

Letter to the Editor

Invasive fungal disease in autosomal-dominant hyper-IgE syndrome

To the Editor:

Autosomal-dominant hyper-IgE (Job) syndrome (AD-HIES), a result of heterozygous dominant-negative mutations in signal transducer and activator of transcription 3 (*STAT3*), is characterized by staphylococcal skin abscesses, eczematous dermatitis, connective tissue defects, and elevated serum IgE.^{1,2} Recurrent bacterial pneumonias, attributed to dysfunctional *STAT3*, frequently lead to bronchiectasis and formation of pneumatoceles. Fungal pneumonias, particularly aspergillosis, have also been reported in patients diagnosed with AD-HIES, albeit by clinical scoring system, and are a cause of mortality.³ However, the few reports of invasive mycoses in patients with HIES predated the ability to confirm the patients' diagnoses genotypically, potentially admixing heterogeneous phenocopies and precluding comprehensive understanding of fungal disease in AD-HIES. We sought to define the epidemiology of invasive mycoses in patients with confirmed *STAT3*-mutant AD-HIES. Further, because *STAT3* is implicated in multiple hematopoietic and nonhematopoietic processes, and the former are critical to antifungal resistance in other at-risk groups, we evaluated myeloid function related to fungal susceptibility.

Medical records of all patients with the diagnosis of AD-HIES enrolled on protocols of the National Institute of Allergy and Infectious Diseases, National Institutes of Health, and confirmed by *STAT3* sequencing² were reviewed. Patients with clinical, computed tomography, and laboratory evidence of "proven" and "probable" invasive fungal disease, based on European Organization for Research and Treatment of Cancer/Mycoses Study Group (EORTC/MSG) consensus definitions,⁴ were identified.

Sixty-four patients with *STAT3* AD-HIES were identified: 23 (36%) had no computed tomography evidence of lung bronchiectasis/cysts/cavities/pneumatoceles, whereas 41 (64%) did. Of those without lung damage, none had concurrent or previous invasive mycoses. Among the 41 patients with parenchymal lung defects, 18 (44%) had at least 1 episode of invasive mold disease, typically involving the respiratory tract. Thus overall, 28.1% of all patients developed an invasive mycosis. The isolated molds were *Aspergillus* spp ($n = 16$; 83.3%), *Scedosporium apiospermum* ($n = 2$), and *Histoplasma capsulatum* ($n = 2$). One patient had aspergillosis and subsequently *S. apiospermum*; another had laryngeal histoplasmosis followed by aspergillosis. The other case of histoplasmosis manifested as a tongue ulcer with right middle lobe lung involvement necessitating lobectomy. Mold disease did not solely manifest as "fungus balls" restricted to lung cavities but also manifested as consolidations without halo sign (Fig 1); 5 patients had disseminated disease. Two cases caused by the yeast *Cryptococcus* sp presented as an esophageal mass⁵ and meningitis, respectively; both occurred in patients with abnormal lung.

Stratification of patients based on location of *STAT3* mutation (DNA-binding domain vs SH2 domain) was performed to identify any genotype-phenotype correlation in regard to the development of fungal infections (Table I). The occurrences of structural lung disease, fungal disease, median age of onset of first fungal pneumonia, and mortality caused by fungal infection were not

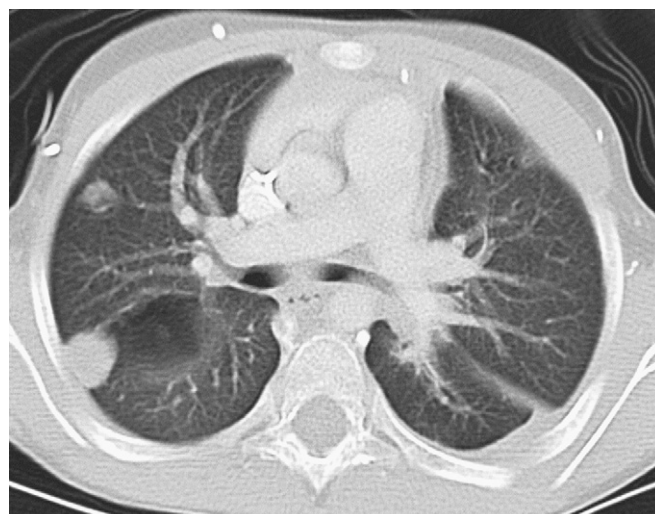


FIG 1. Computed tomography of invasive fungal pneumonia in AD-HIES.

TABLE I. Clinical characteristics of invasive fungal disease in patients with AD-HIES based on location of *STAT3* mutation

	Mutation in DNA-binding domain	Mutation in SH2-binding domain
No. of patients	34	30
Frequency of patients with structural lung disease	23 (67.6%)	18 (60%)
Frequency of patients with invasive fungal disease	10 (29.4%)*	8 (26.7%)*
Median age of onset of first fungal pneumonia (y)	32	30
Mortality as a result of invasive fungal disease	3	0

*All cases occurred in patients with structural lung disease.

statistically different between domains (Fisher exact test). Treatment was variable (systemic antifungal therapy, with/without intracavitary instillation of antifungals or surgical resection). Overall, the mortality rate among those who developed fungal disease was 16.7%.

