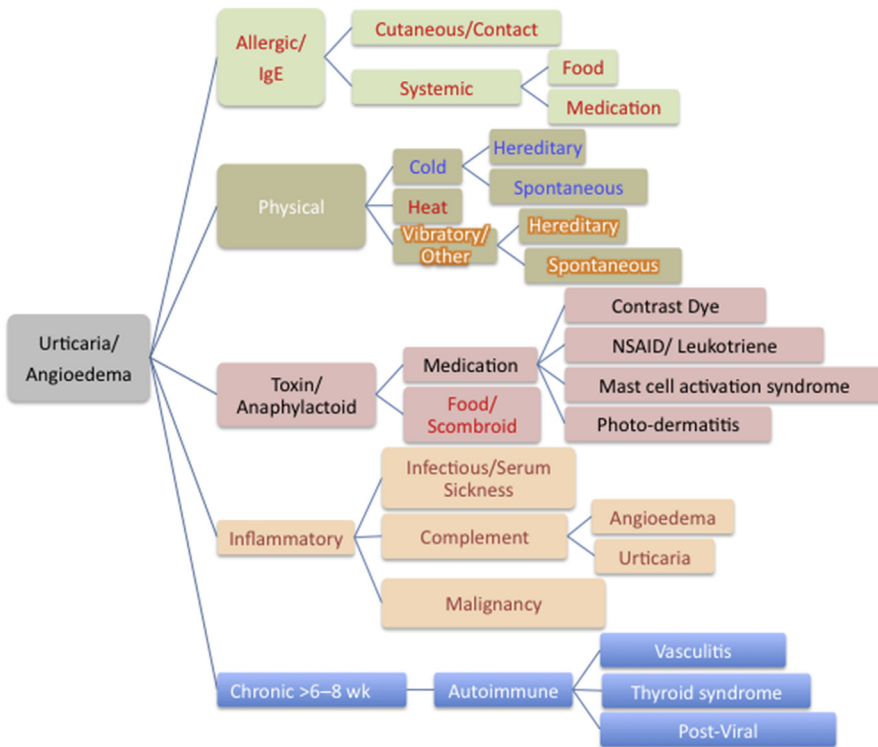


Fig. 1. Phenotypic classification of urticaria and angioedema. This figure is intended to provide a framework for clinical use based on the author's and others' experience that most presentations of urticaria and angioedema can be grouped in distinct categories that can predict prognosis and response to therapy. IgE, immunoglobulin E.

clinical experience most cases fall into 5 basic clinical presentations: (1) allergic or immunoglobulin (Ig)E mediated, (2) physical, (3) toxin mediated, (4) inflammatory, and (5) chronic. Of course, these categories are not mutually exclusive; for example, patients with an inflammatory urticaria owing to a malignancy may have exacerbations owing to allergy, cold, heat or other physical stimuli, medications, or toxins. Each of these categories may in turn be further divided, for example, with physical urticaria owing to cold versus heat, pressure, or vibration.

Often, these phenotypes show little or no overlap; for example, patients with cold urticaria very rarely or never in the author's experience present with simultaneous heat-triggered urticaria. For this reason, some classification schemes have proposed that the underlying mechanisms of, for example, cold-induced physical urticaria might be owing to a cold-sensitive immunoglobulin, whereas heat-induced urticaria (often also termed cholinergic urticaria) might reflect cholinergic neuromediators rather than immunoglobulins. Although published literature beyond the scope of this review supports each of these mechanisms, in the authors opinion terms such as "cholinergic" are mechanistic rather than based on phenotype and should be reserved for cases in which specific neuromediators have been demonstrated. For clinical purposes, it is often sufficient to state that the patient's symptoms are triggered by reduction or elevation of local or core body temperature, or other physical causes without speculating about the mechanism or mechanisms involved.

