Curr Fungal Infect Rep (2017) 11:148–157
DOI 10.1007/s12281-017-0278-5

FUNGAL GENOMICS AND PATHOGENESIS (S SHOHAM, SECTION EDITOR))

Pathogenesis of Invasive Pulmonary Aspergillosis in Transplant Recipients

Palash Samanta[1] · **M. Hong Nguyen**[1,2]

Published online: 27 June 2017

Abstract
Purpose of Review Transplant patients are at high risk for invasive pulmonary aspergillosis, and the associated mortality is high. The purpose of this study is to review the pathogenesis of invasive pulmonary aspergillosis (IPA) in transplant patients.
Recent Findings The pathogenesis of aspergillosis is multifactorial and results from a complex interplay between the pathogen and host. It is well recognized that *Aspergillus* causes IPA in immunocompromised patients. Recent studies have shown that *Aspergillus* might also cause diseases likely attributed to an unmodulated immune response in certain transplant recipients such as bronchopulmonary aspergillosis or bronchiolitis obliterans syndrome in lung transplant recipients.
Summary This review focuses on two crucial axes of the damage response framework applicable to aspergillosis: (1) Aspergillus virulence attributes that enable it to survive and proliferate in the host (thermotolerance, stress and hypoxic response, secretion of secondary metabolites) and (2) host response with specific focus on innate immunity and angiogenesis.

This article is part of the Topical Collection on *Fungal Genomics and Pathogenesis*

✉ M. Hong Nguyen
mhn5@pitt.edu

[1] University of Pittsburgh Medical Center, Pittsburgh, PA, USA

[2] University of Pittsburgh School of Medicine, Scaife Hall, Suite 871, 2335 Terrace Street, Pittsburgh, PA 15261, USA

Keywords Invasive pulmonary aspergillosis · *Aspergillus* pathogenesis · Damage response framework · Transplant recipients

Introduction

Aspergillus species are ubiquitous, saprophytic fungi which live in decaying vegetation and are found in water, soil, dust, and food. They produce small hydrophobic asexual spores or conidia that are dispersed easily in the air, and are able to survive broad range of harsh environmental conditions [1]. Although these conidia are frequently inhaled by humans in hundreds, the majority of the individuals do not typically develop diseases. Indeed, *Aspergillus* has been considered an "opportunistic pathogen" that does not possess classical virulence factors [2], and harms only immunocompromised host. However, it has increasingly been recognized that the clinical spectrum and severity of aspergillosis are determined by the degree of host damage resulting from the interaction between *Aspergillus* and the host [3]. On one hand, in patients with weak immune function, the damage of invasive aspergillosis (IA) is mediated by the pathogen, resulting from proliferation and invasion of fungal elements leading to host tissue destruction and infarction. On the other hand, in patients with strong immune response, the damage is primarily mediated by an unmodulated immune response, resulting in allergic sinus or bronchopulmonary aspergillosis [3].

Over the last few decades, the incidence of IA is on the rise due to exponential increase in the number of transplantation [4••]. In this review, we will discuss the pathogenesis, epidemiology, and clinical spectrum of pulmonary aspergillosis (IPA) among transplant recipients.

Pathogenesis

Out of nearly 200 known species of *Aspergillus*, approximately 20 species are known to cause human diseases. The primary route of infection with *Aspergillus* is via inhalation of airborne fungal spores called conidia, and their deposition in the lower respiratory tract. The vast majority of infections are caused by *Aspergillus fumigatus*, *Aspergillus flavus*, *Aspergillus niger*, and *Aspergillus terreus*, followed by other rare species such as *Aspergillus nidulans* and *Aspergillus calidoustus*. *A. fumigatus* is by far the most common cause of aspergillosis. Although the relative abundance of *A. fumigatus* conidia may partially explain for its success as pathogen, this is not universally true as there are several studies which found *A. fumigatus* to be less prevalent than other *Aspergillus* species in the environment [5–7]. In one study, *A. niger* and *A. fumigatus* accounted for 56 and 0.3% of air isolates, respectively, whereas *A. fumigatus* was the dominant isolate (44%) recovered from patients compared to *A. niger* (17%) [5]. It is likely that, by virtue of smaller size, the conidia of *A. fumigatus* (2.0–3.5 μm) and *A. terreus* (2.0–2.5 μm) can reach lower airways and alveoli more readily than larger conidia of *A. flavus* (3–6 μm) and *A. niger* (4–5 μm). Indeed, the conidial size might explain the fact that *A. fumigatus* and *A. terreus* mostly cause invasive pulmonary aspergillosis (IPA), whereas *A. flavus* and *A. niger* are mainly implicated in paranasal sinusitis and otitis [8–10]. Besides conidial size, several virulence attributes enable *A. fumigatus* to adapt, survive, and proliferate in the host. For example, *A. fumigatus* is more thermotolerant than other *Aspergillus* spp., and this ability to grow and germinate at higher temperatures correlates with its pathogenicity [11]. Higher temperature also induces stress response genes, which in turn confer additional survival benefits in *A. fumigatus.* Other putative virulence factors include cell wall composition and structure, nutritional scavenging mechanism, elaboration of extracellular proteases, and excretion of secondary metabolites [12].

