

Inmaculada Pérez-Rangel, MD
 M. Angeles Gonzalo-Garijo, MD, PhD
 Remedios Pérez-Calderón, MD
 M. Angeles Zambonino, MD
 Silvia Irene Corrales-Vargas, MD
 Allergy Department
 Infanta Cristina University Hospital
 Badajoz, Spain
 magonzaloz@gmail.com

References

- [1] Wong GKY, Huissoon AP, Goddard S, Collins DM, Krishna MT. Wheat dependent exercise induced anaphylaxis: is this an appropriate terminology? *J Clin Pathol*. 2010;63:814–817.
- [2] Yacoub MR, Savi E, Burastero SE, et al. Omega-5 gliadin anaphylaxis: an integrated diagnostic approach. *Eur Ann Allergy Clin Immunol*. 2011;43:92–94.
- [3] Shadick NA, Liang MH, Partridge AJ, et al. The natural history of exercise-induced anaphylaxis: survey results from a 10-year follow-up study. *J Allergy Clin Immunol*. 1999;104:123–127.
- [4] Palosuo K, Varjonen E, Kekki O-M, et al. Wheat omega-5 gliadin is a major allergen in children with immediate allergy to ingested wheat. *J Allergy Clin Immunol*. 2001;108:634–638.
- [5] Jacquenet S, Morisset M, Battais F, et al. Interest of ImmunoCAP system to recombinant omega-5 gliadin for the diagnosis of exercise-induced wheat allergy. *Int Arch Allergy Immunol*. 2009;149:74–80.
- [6] Matsuo H, Dahlström J, Tanaka A, et al. Sensitivity and specificity of recombinant ω -5 gliadin-specific IgE measurement for the diagnosis of wheat-dependent exercise-induced anaphylaxis. *Allergy*. 2008;63:233–236.
- [7] Park H-J, Kim J-H, Kim J-E, et al. Diagnostic value of the serum-specific IgE ratio of omega-5 gliadin to wheat in adult patients with wheat-induced anaphylaxis. *Int Arch Allergy Immunol*. 2012;157:147–150.
- [8] Morita E, Chinuki Y, Takahashi H, Nabika T, Yamasaki M, Shiwaku K. Prevalence of wheat allergy in Japanese adults. *Allergol Int*. 2012;61:101–105.

Success with immunomodulatory therapies in the treatment of recalcitrant chronic urticaria

Estimates suggest that approximately 30% of patients with chronic urticaria (CU) are refractory to antihistamines,¹ a disease characteristic that, in our experience, frequently results in the use of corticosteroids to control acute flares and maintain remission. Reluctance may exist on the part of the patient or physician to use immunomodulatory medications as steroid-sparing agents in the treatment of this frustrating condition because of adverse side effects, need for laboratory monitoring, and uncertainty of outcome. We recently conducted a retrospective chart review to determine the relative effectiveness and safety of immunomodulatory therapies used in our tertiary clinic practice to treat antihistamine-refractory CU.

Under our clinic's standard operating procedure, if a patient did not achieve adequate control with at least twice the recommended daily dose of a second-generation antihistamine, full-dose H2 blocker, highest tolerated dose of an as needed first-generation antihistamine, and in most patients, a leukotriene receptor antagonist, they were deemed recalcitrant to antihistamine therapy, and options for noncorticosteroid second-line therapies were pursued. The initial choice for immunomodulator therapy was usually hydroxychloroquine or sulfasalazine for a 1- to 2-month trial. If the patient's condition failed to respond to either of these medications, consideration was given to mycophenolate mofetil, cyclosporine, and tacrolimus. Patients were asked to return for follow-up within 4 to 8 weeks after starting the medication to assess for level of clinical benefit. Based on their subjective level of improvement, each patient was assigned to 1 of 4 groups; complete resolution of symptoms, improved and did not require additional medications, improved but did require additional medications, and no improvement.

Forty-seven patients with antihistamine-refractory CU were treated in the Vanderbilt Asthma, Sinus and Allergy Program with 1 or more immunomodulatory therapies between 2003 and 2011. This represented 9% of the total CU population managed in our clinic. The average age was 44 years, and 69% were female. The average duration of symptoms at onset of immunomodulatory therapy was 20 months (range, 1.5–60 months). Also in this study, 42 of 47 (89%) patients were taking daily steroids or had received at least 2 prednisone tapers in the 6 months leading up to initiation of a steroid-sparing agent.

Based on our results, patients with CU can anticipate a greater than 50% chance that they will experience an improvement in their symptoms when treated with an immunomodulatory therapy.