Conversely, as shown in **Box 1**, it is important to note whether the patient's urticaria is related to a specific IgE and thus allergic in nature, because in most cases the presence of a specific IgE can be determined by skin testing or immunoassay and this finding may then be considered part of the disease phenotype. Testing for specific IgE is of great importance, for example, in establishing whether a patient who has had 6 months of daily hives has chronic urticaria or instead is suffering from living with a new partner who has pet cats. However, as noted in recent guidelines, extensive and unfocused "screens" for environment or food triggers in patients with urticaria and angioedema are seldom productive and not useful or recommended. Similarly, if all the clients of a certain restaurant are suddenly incapacitated with urticaria and angioedema after eating a certain fish dish, the disease phenotype obtained by history is sufficient in many cases to implicate scrombroid or other food toxins without further testing for a specific IgE to fish.

Also as noted in **Fig. 1**, some phenotypes of urticaria and angioedema are extremely important in suggesting appropriate therapy, for example, angioedema without urticaria should immediately suggest a different approach to therapy, such as discontinuation of angiotensin-converting enzyme inhibitors, evaluation for an occult malignancy, or evaluation of the patient and other family members for hereditary angioedema or a related defect in complement the C1 inhibitor protein, as reviewed elsewhere in this issue. Also as noted, if a specific IgE can be associated with the urticaria angioedema phenotype, or a particular medication such as nonsteroidal antiinflammatory drugs can be implicated, then desensitization may be useful and even life saving in many cases. Urticaria and angioedema related to ingestion of a class of medications such as several different nonsteroidal antiinflammatory drugs is usually related to a metabolic imbalance in leukotriene and prostaglandin metabolism, whereas a reaction to a single nonsteroidal antiinflammatory drug may suggest an IgE-mediated pathology. Often, although not always, IgE-, physical-, or toxin-mediated urticaria and angioedema will respond to conventional antihistamines and oral corticosteroids, whereas inflammatory or chronic forms of urticaria and angioedema often will not respond to conventional therapy.

Box 1

Chronic urticaria/angioedema mechanism based terminology: an outline

1. Acute urticaria and angioedema
 - a. IgE/allergy
 - b. IgG/infection
 - c. Complement
 - d. Innate/mast cell hyperrelease syndrome
- 2.1. Chronic, familial
 - Urticaria predominant/"gene name"
 - Angioedema predominant/"gene name"^a
- 2.2. Chronic, acquired
 - IgG/IgE autoimmune predominant
 - Autoantigen (s)
 - Antibody type
 - Viral or infection related^a
 - Physical predominant
 - Angioedema predominant^a

The author proposes that terms such as "idiopathic" and "spontaneous" should be eliminated and replaced by specific molecular and genetic data to the extent that these are known.

^a These terms are discussed in more detail in the text and figures.

Phenotypic classification schemes as shown in [Fig. 1](#) have been less useful, or in many cases of no use at all in the author's experience, in the case of chronic urticaria. Chronic urticaria often seems to resist any classification at all other than the fact that the urticaria and angioedema goes on for more than 6 to 8 weeks, and often for years. Although some associations with autoimmune disease have been established, it is often difficult to correlate these observations with prognosis or response to therapy, suggesting that there is some a lack of understanding of the mechanism underlying the phenotype. In frustration, many clinicians have adopted terms such as "idiopathic" and/or "spontaneous" to categorize chronic urticaria, terms that in the author's opinion add little or nothing to the diagnosis. Instead, for the remainder of this work, the author focuses on how new technology may clarify the underlying mechanisms of chronic urticaria and angioedema, and suggest that this information should be incorporated into future terminology and differential diagnosis.

DIFFERENTIAL DIAGNOSIS OF CHRONIC URTICARIA AND ANGIOEDEMA SHOULD INCORPORATE TERMS DERIVED FROM ADVANCES IN MOLECULAR BIOLOGY AND GENETICS

As noted, traditionally depending on the phenotype urticaria and angioedema were considered to have either "immunologic" or "nonimmunologic" mechanisms depending on whether immunoglobulin played a role in the disease.^{13,17} More recently, this terminology has become antiquated with the discovery of the innate immune system that does not rely on immunoglobulin but rather conserved "pattern receptors."^{19,20} Thus, the author proposes that the differential diagnosis of hereditary or acquired urticaria and angioedema should contain specific terms indicating whether the disease is primarily mediated by either the innate or the acquired system. Also, complement interacts with both acquired and innate immunity, and complement proteins if responsible should be identified (see [Box 1](#)). Terms such as "idiopathic" or "spontaneous" should be phased out whenever possible in the diagnosis of chronic urticaria, and instead components of the acquired or innate immune system including numerous specific pattern recognition molecules or by immunoglobulins or antibodies generated by the acquired immune should be specified to the extent that this information is known.