The first defense mechanisms against inhaled *Aspergillus* spores are mucociliary apparatus and anatomical and mechanical barriers in the airways. Despite these measures, conidia of ≤2 μm in size are still able to reach terminal airways and alveoli, where they encounter epithelial lining comprising of respiratory epithelial cells (types I and II), alveolar macrophages, interstitial fibroblasts, and endothelial cells. Alveolar macrophages, the first line of innate immunity, phagocytose and kill the conidia. They also activate pro-inflammatory response that recruits neutrophils which in turn kill *Aspergillus* hyphae that escape the intracellular killing by alveolar macrophages. Epithelial cells secrete soluble antimicrobial peptides (AMP), defensins, lysozyme, and lactoferrin, that play a direct role in airway defense [13–16]. Corticosteroid therapy and high salt concentration in the airways of cystic fibrosis (CF) patients can eliminate the effect of the AMPs, favoring the colonization and subsequent invasion in the susceptible patients [16, 17]. Type II pneumocytes, in addition to phagocytosing and killing conidia, secrete surfactant proteins (SP-A, SP-D, and SP-C type lectins) to the alveolar space; these opsonins potentiate the phagocytic effect of alveolar macrophages and neutrophils [18–22]. Interestingly, a small fraction of internalized conidia not only survives within the type II pneumocytes but also inhibits the apoptosis of the host cells [23, 24]. The dual ability of *Aspergillus* conidia of evading the host's immune surveillance and at the same time maintaining the integrity of host cell not only is an important virulence trait of *Aspergillus* but also serves as a potential reservoir for it to cause invasive diseases in appropriate clinical setting.

Conidia germinate into germ tubes and hyphae when exposed to the favorable environment of the airway with optimum moisture and temperature. Germinated conidia and hyphae produce secondary metabolites and mycotoxins, e.g., gliotoxin, fumagillin, and helvolic acid, which damage the integrity of epithelial lining and mucociliary apparatus, and enhance hyphae penetration of epithelial cells and subsequently vascular endothelial cells [25–27]. Angioinvasion, the central feature of pathogenesis of IPA, is associated with endothelial injury, tissue factor expression and activation of platelets, and coagulation cascade [12, 28, 29•, 30]. Collectively, these processes impair vascular perfusion of *Aspergillus*-infected lung tissue, resulting in a necrotic core, where fungal hyphae proliferate abundantly, surrounded by peripheral zone of host immune response cells [31]. The adaptation to hypoxia, an important virulence trait, helps *Aspergillus* to survive in hypoxic environment [32].

Although both *Aspergillus* conidia and hyphae can induce endothelial cell endocytosis, the interaction between *Aspergillus* hyphae and endothelial cells is paramount for angioinvasion. Two different mechanisms of angioinvasion occur in IPA [33]. The most common mechanism is the local invasion of *Aspergillus* hyphae from the lungs into the alveolar-capillary barrier, then onto the blood vessels (abluminal penetration). The less common mechanism occurs in severely immunocompromised hosts, where the blood-circulating hyphal fragments lodge onto the peripheral capillary beds, penetrate the endothelial cell, and establish infection at a distant site (luminal penetration) [33]. In vitro study showed that luminal penetration by hyphae results in greater endothelial cell damage compare to abluminal penetration. On the other hand, abluminal invasion, as primarily happens in the lungs, is associated with greater induction of inflammatory response (such as cytokines, leukocyte adhesion molecules) and thrombosis (tissue factors) than luminal invasion. Altogether, the differences in the endothelial cell responses

to luminal versus abluminal infection may indicate significant differences in the pathogenesis of hematogenously disseminated versus locally invasive aspergillosis [33, 34].

Faced with tissue injury, the host cells trigger angiogenesis to facilitate tissue healing and regeneration via the release of vascular endothelial growth factor (VEGF), basic fibroblast growth factor (bFGF), and other growth factors [34]. In response, *A. fumigatus* secretes gliotoxin and other secondary metabolites to counteract the angiogenic activity, thus leading to further tissue hypoxia and necrosis, and limiting the penetration of immune cells and antifungal drugs into the site of infection [12, 28, 29•, 30]; this process results in abscess formation, which is the hallmark of late stage of IA. Interestingly, recent in vivo experiment showed that treatment with VEGF and fibroblast growth factor (FGF) improved survival of neutropenic mice with IPA, and both growth factors acted synergistically with the antifungal drug amphotericin B to decrease pulmonary fungal burden enhance survival [29•, 35•].

Epidemiology and Risk Factors for Aspergillosis in Specific Transplant Population

Over the past few decades, there has been significant change in the epidemiology of IA. This is mainly attributed to large number of immunosuppressed population including transplant recipients. The patients at risk for IA include those with leukemia and other hematologic disorders, hematopoietic stem cell transplantation (HSCT) recipients, solid organ transplant (SOT) recipients, neutropenic patients, and patients with immunosuppressive therapy for other immunologic conditions [36, 37]. Although there is significant advances in prophylactic antifungal regimens, early diagnosis, and newer antifungal medications, the mortality rate of IA remains as high as 50–60% in highest risk groups [36].

