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Spotlight

Learning while treating: Gain-of-function STAT6 variants in severe allergic disease

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In an era of rapid identification of inborn errors of immunity, Sharma et al.[1] report novel heterozygous gain-of-function variants in the signal transducer and activator of transcription 6 (STAT6) gene in individuals with severe and early onset multi-systemic allergic disease.

Since the discovery of signal transducer and activator of transcription (STAT) in 1988 followed by Janus kinases (JAKs) in 1992,[2] the JAK-STAT pathways are now recognized as central to cell survival, proliferation, differentiation, and development, as well as immune responses and other essential biological functions. Aberrancies of JAK-STAT pathways underly a growing number of heterogeneous human disorders manifesting as impairment of normal immunity, and in some, type 2 immune dysregulation with allergic manifestations. For example, heterozygous loss-of-function (LOF) mutations in STAT3 were first identified as the cause of the autosomal dominant (AD) form of the hyper-immunoglobulin E (IgE) syndrome (HIES).[3] Subsequently, gain-of-function (GOF) mutations in JAK-STAT pathways leading to multiple inflammatory phenotypes have been discovered, including *STAT3*, *STAT5B*, and *JAK1*.[4–7]

Due to recognition of a heterogeneous group of single gene disorders characterized predominately by atopic and allergic disease manifestations, the term "primary atopic disorders (PADs)" has been widely adapted,[8]with more than 40 monogenetic disorders now discovered.[9] Key pathways found to be affected include T cell and B cell receptor signaling, cytokine signaling, and skin barrier and mast cell function.[8,9] In a recent issue of *Journal of Experimental Medicine*, Sharma et al.[1] studied 16 patients from 10 kindreds with severe early-onset allergic disease identified by expert clinicians, all of whom carried monoallelic rare variants in STAT6 (NM_001178079.2) identified by next-generation sequencing using either whole-exome or a targeted gene panel approach. Individuals represented diverse ethnicities inclusive of European, Hispanic, Middle Eastern, and Asian (South, East, and Southeast) ancestry. In 4 patients, *de novo* STAT6 mutations were found, and disease was fully penetrant in the families studied with all STAT6 variant carriers being affected. Based upon pathogenicity prediction models with combined annotation dependent depletion (CADD), sorting intolerant from tolerant (SIFT) and polymorphism phenotyping v2 (Polyphen-2) scores, described variants were all predicted to be highly pathogenic. None of the variants have previously been reported in population databases.

The interleukin 4R (IL-4R)-STAT6 axis plays a critical role in type-2 immunity, and its dysregulation plays a pathogenic role in different allergic diseases. STAT6 is critical in activating cytokine gene expression and cytokine signaling both in the immune cells and in airway epithelia, keratinocytes, and esophageal epithelial cells. STAT6 is activated by the type 2 cytokines IL-4 and IL-13 that are central to the pathogenesis of allergic disorders including asthma, atopic dermatitis, food allergy, and eosinophilic esophagitis (EoE). Through a series of eloquent experiments, transcriptomic/proteomic studies, and functional assays, Sharma et al.[1] demonstrate that the STAT6 germline monoallelic variants underlie severe allergic disease by a GOF mechanism. The variants led to sustained STAT6 phosphorylation due to delayed dephosphorylation, increased STAT6 target gene expression, and a TH2-skewed transcriptional profile.

In addition to this work, individual case reports have identified other *de novo* GOF variants of STAT6 found in children with very similar clinical features, including an initial report that described atopic disease in a father and his two children implicating a germline pathogenic variant.[10] Importantly, the recent work characterized a larger cohort including different families with both sporadic and affected individuals over multiple generations consistent with autosomal dominant (AD) inheritance. These findings highlight the severity of GOF mutations that should be suspected in individuals with early onset of severe allergic disease manifesting within the first year of life and those with refractory disease. Notable clinical features include patients presenting with treatment-resistant dermatitis, food allergies, followed by asthma, eosinophilic gastrointestinal disease and severe episodes of anaphylaxis. Highlighting more global immune dysregulation, half of the patients also presented with recurrent skin, respiratory, and viral infections, although none had history of fatal infections. Like features of HIES, short stature, skeletal issues such as pathologic fractures, and generalized hypermobility were also described. One patient had a history notable for B cell lymphoma, consistent with previous reports that have implicated activating somatic mutations in STAT6. Two separate fatalities due to anaphylaxis and a cerebral aneurysm also occurred. Other aspects of the clinical laboratory and immunological

work were largely unremarkable. T, B, and natural killer cell numbers were all typically in the normal range although clinical evidence of chronic systemic inflammation was documented (i.e., elevations in white blood cell counts, platelets, and serum immunoglobulin levels).

Finally, the authors translated their mechanistic findings into patient treatment with the use of select JAK inhibitors (ruxolitinib and tofacitinib) and the IL-4Rα monoclonal antibody dupilumab. These therapies were selected based upon *in vitro* activity demonstrating reductions in phosphorylated STAT6 in variant transfected HEK293 cells. Both drug classes led to the improvement in clinical outcomes in those who failed to respond to corticosteroids, topical tacrolimus, oral methotrexate, and anti-IL-5 monoclonal antibody. Using primary patient cells, the authors further showed that the JAK inhibitor tofacitinib promoted STAT6 dephosphorylation following IL-4 stimulation. Though the study was not blinded nor powered to determine treatment superiority, the mechanistic insights via GOF STAT6 variants led to dramatic improvements in the patients studied.