Hereditary syndromes are particularly useful and important in understanding underlying mechanisms of chronic urticaria, particularly because there are few or no animal model for chronic urticaria. Increasing numbers of single human gene mutations have been defined in the past several decades. The importance of these mutations is that they show that apparently urticarial phenotype can be generated entirely by an abnormal innate immune response.^{10–12,19,21} Perhaps somatic mutations in these innate immune response genes may also play a role in acquired forms of urticaria and angioedema as well as a diverse population of immunoglobulin-independent "mast cell activation syndromes."^{22,23} As the cost of genome sequencing decreases, it is likely that these innate immune gene mutations will be detected both in germ line tissues and also in somatic tissues contributing to or causing disease.

Similarly, hereditary angioedema without urticaria has historically been categorized as "type 1" resulting from a defect in the C1 inhibitor protein level, "type 2" resulting from a defect in protein function with normal or near normal C1 inhibitor protein levels, and "type 3" resulting from the disease without detected abnormal C1 inhibitor protein levels or function.⁵ The term "type 3" has also sometimes been used to denote a specific form of estrogen-sensitive familial urticaria that in some ethnic groups is associated with a specific abnormality in the factor XII protein.^{24–29} However, most clinicians

who see significant numbers of patients with chronic angioedema encounter patients who do not fall neatly into any of these groups. A more detailed discussion of hereditary angioedema diagnosis, mechanisms and therapy is presented elsewhere in this issue.

To illustrate the limitations of phenotype-based diagnosis of angioedema, for the past decade the author has cared for a family in which females have normal C1 inhibitor protein levels and function by the most sensitive assays, and no described or detectable mutation in factor XII protein and yet have severe estrogen-sensitive angioedema with decreased complement C4 levels during attacks. Females in the family also have severe dysfunctional menstrual bleeding, suggesting that some other component of the coagulation pathway may be involved. Two of the affected females also have allergic urticaria and food-induced IgE-mediated anaphylaxis, possibly unrelated to the hereditary angioedema. Rather than adding more phenotypic subtypes, the author proposes that, like acquired urticaria and angioedema, hereditary angioedema would more accurately be described by the specific mutations in the complement or other proteins when this is known, or by the molecular phenotype such as “hereditary angioedema, estrogen dependent with decreased C4 levels and elevated IgE” in the specific family described.

DIFFERENTIAL DIAGNOSIS OF URTICARIA AND ANGIOEDEMA SHOULD INCLUDE PHARMACOLOGIC INFORMATION INDICATING RESPONSE TO THERAPY

Therapy over the past decade has been revolutionized concurrently with advances in molecular biology and genetics of chronic urticaria. Whereas a decade ago therapy for urticaria was limited largely to antihistamines and occasional oral corticosteroids, most clinicians now use monoclonal antibodies such as omalizumab and immune response-modifying agents such as cyclosporine. Although a full review of on- and off-labels uses for therapy of urticaria and angioedema is beyond the scope of this review, omalizumab may illustrate the complexity of response to these new therapies.