Specifically, 88% of patients treated with cyclosporine experienced some clinical benefit, 75% with sulfasalazine, 60% with tacrolimus, 56% with hydroxychloroquine, and 55% with mycophenolate mofetil. Only 3 patients have been treated with omalizumab in our clinic, but all 3 (100%) have achieved complete resolution of symptoms despite being refractory to multiple other immunomodulators. The various levels of success for each medication are broken down in Figure 1. Adverse events that led to the discontinuation of medications during therapy included 2 patients with drug eruptions and one with a mucosal reaction while on hydroxychloroquine. One patient discontinued sulfasalazine related to gastrointestinal upset, and 1 patient stopped cyclosporine after developing hypertension. Four others had increased blood pressure while on cyclosporine, but this resolved with dose reductions.

An extensive literature search revealed limited existing data assessing the use of immunomodulatory medications in chronic urticaria.^{2–9} In our clinic, hydroxychloroquine was the most common second-line agent used, largely because it is relatively inexpensive and carries a low-risk side effect profile, requiring only a yearly dilated eye examination if long-term therapy is employed. Although mean time to remission was not able to be assessed in many patients examined in this study, a generally observed downside of hydroxychloroquine is its relatively longer duration to peak impact on symptoms, which is frequently in the magnitude of weeks, rather than days, such as when considering cyclosporine. Sulfasalazine resulted in the highest percentage (75%) of patients who were able to discontinue steroid use completely, with an identical percentage of patients achieving some clinical benefit. Mycophenolate mofetil had a lower than expected success rate when compared with prior studies,^{4,5} with only 55% of patients obtaining some clinical benefit, and only 36% achieving complete resolution. However, its use was generally reserved for the more refractory patients who had failed either hydroxychloroquine or sulfasalazine. Cyclosporine has worked well for patients in our clinic (88% improvement rate). All 5 patients who failed hydroxychloroquine and were subsequently placed on cyclosporine achieved subjective clinical benefit. Its use in our clinic has been limited by concerns for the potential for renal toxicity and strict monitoring requirements. Cyclosporine is the only immunomodulatory therapy that currently has placebo-controlled randomized prospective data to support its use in CU.^{7,8} Finally, similar success has been achieved in another smaller study looking at omalizumab,⁹ a drug that is being evaluated in a phase 3 clinical trial for treatment of CU.

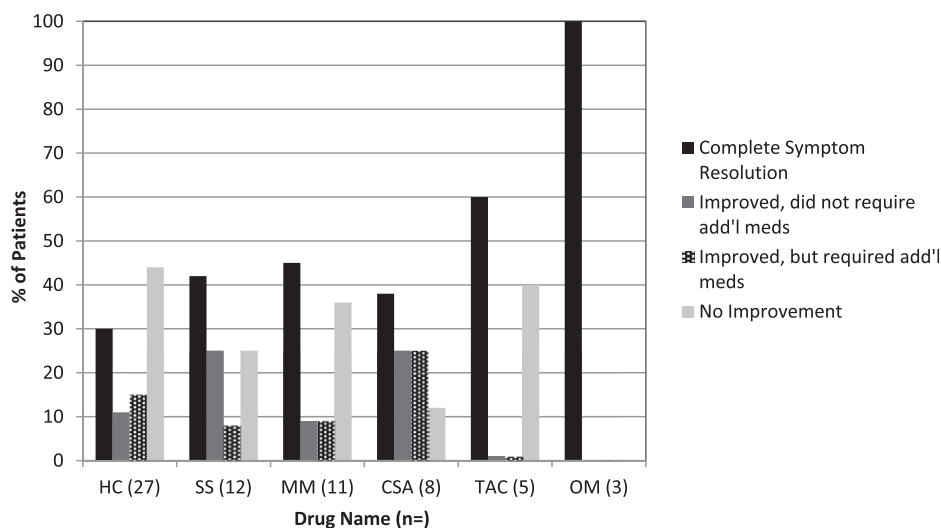


Figure 1. Subjective level of symptom improvement in patients with chronic urticaria, expressed as percentage of total patients treated with each immunomodulatory medication. HC, hydroxychloroquine; SS, sulfasalazine; MM, mycophenolate mofetil; CSA, cyclosporine; TAC, tacrolimus; OM, omalizumab.

Our review does provide evidence that each immunomodulatory medication has some value in the management of chronic urticaria. Methotrexate was not examined in this study because it is not employed for treatment of CU in our clinic, but it has been shown to be beneficial in the treatment of antihistamine-refractory urticaria.¹⁰ Randomized placebo-controlled trials with larger study groups, likely requiring a multicenter effort, are needed in evaluation of this common disease. Given a potential future indication for Omalizumab in CU, these studies will be necessary to validate the efficacy of less expensive immunomodulators.