Hematologic Malignancy and HSCT Recipients

Invasive fungal infection occurs more frequently in patients with acute leukemia compared to chronic leukemia, lymphoma, and multiple myeloma [38]. *Aspergillus* accounts for 40–70% of all invasive fungal diseases in allogenic HSCT recipients [39••]. Recent studies showed that the 12-month cumulative incidence for IA among allogenic HSCT patients ranges from 1.6 to 3%, with overall 1-year survival of 25.4% [40–44]. In the last decades, the shortening of pre-engraftment neutropenia due largely to use of peripheral blood stem cells, growth factor, and nomyeloablative conditioning regimen decreased the incidence of early post-HSCT IA (≤40 days). However, increased age of transplant recipients, the use of alternate source of transplant (cord blood, T cell-depleted, or CD34-selected stem cell product), chronic graft versus host diseases (GVHD) and its prolonged treatment with corticosteroids, prolonged neutropenia, concomitant cytomegalovirus diseases, and respiratory virus infections have dramatically shifted the incidence of IA among HSCT recipients towards late-onset (41–180 days) or very late-onset (> 180 days) post-transplant [45–48]. The mortality rate in HSCT recipients with IA ranges from 66.6 to 80% and does not differ for those with early- versus late-onset post-transplant infections [49]. In HSCT, specific polymorphisms also contribute to host's inability to contain invasive diseases once *A. fumigatus* conidia gain entry to the lungs (Table 1) [50–52].

Solid Organ Transplant Recipients

The Transplant-Associated Infection Surveillance Network (TRANSNET) demonstrates that the overall 12-month cumulative incidence of IA among SOT recipients was 0.65%, which was most common among lung transplant recipients, followed by heart and liver transplant recipients [53, 54••, 55–57]. However, IA can occur 3 years or more post-transplant [58]. The specific risk factors for IA among specific organ transplant recipients are summarized in Table 1. The mortality rate of IA in SOT recipients ranges from 20 to 66% [59].

Lung Transplant Recipients

During lung harvest, the bronchial artery is disrupted, and the donor's bronchus has to rely on the collateral perfusion. Revascularization of the donor lung by the recipient's bronchial arteries may take several weeks. Thus, during early post-transplant, the bronchial anastomosis is devascularized leading to airway ischemia which in turn provides a fertile environment for saprophytic conidia to proliferate. For these reasons, it is not surprising that the rate of *Aspergillus* colonization is high in lung transplant recipients, ranging from 22 to 85%, and highest within 6 months of transplant [60•]. Colonization itself, irrespective of infection, increases patient mortality at 5 years [61]. Furthermore, colonization with small-sized spores of *Aspergillus* has recently been linked to bronchiolitis obliterans syndrome (BOS), a common complication after lung transplantation that leads to progressive allograft dysfunction [62•, 63]. The exact mechanism by which *Aspergillus* colonization leads to BOS is not known. It is speculated that *Aspergillus* spores, similar to other infectious agents previously linked to BOS, like *Cytomegalovirus* and respiratory viruses, trigger the host response that leads to innate immunity activation that ultimately induces BOS [64].

The overall incidence of IA among lung transplant recipients ranges from 4 to 23% [65], a rate as high as the reported rate among donor-mismatched allogeneic HSCT recipients [44, 53]. This emphasizes the highest risk status of IA among lung transplant recipients despite widespread use of antifungal prophylaxis. Lung transplant recipients possess unique risk

Table 1 Risk factors for invasive aspergillosis in transplant recipients

General risk factors
- Neutropenia
- Prolonged corticosteroid treatment
- Post-transplant rejection and augmented immunosuppressive therapy
- Older age of transplant recipients
- Renal dysfunction
- CMV infections
- Respiratory viral infections

Hematologic malignancies and HSCT recipients
- Hematological malignancy
  - Acute (compared to chronic) leukemia
  - Myelodysplasia
  - Aplastic anemia
- Myeloablative conditioning regimen
  - Alemtuzumab
- Transplant type
  - Allogenic (compared to autologous) transplant
  - Bone marrow (compared to peripheral stem cell) transplant
  - Cord blood transplant
  - T cell-depleted or CD34-selected stem cell product
  - HLA-mismatched transplant
- Graft versus host diseases and its treatment
  - Corticosteroids (>0.5 mg/kg/day)
  - Infliximab
- Polymorphisms
  - Toll-like receptor 4 polymorphism
  - Dectin-1 Y238X heterozygosity
  - Deficiency of soluble pattern-recognition receptor, pentraxin 3 (PTX3)
- Others
  - CMV disease

Lung transplant recipients
- Pre- and post-transplant *Aspergillus* colonization
- Single lung transplant
- Early ischemia at the anastomosis and airway stenosis
- Hypogammaglobulinemia (IgG < 400 mg/dl)
- Uncontrolled CMV infection
- Augmentation of immunosuppression

Heart transplant recipients
- Reoperation
- Isolation of *Aspergillus* from respiratory culture
- Post-transplant hemodialysis
- An episode of IA in the heart transplant institute 2 months pre- or post-transplant date

Liver transplant
- Retransplantation
- Transplantation for fulminant hepatic failure
- Renal replacement therapy
- Allograft dysfunction
- Recurrent HCV infection
- Human herpes virus 6 (HHV 6) infection

Renal transplant
- Allograft dysfunction
  - Requiring renal replacement therapy
- Delayed graft function
- Pre-transplant diagnosis of chronic obstructive pulmonary diseases (COPD)
- Occurrence of post-transplant pneumonia

factors that predispose them to IA. First, they typically remain on higher dose and longer duration of immunosuppression than other organ transplant. Second, the continuous exposure of the lungs to the environment and inhaled pathogens, coupled with blunted cough response and impaired ciliary function, contributes to susceptibility to IA. The overall mortality is 20%, ranging from 23 to 29% in patients with tracheobronchitis to as high as 67 to 82% in patients with IPA [49, 66].

