

# Fatal Drug-Resistant Invasive Pulmonary *Aspergillus fumigatus* in a 56-Year-Old Immunosuppressed Man

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Immune status, severity or burden of disease, appropriate dosing of medication, and drug resistance are important considerations when treating immunosuppressed patients.

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**H**istorically, aspergillosis in patients with hematopoietic stem cell transplantation (HSCT) has carried a high mortality rate. However, recent data demonstrate a dramatic improvement in outcomes for patients with HSCT: 90-day survival increased from 22% before 2000 to 45% over the past 15 years.<sup>1</sup> Improved outcomes coincide with changes in transplant immunosuppression practices, use of cross-sectional imaging for early disease identification, galactomannan screening, and the development of novel treatment options.

Voriconazole is an azole drug that blocks the synthesis of ergosterol, a vital component of the cellular membrane of fungi. Voriconazole was approved in 2002 after a clinical trial demonstrated an improvement in 50% of patients with invasive aspergillosis in the voriconazole arm vs 30% in the amphotericin B arm at 12 weeks.<sup>2</sup> Amphotericin B is a polyene antifungal drug that binds with ergosterol, creating leaks in the cell membrane that lead to cellular demise. Voriconazole quickly became the first-line therapy for invasive aspergillosis and is recommended by both the Infectious Disease Society of American (IDSA) and the European Conference on Infections in Leukemia.<sup>3</sup>

## CASE PRESENTATION

A 55-year-old man with high-risk chronic myelogenous leukemia (CML) underwent a 10 of 10 human leukocyte antigen allele and antigen-matched peripheral blood allogeneic HSCT with a myeloablative-conditioning regimen of busulfan and cyclophosphamide, along with prophylactic voriconazole, sulfamethoxazole/trimethoprim, and acyclovir. After successful engraftment (without significant neutropenia),

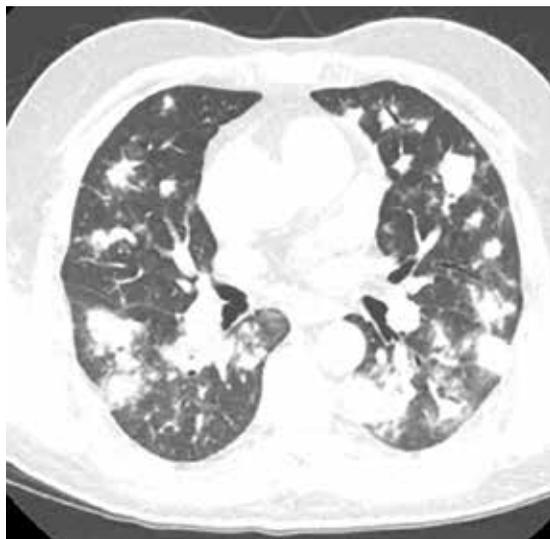
his posttransplant course was complicated by grade 2 graft vs host disease (GVHD) of the skin, eyes, and liver, which responded well to steroids and tacrolimus. Voriconazole was continued for 5 months until immunosuppression was minimized (tacrolimus 1 mg twice daily). Two months later, the patient's GVHD worsened, necessitating treatment at an outside hospital with high-dose prednisone (2 mg/kg/d) and cyclosporine (300 mg twice daily). Voriconazole prophylaxis was not reinitiated at that time.

One year later, at a routine follow-up appointment, the patient endorsed several weeks of malaise, weight loss, and nonproductive cough. The patient's immunosuppression recently had been reduced to 1 mg/kg/d of prednisone and 100 mg of cyclosporine twice daily. A chest X-ray demonstrated multiple pulmonary nodules; follow-up chest computed tomography (CT) confirmed multiple nodular infiltrates with surrounding ground-glass opacities suspicious with a fungal infection (Figure 1). Bronchoscopy with bronchoalveolar lavage (BAL) was significant for a positive *Aspergillus fumigatus* (*A fumigatus*) DNA polymerase chain reaction (PCR) assay and a BAL galactomannan level of > 5.3 optical density index (ODI) (normal, < 0.5). Bacterial and fungal cultures were negative, and serum galactomannan testing was not performed.

Treatment with oral voriconazole (300 mg twice daily) was initiated for probable pulmonary aspergillosis. Cyclosporine (150 mg twice daily) and prednisone (1 mg/kg/d) were continued throughout treatment out of concern for hepatic GVHD. The patient's symptoms improved over the next 10 days, and follow-up chest imaging demonstrated improvement.