Omalizumab was first developed and FDA approved for asthma, and later found effective in chronic urticaria.⁶ Omalizumab interferes with free IgE binding to high- and low-affinity IgE receptors on mast cells, basophils, and other hematopoietic cells; however, the effects of xolair in chronic urticaria may or may not be related to its effects on IgE receptors.^{16,30,31} Almost immediately it was noted that most but not all patients with phenotypically similar chronic urticaria responded to omalizumab. In some patients, response was complete, in others partial, and in some not at all.³² In addition, it was soon noted that there is not a simple relationship between serum IgE levels or other autoantibodies such as antithyroid antibodies and response to therapy.³³ This has led to the hypothesis that more complicated or indirect effects such as the downregulation of other cytokine receptors or endogenous viruses may contribute to the response to omalizumab.³⁴

In addition, other forms of chronic urticaria such as physical urticarias triggered by cold seem in many cases to respond rapidly to omalizumab despite no apparent direct mechanistic link between these physical urticarias and IgE.^{35–39} So-called mast cell activation syndromes, although relatively IgE and immunoglobulin independent, may also respond to omalizumab.^{22,23} To help clarify these mysteries, some inclusion of response to therapy would be helpful; for example, it may be that chronic idiopathic or spontaneous urticaria that responds to xolair uses different molecular pathways than the same phenotype or apparent condition that does not respond to xolair. Alternatively, the same mechanism, such as alteration of inflammatory cytokines in the skin might be involved in both responsive and unresponsive forms, but with a dose-related

window of efficacy.^{33,40,41} Alternatively, autoantibodies might be present in some patients that neutralize omalizumab. Similarly, characterization of a response to therapy should be included in the differential diagnosis and description of “mast cell activation” syndromes.^{22,23,42–45}

Hereditary and acquired angioedema had no FDA-approved treatment a decade ago and a single specific therapy, androgens; however, there has been a remarkable increase in both molecular diagnosis and therapy of these conditions as been reviewed extensively elsewhere.⁵ Remarkably, in the case of hereditary angioedema, response among patients is variable with some having almost complete cessation of attack with addition of prophylactic administration of replacement C1 inhibitor protein, whereas others require a second rescue medication for treatment. Obviously, as in the case of chronic urticaria, in hereditary angioedema it would also be helpful to have a diagnostic schema including previous response to therapy to facilitate care among multiple providers and to develop more effective prophylactic therapy.

In the acquired form of angioedema, there is normal to increased synthesis of normal C1 inhibitor protein and either an excessive consumption of C1 inhibitor protein or a formation of circulating anti-C1 inhibitor protein antibodies that cleaves or otherwise transforms C1 inhibitor protein into a nonfunctional form.¹³ In theory, acquired angioedema should not respond well to additional replacement C1 inhibitor proteins because the circulating abnormal proteins or immunoglobulins should inactivate the replacement protein rapidly; however, this has not been studied systematically.⁵ The acquired form of C1 inhibitor protein has been divided into types 1 and 2, a distinction that may influence therapy.¹³

Type 1 acquired angioedema is associated with lymphoproliferative disorders and thus require chemotherapy of the underlying malignant or premalignant condition and responds to therapy directed at the underlying disease, whereas type 2 has been associated with autoimmune syndromes. In patients with acquired angioedema related to autoimmune disease, a wide variety of therapies have been attempted including therapy with androgens also useful in hereditary angioedema, unsuccessful depletion of autoreactive antibodies through plasmapheresis, replacement of C1 INH, etanercept blocking inflammatory tumor necrosis factor, as well as agents that interfere with generation of bradykinin. Remarkably, patients with both types of acquired angioedema seem to respond to depletion of B-lymphocytes with chimeric antibodies such as rituximab.¹³ Rituximab, which depletes memory B-lymphocytes has been used successfully (and sometimes unsuccessfully) in both chronic urticaria and other autoimmune diseases such as rheumatoid arthritis, idiopathic thrombocytopenic purpura, idiopathic urticaria, autoimmune hemolytic anemia, systemic lupus erythematosus, multiple sclerosis, and acquired hemophilia.⁴⁶

DIFFERENTIAL DIAGNOSIS OF URTICARIA AND ANGIOEDEMA SHOULD INCLUDE INFORMATION FROM NOVEL PROTEOMIC TECHNOLOGY

In many cases, chronic urticaria is a disease primarily of the acquired immune system associated with generation of abnormal or pathologic immunoglobulins in conditions previously termed “idiopathic” or “spontaneous.”^{14,15,47–49} These autoreactive antibodies have been proposed to trigger mast cells and basophils by binding and cross-linking surface IgE leading to release of mediators responsible for urticaria and angioedema. An advance has been the clinical availability of assays for basophil activation using donor basophils exposed to serum from patients with chronic urticaria.⁴⁹ Basophil activation tests or autologous skin tests that suggest the presence of serum-mediated activation of mast cells may prove useful particularly if positive,

for example, to differentiate between angioedema mediated by complement pathways versus autoreactive antibodies as a guide for therapy.