Heart Transplant Recipients

Aspergillosis is the most commonly occurring invasive mycoses among heart transplant recipients, with incidence ranging from 1 to 14% [53, 67]. The data on IA among heart transplant recipients are relatively scarce. In a recent descriptive study of 479 consecutive heart transplant recipients from 1988 to 2011 in a single institution in Spain, the overall incidence of IA was 6.5%. Incidence decreased from 8.7% in the period 1988 to 2000 to 3.5% thereafter. The overall mortality was 61%, and the attributable mortality was 36%, with significant decrease from 46% in the historical cohort (1988–2000) to 0% in the present cohort. The proportion of early episodes (<90 days) in the historical cohort and present cohort was 71 and 86%, respectively [68]. Recovery of *Aspergillus* from respiratory tract cultures, particularly of *A. fumigatus*, is highly predictive of IA in heart transplant recipients [49, 69]. Other risk factors include reoperation, CMV diseases, post-transplant hemodialysis, and an episode of IA in the institutions' heart transplant program 2 months pre- or post-transplant date (Table 1) [49, 69].

Liver Transplant Recipients

IA has been reported in 1–9.2% of liver transplant recipients. Although IA in general occurs within the first month of transplant, the timing can range from a few weeks to years after [57, 70]. More recent data, however, documented later-onset IA (>90 days), which is largely due to advanced surgical technique, delayed onset of post-transplant risk factors of IA (e.g., CMV infection), and allograft rejection due to hepatitis C virus (HCV) infection [57]. The other risk factors for IA include retransplantation and renal replacement therapy which confer 30-fold and 15- to 25-fold higher risk of IA, respectively (Table 1) [49, 57, 71, 72]. The overall mortality rate of IA is 60 to 80%, particularly among those undergoing retransplantation within 30 days of primary transplant.

Renal Transplant Recipients

The risk of IA after renal transplant is relatively lower compared to that of other SOT recipients, with incidence rate <0.5% in most of the studies and ranging up to 4% [54••, 55, 73, 74]. Approximately 45% of IA cases were diagnosed within the first 6 months post-transplant. The 6- and 12-week survival rates were 68.8 and 60.7%, respectively, and 22.1% of survivors experienced graft loss.

Clinical Spectrum of Aspergillosis

The clinical manifestations and outcome of *Aspergillus* infection are governed by the host- and pathogen-mediated events. *Aspergillus* is categorized as a class 4 pathogen according to the damage response framework, meaning that the damage to the host can occur at the extremes of both weak and strong immune response (Table 2) [3]. Indeed, the spectrum of clinical diseases varies from asymptomatic colonization in immunocompetent or minimally immunosuppressed individuals to symptomatic and, at times, severe diseases, in individuals with depressed immune response (Table 2). In the other extreme of the immune system, an exuberant and dysregulated host response to *Aspergillus* antigens could result in hypersensitivity reactions in the form of allergic sinus and bronchopulmonary aspergillosis (ABPA).

Invasive Pulmonary Aspergillosis

Invasive pulmonary aspergillosis (IPA) is associated with a high mortality rate in immunocompromised patients, particularly those with hematologic malignancies, those with chemotherapy-induced prolonged and profound neutropenia, and those recipients of HSCT or SOT. However, neutropenic and HSCT patients are afflicted with different defects in host immune system than non-neutropenic SOT patients. Histopathology of IPA in neutropenic and HSCT patients is characterized by abundant *Aspergillus* hyphae invading blood vessels and tissue, thrombosis, coagulative necrosis, intra-alveolar hemorrhage, a scant mononuclear inflammatory infiltrate, and eventual dissemination [75, 76]. In contrast, in non-neutropenic non-HSCT patients receiving corticosteroids, IPA is characterized by few hyphal elements with conidia in various stages of germination, neutrophilic and monocytic infiltrates, areas of pneumonia and bronchiolitis, inflammatory necrosis, and scant intra-alveolar hemorrhage [75–78].

The histopathologic patterns of IPA correlate with the computed tomography (CT) findings in these cohorts of patients. In neutropenic or HSCT patients, the common CT findings of IPA are macronodules of >1 cm in diameter, which are present in ~94% of patients [79]. Nodules are characteristic of angioinvasion (Fig. 1a), a form of IPA that typically occurs in neutropenic or severely immunocompromised patients. Nodules may be surrounded by a halo sign (Fig. 1b) which

Table 2 Classification of clinical manifestations of aspergillosis according to damage response framework

Aspergillus colonization				
Host response	Severe immunosuppression	Non-neutropenic, chronic steroid administration +/− other immunosuppression	Normal immunity to mild immunosuppression , with broncho-pulmonary disorders[a]	Hyperactive immunity
Examples of clinical scenerios	Neutropenia Acute leukemia HSCT	Solid organ transplant	Cavitary lung disease Bronchiectasis Cystic fibrosis	- Asthma - Exposure to *Aspergillus*
Histopathology and host damage	- High fungal burden, angioinvasion, intra-alveolar hemorrhage - Severe tissue destruction	- Few hyphae and conidia, neutrophilic and monocytic infiltrates - Areas of pneumonia and bronchiolitis	Colonization	- Allergic - Chronic immune activation
Examples of clinical entities	- IPA - ITBA, pseudomembranous	- IPA - ATB, - ITBA, ulcerative	- Aspergilloma - Airway colonization	- ABPA - ? BOS
Treatment	Antifungal Surgical resection	Antifungal Surgical resection	None if asymptomatic	Steroid and antifungal

[a] Patients with pre-existing pulmonary cavities, cystic fibrosis, and other bronchopulmonary disorders like chronic obstructive pulmonary diseases and bronchiectasis are prone to *Aspergillus* colonization. However, these conditions alone are not sufficient to cause IPA, unless there is an epithelial damage or the patient receives immunosuppressive therapy