Two weeks after initiation of voriconazole

**FIGURE 1**  
**Chest Computed Tomography**  
**Showing Multiple Pulmonary Nodules**

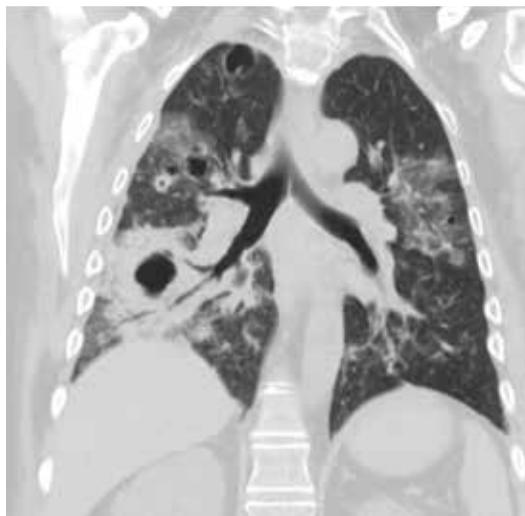


Surrounding ground-glass opacities ("halo sign") suggestive of multifocal fungal infection.

treatment, the patient developed a new productive cough and dyspnea, associated with fevers and chills. Repeat imaging revealed right lower-lobe pneumonia. The serum voriconazole trough level was checked and was 3.1 mg/L, suggesting therapeutic dosing. The patient subsequently developed acute respiratory distress syndrome and required intubation and mechanical ventilation. Repeat BAL sampling demonstrated multidrug-resistant *Escherichia coli*, a BAL galactomannan level of 2.0 ODI, and negative fungal cultures. The patient's hospital course was complicated by profound hypoxemia, requiring prone positioning and neuromuscular blockade. He was treated with meropenem and voriconazole. His immunosuppression was reduced, but he rapidly developed acute liver injury from hepatic GVHD that resolved after reinitiation of cyclosporine and prednisone at 0.75 mg/kg/d.

The patient improved over the next 3 weeks and was successfully extubated. Repeat chest CT imaging demonstrated numerous pneumatoceles in the location of previous nodules, consistent with healing necrotic fungal disease, and a new right lower-lobe cavitary mass (Figure 2). Two days after transferring out of the intensive care unit, the patient again developed hypoxemia and fevers to 39° C. Bronchoscopy with BAL of the right lower lobe revealed positive *A fumigatus* and *Rhizopus* sp polymerase chain reac-

**FIGURE 2**  
**Follow-up Chest Computed**  
**Tomography**



Multiple pneumatoceles in the location of previous nodules are consistent with healing necrotic fungal disease and a new right lower lobe cavitary mass.

tion (PCR) assays, although fungal cultures were positive only for *A fumigatus*. Liposomal amphotericin B (5 mg/kg) was added to voriconazole therapy to treat mucormycosis and to provide a second active agent against *A fumigatus*.

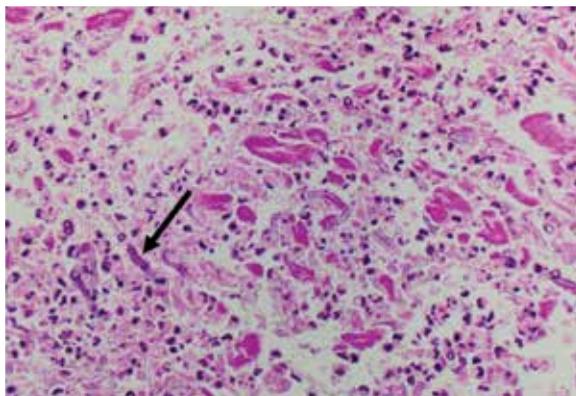
Unfortunately, the patient's clinical status continued to deteriorate with signs of progressive respiratory failure and infection despite empiric, broad-spectrum antibiotics and dual antifungal therapy. His serum voriconazole level continued to be therapeutic at 1.9 mg/L. The patient declined reintubation and invasive mechanical ventilation, and he ultimately transitioned to comfort measures and died with his family at the bedside.

Autopsy demonstrated widely disseminated *Aspergillus* infection as the cause of death, with evidence of myocardial, neural, and vascular invasion of *A fumigatus* (Figures 3 and 4). *Rhizopus* sp was identified in the large right lower lobe cavity without signs of angioinvasion, suggestive of cavity colonization. Follow-up sensitivity data (University of Texas, San Antonio, CLSI M38 A2, broth microdilution) of the *A fumigatus* demonstrated voriconazole sensitivity (MIC 0.25 µg/dL) but surprisingly, amphotericin B resistance (MIC > 2 µg/dL).

## DISCUSSION

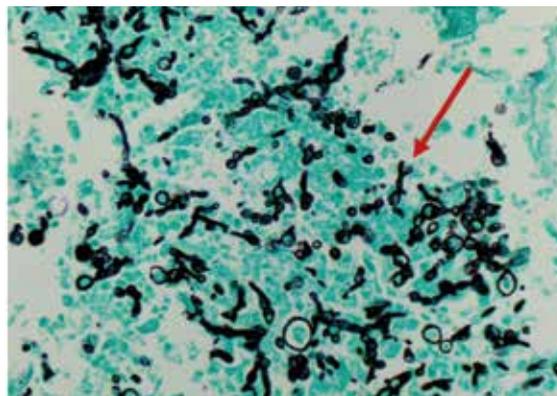
This case of fatal, progressive, invasive, pulmonary aspergillosis demonstrates several

**FIGURE 3**  
**Periodic Acid-Schiff Staining of Heart,**  
**x 40 Magnification**



Arrow indicates hyphae with acute angle branching consistent with aspergillosis.