However, basophil activation assays provide only a qualitative rather than quantitative answer and often are negative in the authors experience despite other evidence suggesting a component of an autoreactive immunoglobulin component, such as a comorbid autoimmune condition. Similarly, although in some cases of acquired angioedema a specific mechanism of complement depletion such as a premalignant lymphoma or complement autoreactive antibody can be detected, often these methods lack sensitivity, specificity and are not clinically available, except at a few specialized laboratories.¹³ As shown in Fig. 2, it is likely that the next clinical advance will include proteomic methods in development for autoimmune diseases that could also radically alter classification of urticaria and angioedema.⁵⁰

For example, the author has used proteomic technology for identification of novel sperm autoantigens in chronic urticaria after vasectomy (unpublished observations). After more than a year of daily urticaria after a vasectomy, a patient's serum was used to probe a sperm-specific protein library generated by gel electrophoresis. Using proteomic analysis of sperm proteins, 2 proteins were identified that could serve as triggering sperm antigens in this putative syndrome: (1) testis-specific protein 1 (NCBI accession # NP_003287) and (2) similar to the alpha subunit, or the macropain iota subunit of the proteasome multicatalytic endopeptidase complex (NCBI accession # CAA43964 and # AAH5552). Evidence of IgE autoantibodies to both proteins were present in addition to autoreactive IgG. Antithyroid microsomal and peroxidase antibodies and a positive basophil activation test were also present. A full discussion of this patient will be recounted elsewhere; however, the power of

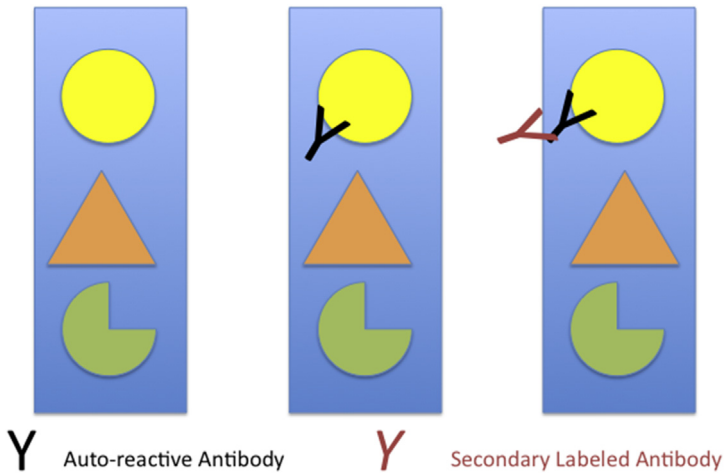


Fig. 2. Replacement of labor-intensive proteomic techniques such as gel electrophoresis with more advanced technology to identify autoantibodies is predicted to occur through arrays of antigen printed microchips. As shown, individual proteins or peptides (shown as 3 different shapes), are bound to a chip and incubated with patient and control serum. After washing, bound autoantibodies are detected with secondary antibody binding only to 1 of the 3 proteins (*top circle*). With miniaturization of this new technology, thousand of autoantigens and corresponding autoantibodies can be bound chronic urticaria to serum immunoglobulin, washed, and labeled to detect and characterize bound autoantibodies in mass production.

proteomics has recently been established independently in published peer-reviewed studies.^{15,33} For example, in chronic urticaria associated with thyroid autoimmunity, organ-specific IgE as well as IgG can be demonstrated. Whatever the mechanisms responsible for the generation of these abnormal disease-associated antibodies, it will be of great interest to see to what extent specific autoimmune IgE and IgG are correlated with disease prognosis and response to therapy.