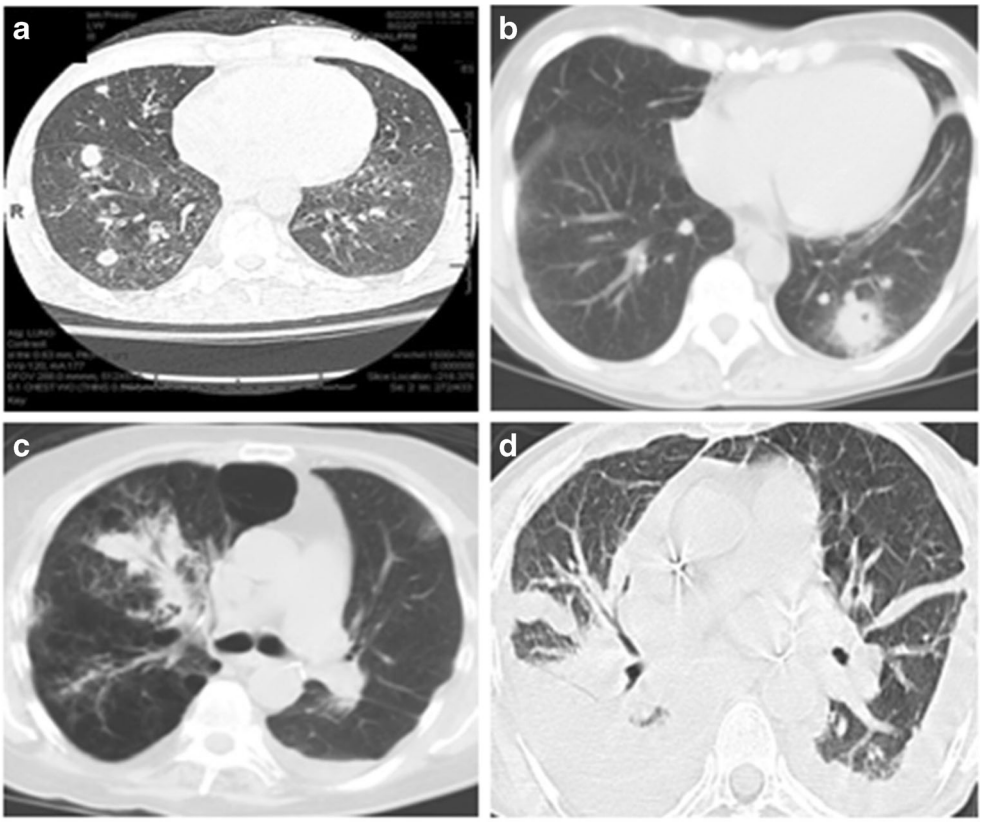

Fig. 1 CT scan manifestations of IPA among transplant recipients. The most common CT findings of IPA among the neutropenic patients and HSCT patients are macronodules (**a**). Nodules may be surrounded by a halo sign (**b**). Among SOT patients, the most common CT manifestations of IPA are macronodules (**a**) and peribronchial infiltrates or consolidations (**c**, **d**)

is an area of ground glass opacity of non-inflammatory alveolar edema or hemorrhage [80]. In severely immunocompromised hosts, the halo sign is highly suggestive of infection due to angioinvasive fungi. Among SOT patients, the most common CT manifestations of IPA are ground glass opacification, macronodules, peribronchial consolidation, and mass-like consolidation (Fig. 1c, d) [81]; halo and air-crescent signs are uncommon.

Invasive *Aspergillus* Tracheobronchitis

Invasive *Aspergillus* tracheobronchitis (IATB), an infrequent form of IA, is most common in lung transplant recipients, although it is seen in other immunocompromised patients (SOT and HSCT recipients and neutropenic patients) and rarely in immunocompetent patients [82, 83]. Based on bronchoscopic and pathologic appearance, three different forms of IATB have been described: *Aspergillus* tracheobronchitis (AT), ulcerative form, and pseudomembranous form [83]. AT is characterized by bronchial and/or tracheal inflammation and excessive mucus production without invasion of the bronchial mucosa on biopsy [83]. The ulcerative form is characterized by the presence of ulceration or plaque-like lesions in the bronchial walls (endobronchial aspergillosis) and most commonly associated with lung transplant recipients [83–85]. The pseudomembranous form is the most severe form of IATB and affects severely immunocompromised or neutropenic patients. They are characterized by pseudomembranes comprising of sloughing off necrotic epithelium and endobronchial mucous overlying the mucosal surface of the airways [83]. These three forms may overlap, or represent a progressive evolution of the disease. TBA may also progress to invasive and disseminated disease.

IATB affects 4–5% of lung transplant recipients, and the incidence is highest in the first year after transplantation [83, 85]. Virtually all cases were diagnosed within the first 3 to 6 months after transplantation with a medial interval of 2.7 months. Recently, IATB has been described to be associated with the use of belatacept, a CD-28 co-stimulation blocker, in patient with double lung transplant [86•]. TBA can be asymptomatic and detected only by surveillance bronchoscopy. Reported clinical findings have included fever, cough, wheezing, and hemoptysis. The overall outcome of IATB in lung transplant recipients is better than that of IPA, and treatment includes systemic and inhaled antifungal therapy along with bronchoscopic debridement, balloon dilatation, laser treatment, and/or stent placement.