**FIGURE 4**  
**Gomori Methenamine Silver Stain of**  
**Brain, x 40 Magnification**



Arrow indicates numerous hyphae with acute angle branching consistent with *Aspergillus fumigatus*.

important factors in the treatment of patients with this disease. Treatment failure usually relates to any of 4 possible factors: host immune status, severity or burden of disease, appropriate dosing of antifungal agents, and drug resistance. This patient's immune system was heavily suppressed for a prolonged period. Attempts at reducing immunosuppression to the minimal required dosage to prevent a GVHD flare were unsuccessful and became an unmodifiable risk factor, a major contributor to his demise.

The risks of continuous high-dose immunosuppression in steroid-refractory GVHD is well understood and has been previously demonstrated to have up to 50% 4-year nonrelapse mortality, mainly due to overwhelming bacterial, viral, and fungal infections.<sup>4</sup> All attempts should be made to cease or reduce immunosuppression in the setting of a severe infection, although this is sometimes impossible as in this case.

The patient's disease burden was significant as evidenced by the bilateral, multifocal pulmonary nodules seen on chest imaging and the disseminated disease found at postmortem examination. His initial improvement in symptoms with voriconazole and the evolution of his images (with many of his initial pulmonary nodules becoming pneumatoceles) suggested a temporary positive immune response. The authors believe that the *Rhizopus* in his sputum represents noninvasive colonization of one of his pneumatoceles, because postmortem examination failed to reveal *Rhizopus* at any other location.

Voriconazole has excellent pulmonary and central nervous system penetration: In this pa-

tient serum levels were well within the therapeutic range. His peculiar drug resistance pattern (sensitivity to azoles and resistance to amphotericin) is unusual. Azole resistance in leukemia and patients with HSCT is more common than is amphotericin resistance, with current estimates of azole resistance close to 5%, ranging between 1% and 30%.<sup>5,6</sup> Widespread use of antifungal prophylaxis with azoles likely selects for azole resistance.<sup>6</sup>

Despite this concern of azole resistance, current IDSA guidelines recommend against routine susceptibility testing of *Aspergillus* to azole therapy because of the current lack of consensus between the European Committee on Antibiotic Susceptibility Testing and Clinical and Laboratory Standards Institute on break points for resistance patterns.<sup>3,7</sup> This is an area of emerging research, and proposed cut points for declaration of resistance do exist in the literature even if not globally agreed on.<sup>8</sup>

Combination antifungal therapy is an option for treatment in cases of possible drug resistance. Nonetheless, a recent randomized, double-blind, placebo-controlled, multicenter trial comparing voriconazole monotherapy with the combination of voriconazole and anidulafungin failed to demonstrate an overall mortality benefit in the primary analysis, although secondary analysis showed a mortality benefit with combination therapy in patients at highest risk for death.<sup>9</sup>

Despite the lack of unified standards with susceptibility testing, it may be reasonable to perform such tests in patients with demonstrating progressive disease. In this patient's case,

amphotericin B was added to treat the *Rhizopus* species found in his sputum, and while not the combination studied in the previously mentioned study, the drug should have provided an additional active agent for *Aspergillus* should this patient have had azole resistance.

Surprisingly, subsequent testing demonstrated the *Aspergillus* species to be resistant to amphotericin B. De novo amphotericin B-resistant *A fumigatus* is extremely rare, with an expected incidence of 1% or less.<sup>10</sup> The authors believe the patient may have demonstrated induction of amphotericin-B resistance through activation of fungal stress pathways by prior treatment with voriconazole. This has been demonstrated in vitro and should be considered should combination salvage therapy be required for the treatment of a refractory *Aspergillus* infection especially if patients have received prior treatment with voriconazole.<sup>11</sup>

## CONCLUSION

This fatal case of invasive pulmonary aspergillosis illustrates the importance of considering the 4 main causes of treatment failure in an infection. Although the patient had a high burden of disease with a rare resistance pattern, he was treated with appropriate and well-dosed therapy. Ultimately, his unmodifiable immunosuppression was likely the driving factor leading to treatment failure and death. The indication for and number of bone marrow transplants continues to increase, thus exposure to and treatment of invasive fungal infections will increase accordingly. As such, providers should ensure that all causes of treatment failure are considered and addressed.

## AUTHOR DISCLOSURES

The authors report no actual or potential conflicts of interest with regard to this article.

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