DIFFERENTIAL DIAGNOSIS OF CHRONIC URTICARIA AND ANGIOEDEMA SHOULD INCLUDE MECHANISMS IDENTIFIED IN OTHER SYSTEMIC AUTOIMMUNE AND INFLAMMATORY DISEASES AND SYNDROMES

Viral proteins, like sperm proteins, frequently contain enzymatic activities such as proteases and nucleases. Also like sperm, viral proteins are foreign to the immune system developed in infancy through the acquired immune system. Thus, like sperm proteins that come in contact with the immune system after vasectomy, viral proteins from acute viral infection or reactivation may trigger some forms of chronic urticaria.^{34,46} Although some patients with chronic urticaria respond to antiviral therapy such as acyclovir, most do not, suggesting that once autoantibodies and autoreactive inflammatory processes are present, they may trigger pathology independent of active infection.

Extensive observations suggest that systemic lupus erythematosus and other systemic syndromes such as scleroderma causing epithelial disease are associated with abnormal response to chronic infection with some herpesviridae, specifically human herpes virus (HHV)-4 and HHV-6.^{46,51} In other autoimmune syndromes affecting the neuroepithelium such as multiple sclerosis, serology to HHV-4 antigens is

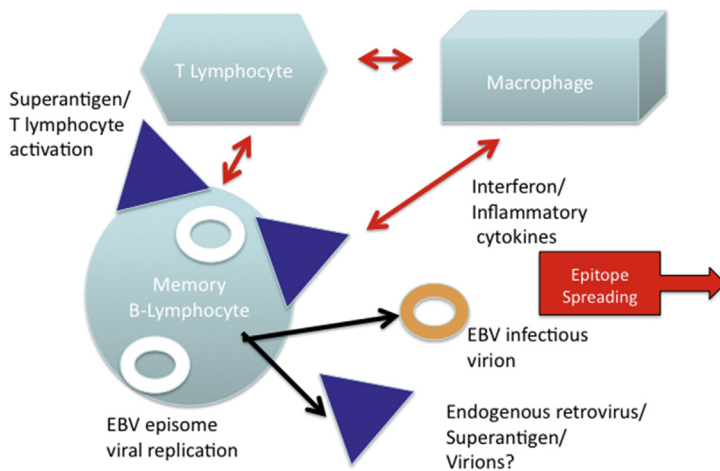


Fig. 3. Possible role of human endogenous retrovirus (HERV) in autoimmune disease. Both gamma herpes viruses such as human herpes virus (HHV)-4 (Epstein-Barr virus [EBV]) latent in memory B lymphocytes and HHV-6 in infected CD4 T-lymphocytes transactivate HERV superantigens that can both activate and suppress immune responses during viral reactivation as discussed in more detail in the text. Herpes reactivation may also reflect altered T helper cell type 2 cytokine balance in the host owing to hygiene, other illness, or factors; chronic urticaria thus could provide a useful means to study human autoimmune disease in situ in humans because a skin biopsy can be obtained much more readily than a brain biopsy in multiple sclerosis or other systemic autoimmune syndromes.

detected years or decades before presentation of clinical disease and may correlate with disease exacerbations.⁴⁶ Possibly, a common mechanism or mechanism may underlie autoimmune pathogenesis because beta and gamma herpesviruses are capable of triggering both systemic and localized inflammation through a variety of mechanisms including viral encoded cytokines (virokines), and viral encoded microRNA.¹⁸

As shown in **Fig. 3**, an intriguing possibility is that HHV-6 and HHV-4 viruses are particularly associated with autoimmune syndromes because they replicate in lymphocytes and activate endogenous superantigens expressed on lymphocytes encoded by extinct retroviruses or human endogenous retroviruses.⁴⁶ Herpes virus replication and gene expression are not independent of the host, but instead influenced by host T helper cell type 2 cytokine and other inflammatory cytokine expression, and viral gene expression in turn interacts with host T helper cell type 2 cytokine expression in epithelial tissues.⁴¹ As the prevalence of both T helper cell type 2-mediated atopic disease and also autoimmune disease increase in modern societies, it is