Allergic Bronchopulmonary Aspergillosis

Allergic bronchopulmonary aspergillosis (ABPA) is characterized by type I hypersensitivity reaction to *Aspergillus*

antigens in particular individual with clinical symptoms of asthma or COPD [31]. ABPA has been reported to develop after lung transplant in two patients with cystic fibrosis [87, 88]. Recently, there have been several case reports describing a syndrome suggestive of ABPA following lung transplant [89, 90]. These patients presented with features of obstruction of the central bronchi, obstructive patterns on their pulmonary function tests, and subsequently *Aspergillus* recovery from mucus plugs. Although radiological changes characteristic of ABPA, such as consolidation, segmental or lobar atelectasis, and shadows from mucous impaction, were notably absent in most of the patients, some patients had elevated *Aspergillus*-specific IgE level and positive intradermal *Aspergillus* skin test [89, 90]. Immunosuppression in these patients may account for varying presentation [89]. All these patients responded to increased dose of corticosteroid and antifungal therapy.

Conclusion

Aspergillus is the second most common cause of invasive mycoses in transplant recipients. Despite recent advances in diagnostics and treatment modalities, the mortality of IA remains high. The interaction of host immunity and *Aspergillus* is complex and determines the clinical spectrum. Even among the immunocompromised population, the disease process is heterogeneous and depends on the deficiency of specific immune components of the host. Recent studies also enlighten the role of genetic components in the pathogenesis of IA. It is apparent that innate immunity has an important role in containment of infection. Furthermore, *Aspergillus* is able to develop mechanisms to evade host immune response. With advent of newer, potent immunosuppressive drugs, humanized monoclonal antibodies, and novel anticancer therapy, the population susceptible to IA is expanding. Thus, newer studies in pathogenesis of IA are of utmost importance. A major challenge in moving forward is how to harness the recent developments for newer therapeutic interventions.

Compliance with Ethical Standards

Conflict of Interest Palash Samanta declares no conflict of interest.
M. Hong Nguyen has received grants from Merck, Pfizer, and Astellas.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

References

Papers of particular interest, published recently, have been highlighted as:
• Of importance
•• Of major importance

1. Latge JP. The pathobiology of Aspergillus fumigatus. Trends Microbiol. 2001;9(8):382–9.
2. McCormick A, Loeffler J, Ebel F. Aspergillus fumigatus: contours of an opportunistic human pathogen. Cell Microbiol. 2010;12(11):1535–43.
3. Casadevall A, Pirofski LA. The damage-response framework of microbial pathogenesis. Nat rev Microbiol. 2003;1(1):17–24.
4.•• Patterson TF, Thompson GR 3rd, Denning DW, Fishman JA, Hadley S, Herbrecht R, et al. Practice guidelines for the diagnosis and management of aspergillosis: 2016 update by the Infectious Diseases Society of America. Clin Infect dis. 2016;63(4):e1–e60. **This is a current IDSA guideline on diagnosis and management of aspergillosis.**
5. Schmitt HJ, Blevins A, Sobeck K, Armstrong D. Aspergillus species from hospital air and from patients. Mycoses. 1990;33(11–12):539–41.
6. Hospenthal DR, Kwon-Chung KJ, Bennett JE. Concentrations of airborne Aspergillus compared to the incidence of invasive aspergillosis: lack of correlation. Med Mycol. 1998;36(3):165–8.
7. Panagopoulou P, Filioti J, Petrikkos G, Giakouppi P, Anatoliotaki M, Farmaki E, et al. Environmental surveillance of filamentous fungi in three tertiary care hospitals in Greece. J Hosp Infect. 2002;52(3):185–91.
8. Marr KA, Patterson T, Denning D. Aspergillosis. Pathogenesis, clinical manifestations, and therapy. Infect dis Clin N am. 2002;16(4):875–94. **vi**
9. Augustowska M, Dutkiewicz J. Variability of airborne microflora in a hospital ward within a period of one year. Ann Agric Environ med. 2006;13(1):99–106.
10. Xavier MO, Sales Mda P, Camargo Jde J, Pasqualotto AC, Severo LC. Aspergillus niger causing tracheobronchitis and invasive pulmonary aspergillosis in a lung transplant recipient: case report. Rev Soc Bras med Trop. 2008;41(2):200–1.
11. Dagenais TR, Keller NP. Pathogenesis of Aspergillus fumigatus in invasive aspergillosis. Clin Microbiol rev. 2009;22(3):447–65.
12. Ben-Ami R, Lewis RE, Kontoyiannis DP. Enemy of the (immunosuppressed) state: an update on the pathogenesis of Aspergillus fumigatus infection. Br J Haematol. 2010;150(4):406–17.
13. Alekseeva L, Huet D, Femenia F, Mouyna I, Abdelouahab M, Cagna A, et al. Inducible expression of beta defensins by human respiratory epithelial cells exposed to Aspergillus fumigatus organisms. BMC Microbiol. 2009;9:33.
14. Agerberth B, Grunewald J, Castanos-Velez E, Olsson B, Jornvall H, Wigzell H, et al. Antibacterial components in bronchoalveolar lavage fluid from healthy individuals and sarcoidosis patients. Am J Respir Crit Care med. 1999;160(1):283–90.
15. Schnapp D, Harris A. Antibacterial peptides in bronchoalveolar lavage fluid. Am J Respir Cell Mol Biol. 1998;19(3):352–6.
16. Travis SM, Conway BA, Zabner J, Smith JJ, Anderson NN, Singh PK, et al. Activity of abundant antimicrobials of the human airway. Am J Respir Cell Mol Biol. 1999;20(5):872–9.
17. Bellanger AP, Millon L, Khoufache K, Rivollet D, Bieche I, Laurendeau I, et al. Aspergillus fumigatus germ tube growth and not conidia ingestion induces expression of inflammatory mediator genes in the human lung epithelial cell line A549. J med Microbiol. 2009;58(Pt 2):174–9.