The authors justifiably propose that the described heterozygous GOF variants in STAT6 be added to the list of AD causes of the hyper-IgE phenotype and recommend genetic testing as the most efficient initial diagnostic approach for patients who experience severe allergic disease very early in life. The availability of genomic sequencing is becoming more accessible and, with commercially available gene panels, such diagnostic tools have heavily informed treatment strategy while continuing the search for reliable prognostic clinical biomarkers. This study also highlights the need for streamlined approaches to clinically correlate gene variants for which pathogenicity has not been described, possibly using pathogenicity prediction models along with functional assays. Although a portion of clinical immunologists have access to research laboratories with such resources, there remains unmet needs including lack of awareness of researchers willing to collaborate and expense of studies not covered by insurance. Because allergic disease phenotypes show a spectrum of severity, significant questions remain regarding who should be tested for STAT GOF variants, timing of testing, and how treatment choices might be affected by results. However, increased awareness of this work and similar reports will hopefully increase pediatric, primary care, and community allergy referrals to improve upon early diagnosis, initiation of effective treatment, and clinical outcomes.

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DECLARATION OF INTERESTS

The authors declare no competing interests.

REFERENCES

1. Sharma, M., Leung, D., Momenilandi, M., Jones, L.C.W., Pacillo, L., James, A.E., Murrell, J.R., Delafontaine, S., Maimaris, J., Vaseghi-Shanjani, M., et al. (2023). Human germline heterozygous gain-of-function STAT6 variants cause severe allergic disease. J. Exp. Med. *220*, e20221755. https://doi.org/10.1084/jem.20221755.
2. Seif, F., Khoshmirsafa, M., Aazami, H., Mohsenzadegan, M., Sedighi, G., and Bahar, M. (2017). The role of JAK-STAT signaling pathway and its regulators in the fate of T helper cells. Cell Commun. Signal. *15*, 23. https://doi.org/10.1186/s12964-017-0177-y.
3. Holland, S.M., DeLeo, F.R., Elloumi, H.Z., Hsu, A.P., Uzel, G., Brodsky, N., Freeman, A.F., Demidowich, A., Davis, J., Turner, M.L., et al. (2007). STAT3 mutations in the hyper-IgE syndrome. N. Engl. J. Med. *357*, 1608–1619. https://doi.org/10.1056/NEJMoa073687.
4. Del Bel, K.L., Ragotte, R.J., Saferali, A., Lee, S., Vercauteren, S.M., Mostafavi, S.A., Schreiber, R.A., Prendiville, J.S., Phang, M.S., Halparin, J., et al. (2017). JAK1 gain-of-function causes an autosomal dominant immune dysregulatory and hypereosinophilic syndrome. J. Allergy Clin. Immunol. *139*, 2016–2020.e5. https://doi.org/10.1016/j.jaci.2016.12.957.
5. Kasap, N., Aslan, K., Karakurt, L.T., Bozkurt, H., Canatan, H., Cavkaytar, O., Eken, A., and Arga, M. (2022). A novel gain-of-function mutation in STAT5B is associated with treatment-resistant severe atopic dermatitis. Clin. Exp. Allergy *52*, 907–910. https://doi.org/10.1111/cea.14148.
6. Milner, J.D., Vogel, T.P., Forbes, L., Ma, C.A., Stray-Pedersen, A., Niemela, J.E., Lyons, J.J., Engelhardt, K.R., Zhang, Y., Topcagic, N., et al. (2015). Early-onset lymphoproliferation and autoimmunity caused by germline STAT3 gain-of-function mutations. Blood *125*, 591–599. https://doi.org/10.1182/blood-2014-09-602763.
7. Liu, L., Okada, S., Kong, X.F., Kreins, A.Y., Cypowyj, S., Abhyankar, A., Toubiana, J., Itan, Y., Audry, M., Nitschke, P., et al. (2011). Gain-of-function human STAT1 mutations impair IL-17 immunity and underlie chronic mucocutaneous candidiasis. J. Exp. Med. *208*, 1635–1648. https://doi.org/10.1084/jem.20110958.
8. Lyons, J.J., and Milner, J.D. (2018). Primary atopic disorders. J. Exp. Med. *215*, 1009–1022. https://doi.org/10.1084/jem.20172306.
9. Vaseghi-Shanjani, M., Snow, A.L., Margolis, D.J., Latrous, M., Milner, J.D., Turvey, S.E., and Biggs, C.M. (2022). Atopy as immune dysregulation: Offender genes and targets. J. Allergy Clin. Immunol. Pract. *10*, 1737–1756. https://doi.org/10.1016/j.jaip.2022.04.001.
10. Suratannon, N., Ittiwut, C., Dik, W.A., Ittiwut, R., Meesilpavikkai, K., Israsena, N., Ingrungruanglert, P., Dalm, V.A.S.H., van Daele, P.L.A., Sanpavat, A., et al. (2023). A germline STAT6 gain-of-function variant is associated with early-onset allergies. J. Allergy Clin. Immunol. *151*, 565–571.e9. https://doi.org/10.1016/j.jaci.2022.09.028.