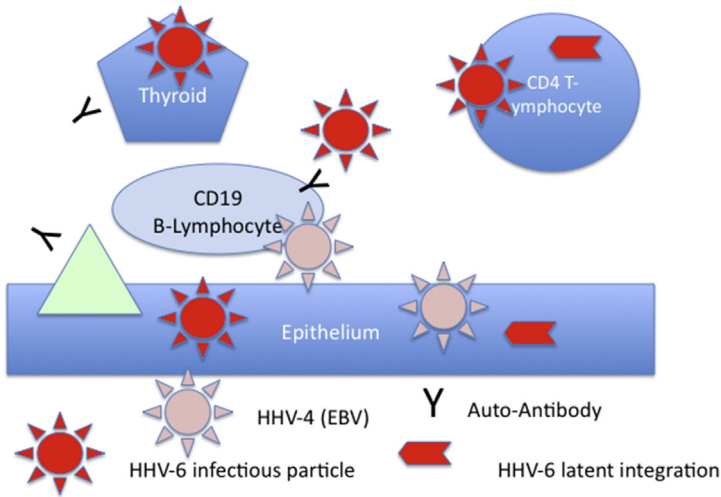


Fig. 4. The chronic viral urticaria (CVU) paradigm. In the CVU model, the initial triggering event in chronic urticaria (CU) is localized cutaneous inflammation owing to reactivation of beta and gamma herpes viruses that subsequently activate superantigens from endogenous human endogenous retroviruses (HERV). Perhaps many years after primary human herpes virus (HHV)-6 infection, reactivation of gene expression from HHV-6-infected CD4 lymphocytes and epithelial cells could trigger a chronic inflammatory state associated with HERV reactivation and clinically observed autoimmune pathology as well as depletion of mast cells in the skin and basophils in the serum. This inflammatory state would be sufficient to trigger urticaria analogous to the exanthem of primary herpes infection. Long-term cutaneous inflammation would also stimulate proliferation in situ of local autoreactive HHV-4 positive B lymphocyte clones. Many questions remain unresolved, for example, whether use of omalizumab is correlated with decreased viral reactivation of herpes and herpes and HERV gene expression in the skin. Related questions include whether level of elevation of initial HHV-6 and HHV-4 titers predict a clinical response to omalizumab and a response to anti-viral therapy, and to what extent mechanisms of chronic urticaria are shared with severe drug allergy syndromes such as drug reaction with eosinophilia and systemic symptoms and/or delayed hypersensitivity with systemic symptoms (see text).

probable that increased T helper cell type 2 bias in the skin would also predict an increased prevalence of chronic urticaria.¹⁸

Based on analysis of a response to antiviral therapy in some patients with chronic urticaria, the author recently presented a working model of urticaria pathogenesis in some cases as a syndrome termed chronic viral urticaria related to partial or complete reactivation of a beta or gamma herpes virus or viruses in both endocrine and cutaneous tissues^{34,46} (Fig. 4). An important and long-standing observation in chronic urticaria is an association with autoimmune thyroid disease that may also have HHV-6 as a cofactor.⁵² HHV-4 and HHV-6 reactivation also is implicated in severe cutaneous drug allergy syndromes termed drug reaction with eosinophilia and systemic symptoms and/or delayed hypersensitivity with systemic symptoms driven by increased gene expression of a chronic viral pathogen.³⁴ A common underlying etiology for the underlying inflammation in thyroid disease, drug reaction with eosinophilia and systemic symptoms and chronic urticaria could open a new window into definitive or curative therapies both in all of these autoimmune conditions.

Long-standing clinical observations have suggested that patients who report multiple drug and antibiotics allergy often have underlying chronic urticaria, supporting a common mechanism that might be a target for new therapy. A related and unresolved clinical question concerns whether all forms of chronic urticaria and angioedema, including physical urticaria syndromes, share some common inflammatory pathway because physical urticaria, although phenotypically diverse, also often responds to omalizumab. If in some forms of chronic urticaria the underlying defect is related to abnormal gene expression of reactivated herpes viruses and human endogenous retroviruses in cutaneous tissues, then monitoring of herpes and human endogenous retroviruses serology or related markers for viral infection could be useful in predicting response to both conventional and antiviral therapy. Similarly, proteomics, and messenger RNA or other gene profiling methods might be helpful in discriminating between patients with active cutaneous viral gene expression and replication and those with inflammation not related to active viral replication to predict response to therapy.