18. Madan T, Eggleton P, Kishore U, Strong P, Aggrawal SS, Sarma PU, et al. Binding of pulmonary surfactant proteins A and D to Aspergillus fumigatus conidia enhances phagocytosis and killing by human neutrophils and alveolar macrophages. Infect Immun. 1997;65(8):3171–9.
19. Madan T, Kishore U, Singh M, Strong P, Hussain EM, Reid KB, et al. Protective role of lung surfactant protein D in a murine model of invasive pulmonary aspergillosis. Infect Immun. 2001;69(4): 2728–31.
20. Madan T, Reid KB, Clark H, Singh M, Nayak A, Sarma PU, et al. Susceptibility of mice genetically deficient in SP-A or SP-D gene to invasive pulmonary aspergillosis. Mol Immunol. 2010;47(10): 1923–30.
21. Wright JR. Immunoregulatory functions of surfactant proteins. Nat rev Immunol. 2005;5(1):58–68.
22. Singh M, Madan T, Waters P, Sonar S, Singh SK, Kamran MF, et al. Therapeutic effects of recombinant forms of full-length and truncated human surfactant protein D in a murine model of invasive pulmonary aspergillosis. Mol Immunol. 2009;46(11–12):2363–9.
23. Wasylnka JA, Moore MM. Aspergillus fumigatus conidia survive and germinate in acidic organelles of A549 epithelial cells. J Cell Sci. 2003;116(Pt 8):1579–87.
24. Berkova N, Lair-Fulleringer S, Femenia F, Huet D, Wagner MC, Gorna K, et al. Aspergillus fumigatus conidia inhibit tumour necrosis factor- or staurosporine-induced apoptosis in epithelial cells. Int Immunol. 2006;18(1):139–50.
25. Tomee JF, Wierenga AT, Hiemstra PS, Kauffman HK. Proteases from Aspergillus fumigatus induce release of proinflammatory cytokines and cell detachment in airway epithelial cell lines. J Infect dis. 1997;176(1):300–3.
26. Amitani R, Murayama T, Nawada R, Lee WJ, Niimi A, Suzuki K, et al. Aspergillus culture filtrates and sputum sols from patients with pulmonary aspergillosis cause damage to human respiratory ciliated epithelium in vitro. Eur Respir J. 1995;8(10):1681–7.
27. Amitani R, Taylor G, Elezis EN, Llewellyn-Jones C, Mitchell J, Kuze F, et al. Purification and characterization of factors produced by Aspergillus fumigatus which affect human ciliated respiratory epithelium. Infect Immun. 1995;63(9):3266–71.
28. Ben-Ami R. Angiogenesis at the mold-host interface: a potential key to understanding and treating invasive aspergillosis. Future Microbiol. 2013;8(11):1453–62.
29.• Ben-Ami R, Albert ND, Lewis RE, Kontoyiannis DP. Proangiogenic growth factors potentiate in situ angiogenesis and enhance antifungal drug activity in murine invasive aspergillosis. J Infect dis. 2013;207(7):1066–74. **This murine study suggests that vasculogenic pathways are unexplored targets for the treatment of invasive pulmonary aspergillosis.**
30. Ben-Ami R, Lewis RE, Leventakos K, Kontoyiannis DP. Aspergillus fumigatus inhibits angiogenesis through the production of gliotoxin and other secondary metabolites. Blood. 2009;114(26): 5393–9.
31. Shibuya K, Ando T, Hasegawa C, Wakayama M, Hamatani S, Hatori T, et al. Pathophysiology of pulmonary aspergillosis. J Infect Chemother. 2004;10(3):138–45.
32. Willger SD, Puttikamonkul S, Kim KH, Burritt JB, Grahl N, Metzler LJ, et al. A sterol-regulatory element binding protein is required for cell polarity, hypoxia adaptation, azole drug resistance, and virulence in Aspergillus fumigatus. PLoS Pathog. 2008;4(11):e1000200.
33. Kamai Y, Chiang LY, Lopes Bezerra LM, Doedt T, Lossinsky AS, Sheppard DC, et al. Interactions of Aspergillus fumigatus with vascular endothelial cells. Med Mycol. 2006;44(Suppl 1):S115–7.
34. Kontoyiannis RB-AaDP. Aspergillosis: from diagnosis to prevention. Editors: Comarú Pasqualotto, Alessandro (Ed) 2009:pp 345–79. Springer.
35.• Osherov N, Ben-Ami R. Modulation of host angiogenesis as a microbial survival strategy and therapeutic target. PLoS Pathog. 2016;12(4):e1005479. **This review summarizes the emerging roles of vasculopathy and angiogenesis in the pathogenesis of invasive aspergillosis.**
36. Gregg KS, Kauffman CA. Invasive aspergillosis: epidemiology, clinical aspects, and treatment. Semin Respir Crit Care med. 2015;36(5):662–72.
37. Samanta P, Singh N. Complications of invasive mycoses in organ transplant recipients. Expert review of anti-infective therapy. 2016.
38. Segal BH, Bow EJ, Menichetti F. Fungal infections in nontransplant patients with hematologic malignancies. Infect dis Clin N am. 2002;16(4):935–64. **vii**
39.•• Papadopoulou A, Kaloyannidis P, Yannaki E, Cruz CR. Adoptive transfer of Aspergillus-specific T cells as a novel anti-fungal therapy for hematopoietic stem cell transplant recipients: progress and challenges. Crit rev Oncol Hematol. 2016;98:62–72. **This review highlights the recent advances and the critical issues involved in development and protocol design of Aspergillus-specific T cell immunotherapy.**
40. Mikulska M, Raiola AM, Bruno B, Furfaro E, Van Lint MT, Bregante S, et al. Risk factors for invasive aspergillosis and related mortality in recipients of allogeneic SCT from alternative donors: an analysis of 306 patients. Bone Marrow Transplant. 2009;44(6): 361–70.
41. Neofytos D, Horn D, Anaissie E, Steinbach W, Olyaei A, Fishman J, et al. Epidemiology and outcome of invasive fungal infection in adult hematopoietic stem cell transplant recipients: analysis of Multicenter Prospective Antifungal Therapy (PATH) Alliance registry. Clin Infect dis. 2009;48(3):265–73.
42. Neofytos D, Treadway S, Ostrander D, Alonso CD, Dierberg KL, Nussenblatt V, et al. Epidemiology, outcomes, and mortality predictors of invasive mold infections among transplant recipients: a 10-year, single-center experience. Transpl Infect dis. 2013;15(3):233–42.
43. Nucci M, Garnica M, Gloria AB, Lehugeur DS, Dias VC, Palma LC, et al. Invasive fungal diseases in haematopoietic cell transplant recipients and in patients with acute myeloid leukaemia or myelodysplasia in Brazil. Clin Microbiol Infect. 2013;19(8):745–51.
44. Kontoyiannis DP, Marr KA, Park BJ, Alexander BD, Anaissie EJ, Walsh TJ, et al. Prospective surveillance for invasive fungal infections in hematopoietic stem cell transplant recipients, 2001–2006: overview of the Transplant-Associated Infection Surveillance Network (TRANSNET) Database. Clin Infect dis. 2010;50(8):1091–100.
45. Marr KA, Carter RA, Boeckh M, Martin P, Corey L. Invasive aspergillosis in allogeneic stem cell transplant recipients: changes in epidemiology and risk factors. Blood. 2002;100(13):4358–66.
46. Marr KA, Carter RA, Crippa F, Wald A, Corey L. Epidemiology and outcome of mould infections in hematopoietic stem cell transplant recipients. Clin Infect dis. 2002;34(7):909–17.
47. Passweg JR, Baldomero H, Bader P, Bonini C, Cesaro S, Dreger P, et al. Hematopoietic SCT in Europe 2013: recent trends in the use of alternative donors showing more haploidentical donors but fewer cord blood transplants. Bone Marrow Transplant. 2015;50(4):476–82.
48. Bensinger WI, Deeg HJ. Blood or marrow? Lancet. 2000;355(9211):1199–200.
49. Singh N, Paterson DL. Aspergillus infections in transplant recipients. Clin Microbiol rev. 2005;18(1):44–69.
50. Cunha C, Aversa F, Lacerda JF, Busca A, Kurzai O, Grube M, et al. Genetic PTX3 deficiency and aspergillosis in stem-cell transplantation. N Engl J med. 2014;370(5):421–32.
51. Bochud PY, Chien JW, Marr KA, Leisenring WM, Upton A, Janer M, et al. Toll-like receptor 4 polymorphisms and aspergillosis in stem-cell transplantation. N Engl J med. 2008;359(17):1766–77.