Raymond M. Pongonis, Jr., MD

John M. Fahrenholz, MD

Division of Allergy, Pulmonary, and Critical Care Medicine

Vanderbilt University Medical Center

Nashville, Tennessee

pogie15@gmail.com

References

- [1] Najib U, Bajwa Z, Ostro M, Sheikh J. A retrospective review of clinical presentation, thyroid autoimmunity, laboratory characteristics, and therapies

used in patients with chronic idiopathic urticaria. *Ann Allergy Asthma Immunol.* 2009;103:496–501.

- [2] Reeves G, Boyle MJ, Bonfield J, Dobson P, Loewenthal M. Impact of hydroxychloroquine therapy on chronic urticaria: chronic autoimmune urticaria study and evaluation. *Int Med J.* 2004;34:182–186.
- [3] McGirt L, Vasagar K, Gober L, Saini SS, Beck LA. Successful treatment of recalcitrant chronic idiopathic urticaria with sulfasalazine. *Arch Dermatol.* 2006;142:1337–1342.
- [4] Zimmerman AB, Berger EM, Elmariah SB, Soter NA. The use of mycophenolate mofetil for the treatment of autoimmune and chronic idiopathic urticaria: experience in 19 patients. *J Am Acad Dermatol.* 2012;66:767–770.
- [5] Shahar E, Bergman R, Guttman-Yassky E, Pollack S. Treatment of severe chronic idiopathic urticaria with oral mycophenolate mofetil in patients not responding to antihistamines and/or corticosteroids. *Int J Dermatol.* 2006;45:1224–1227.
- [6] Vena G, Cassano N, Colombo D, Peruzzi E, Pigatto P. Cyclosporine in chronic idiopathic urticaria: a doubleblind, randomized, placebo-controlled trial. *J Am Acad Dermatol.* 2006;55:705–709.
- [7] Grattan C, O'Donnell B, Francis D, et al. Randomized doubleblind study of cyclosporine in chronic 'idiopathic' urticaria. *Br J Dermatol.* 2000;143:365–372.
- [8] Kessel A, Bamberger E, Toubi E. Tacrolimus in the treatment of severe chronic idiopathic urticaria: an open-label prospective study. *J Am Acad Dermatol.* 2005;52:145–148.
- [9] Kaplan AP, Joseph K, Maykut RJ, Geba GP, Zeldin RK. Treatment of chronic autoimmune urticaria with omalizumab. *J Allergy Clin Immunol.* 2008;122:569–573.
- [10] Sagi L, Solomon M, Baum S, Lyakhovitsky A, Trau H, Barzilai A. Evidence for methotrexate as a useful treatment for steroid dependent chronic urticaria. *Acta Dermatol Venereol.* 2011;91:303–306.

Glucoamylase is a potential allergen in products expressed in *Neurospora crassa*

Neurospora crassa, a well-characterized filamentous fungus, has been proposed as a vaccine production platform because of low cost, rapid expression, and molecular flexibility. Vaccine antigens are expressed and secreted by *N. crassa* in liquid culture, and the supernatant is then filtered and processed.¹ Because such vaccines are intended for human use, the presence of fungal allergens that might co-purify with vaccine antigens needs to be ascertained and minimized. Our goal was to screen *N. crassa* conditioned medium (CM) for the presence of such potential allergens. In the process we

identified the presence of *Neurospora* glucoamylase I precursor, a putative allergen of possible clinical significance.

Three-day *N. crassa* CM and growth media alone (containing Soytone, maltose, glucose, 2-(N-morpholino) ethanesulfonic acid buffer, Tween 80, and L-arginine) were used as antigen source and control. A highly complex (1.42×10^9) naïve human single-chain variable fragment (scFv) library was panned and screened against CM. Eighty specific clones were obtained after 3 rounds of biopanning and enrichment. Sequence analysis revealed 4 unique clones (NC3, NC14, NC65, and NC81) with greatest divergences in CDR2 of V_H and CDR3 of V_L chains (eFig 1A). Selected scFv antibodies were expressed and purified from *Escherichia coli* with hexahistidine at the N-termini and c-myc at the C-termini by nickel affinity chromatography (eFig 1B). All 4 scFv antibodies were reactive with a ~70 kDa band (eFig 1C).

Disclosures: Authors have nothing to disclose.

Funding Sources: This work was supported by intramural funding from Center for Biologics Evaluation and Research, FDA; and the Defense Advanced Research Projects Agency.