SUMMARY

New molecular and pharmacologic techniques and therapies have blurred many of the traditional categories of differential diagnosis and therapy particularly for chronic urticaria and angioedema. Rather than the impossible task of attempting to review all of these developments, instead the author has used selective examples from his own literature review and clinical experience to illustrate the limitations of phenotype based diagnosis and corresponding complexity of urticaria and angioedema diagnosis by molecular mechanism and response to therapy. Apparently, new genomic methods such as whole exome and whole genome sequencing both of germ line and somatic tissues, proteomics, and new targeted therapies will permit very detailed characterization of chronic urticaria and angioedema and this information must somehow be incorporated into new classification schemes.

The author has focused also on the role of inflammation in triggering chronic urticaria, and the author suggests there may be significant overlap between chronic urticaria and other autoimmune conditions detected by proteomic technology in the future. Chronic urticaria may also yield insights into acute urticaria, either IgE mediated or postviral, physical urticaria and mast cell activation syndromes, as well as severe drug allergy such as drug reaction with eosinophilia and systemic symptoms and/or delayed hypersensitivity with systemic symptoms, which may also share innate and

acquired immune response owing to chronic viral reactivation.^{34,46} Importantly, many or even most phenotypically distinct cases of familial urticaria and angioedema can now be characterized as the results of specific gene mutations, often in the innate and complement components of the immune system rather than resulting from defects in the acquired immune system (see **Box 1**). Because of these advances in molecular biology and pharmacology, the author suggests that poorly defined terms such as “idiopathic” and “spontaneous” do not provide any additional information and should be eliminated from future terminology.

Classification of response to therapy is also useful in differential diagnosis of urticaria although often surprisingly unpredictable (**Box 2**). With respect to the role of the acquired immune system, autoreactive antibodies are certainly present in many acquired chronic urticaria and angioedema phenotypes, but a causal role for these antibodies is more difficult to establish and may benefit in the future from more advanced combinations of proteomic and immunologic technologies (see **Fig. 2**). As shown in **Fig. 2**, the proteomic methods outlined also can be combined with immunologic methods into very powerful screens to detect hundreds or thousands of autoreactive antigens and antibodies.

As increasing data regarding specific autoantibody specificity emerges from novel technology, it will be interesting to what extent these observations can be integrated into diagnostic algorithms (see **Box 1**) and correlated with response to therapy (see **Box 2**). It is likely that correlations with other cutaneous autoimmune and inflammatory syndromes will also emerge from this data. In summary, many different phenotypic forms of chronic urticaria, particularly nonhereditary forms, may result from complex and chaotic interactions between the genome, pharmacologic therapy, and the microbiome and metagenome (see **Figs. 3** and **4**). Clinicians seeking accurate differential diagnosis of chronic urticaria and angioedema should continue to use phenotypic classification schemes as outlined in **Fig. 1** of this work, but also bear in mind Jonathan Swift (1667–1745) who wrote in his poem for children “Syphonaptera” more than 300 years ago shortly after the first microscopes revealed unexpected complexity of microscopic organisms:

*Big fleas have little fleas,
Upon their backs to bite 'em,
And little fleas have lesser fleas,
and so, ad infinitum.*

Box 2
Classification of chronic urticaria subtypes by response to therapy: an outline

Antihistamine responsive, unresponsive

Antiviral/antibiotic responsive, unresponsive

Corticosteroid responsive, unresponsive

Cyclosporine responsive, unresponsive

Omalizumab responsive, unresponsive

Other responsive, unresponsive

The author proposes that terms such as “idiopathic” and “spontaneous” should be eliminated and replaced by specific pharmacologic response data to the extent that this is known. This box is not intended to provide an exhaustive list of all possible reported therapy for chronic urticaria, but instead to provide a framework for more definitive future nomenclature.

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