The integrity of myeloid functions associated with susceptibility to invasive mycoses was investigated. Neutrophils, PBMCs, and plasma were isolated from patients (3 AD-HIES; 3 chronic granulomatous disease [CGD]) and healthy donors ($n = 4$), and *Aspergillus fumigatus* B-5233, a pulmonary isolate from a leukemia patient, was prepared as previously described.⁶ Patients' neutrophils were previously investigated for reactive oxygen species production via nitroblue tetrazolium histochemical staining, cytochrome c reduction, and dihydrorhodamine-123 flow cytometry; no deficiency was detected in the patients with AD-HIES. The capacities of neutrophils or PBMCs to inhibit growth of *Aspergillus* swollen conidia (the earliest metabolically active spore form) and of germlings (the earliest hyphal form) in autologous plasma were assessed by the modified 2,3-bis(2-methoxy-4-nitro-5-sulfophenyl)-5-[(phenylamino) carbonyl]-2 H-tetrazolium hydroxide

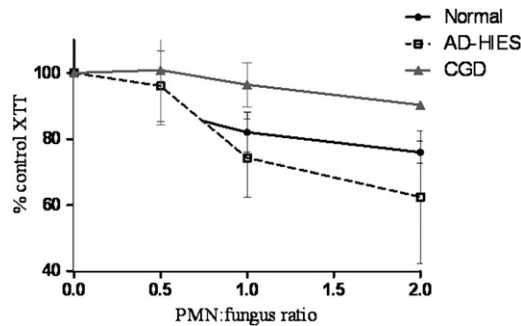


FIG 2. Growth inhibition of *A fumigatus* hyphae by neutrophils from patients with AD-HIES compared with normal donors and patients with CGD.

(XTT) method at leukocyte:fungus ratios of 0, 0.5, 1, and 2.⁶ This method has previously been used to demonstrate that neutrophils from patients with chronic granulomatous disease have impaired ability to control *A fumigatus* germings,⁶ recapitulating the *in vivo* phenomenon. Data analysis was performed by using GraphPad Prism software (v.5) (GraphPad Software, La Jolla, Calif).

Swollen conidia were inhibited equally well by neutrophils from controls and patients (data not shown). Neutrophils from patients with AD-HIES inhibited the growth of germings as effectively as normal donors, whereas neutrophils from patients with CGD were significantly less effective (Fig 2). PBMCs from controls and patients equally suppressed swollen conidia but mediated no significant inhibition of germings at ratios tested (data not shown).

Among primary immunodeficiencies, only 2 demonstrate consistent susceptibility to invasive aspergillosis: CGD and AD-HIES. The former results from mutations in the phagocyte reduced nicotinamide adenine dinucleotide phosphate (NADPH) oxidase complex, impairing their ability to generate reactive oxygen species. Consequently, patients with CGD demonstrate a high frequency of invasive aspergillosis (25% to 35%), whereby fungal disease typically occurs within the first 2 decades of life in otherwise normal lung. Patients with AD-HIES are also at risk for invasive mold infections (28% of this cohort), most commonly *Aspergillus*. In contrast with CGD, invasive fungal disease typically occurred in the fourth decade of life and only when there were concurrent anatomical lung defects from previous bacterial pneumonias. Despite therapy, mortality from invasive fungal disease was significant (~17%).

Late-onset invasive fungal disease in the setting of anatomical lung defects suggests that susceptibility to molds in AD-HIES is distinct from that in CGD. In agreement with this hypothesis, leukocytes from patients with AD-HIES performed as well as those from healthy donors in *in vitro* killing of *Aspergillus* conidia and hyphae and markedly better than CGD, suggesting that an intrinsic myeloid defect is not the prime reason these patients are susceptible to molds. Furthermore, chemotaxis of AD-HIES neutrophils toward *A fumigatus* was not different from controls (see this article's Videos E1 and E2 in the Online Repository at www.jacionline.org). Therefore, STAT3 mutations do not appear to impair the capacity of neutrophils or PBMCs to inhibit

Aspergillus in vitro. However, STAT3 may be operational in the myeloid control of intracellular fungi (eg, *Histoplasma*).

STAT3 enables the epithelium to respond to T_H17-based cytokines by upregulating antimicrobial peptides; defects in this pathway likely account for susceptibility to bacterial infections.⁷ STAT3 also has numerous roles in lung processes relevant to epithelial integrity after injury,^{8,9} such as bacterial infection; defects in these healing processes likely create a nidus for subsequent susceptibility to fungal pneumonias, similar to cystic fibrosis or posttuberculosis cavities.

AD-HIES confers a significant late-onset risk of invasive fungal disease despite preserved myeloid functions. This temporally dynamic susceptibility is associated with bronchiectasis/pneumatocoles, likely related to STAT3's role in lung epithelial homeostasis and defense.

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