52. Chai LY, de Boer MG, van der Velden WJ, Plantinga TS, van Spriel AB, Jacobs C, et al. The Y238X stop codon polymorphism in the human beta-glucan receptor dectin-1 and susceptibility to invasive aspergillosis. J Infect dis. 2011;203(5):736–43.
53. Pappas PG, Alexander BD, Andes DR, Hadley S, Kauffman CA, Freifeld A, et al. Invasive fungal infections among organ transplant recipients: results of the Transplant-Associated Infection Surveillance Network (TRANSNET). Clin Infect dis. 2010;50(8):1101–11.
54.•• Lopez-Medrano F, Fernandez-Ruiz M, Silva JT, Carver PL, van Delden C, Merino E, et al. Clinical presentation and determinants of mortality of invasive pulmonary aspergillosis in kidney transplant recipients: a multinational cohort study. Am J Transplant. 2016. **This multicenter study shows that the combination antifungal therapy has no mortality benefit for kidney transplant patients with invasive aspergillosis.**
55. Lopez-Medrano F, Silva JT, Fernandez-Ruiz M, Carver PL, van Delden C, Merino E, et al. Risk factors associated with early invasive pulmonary aspergillosis in kidney transplant recipients: results from a multinational matched case-control study. Am J Transplant. 2016;16(7):2148–57.
56. Gavalda J, Len O, San Juan R, Aguado JM, Fortun J, Lumbreras C, et al. Risk factors for invasive aspergillosis in solid-organ transplant recipients: a case-control study. Clin Infect dis. 2005;41(1):52–9.
57. Singh N, Avery RK, Munoz P, Pruett TL, Alexander B, Jacobs R, et al. Trends in risk profiles for and mortality associated with invasive aspergillosis among liver transplant recipients. Clin Infect dis. 2003;36(1):46–52.
58. Lortholary O, Gangneux JP, Sitbon K, Lebeau B, de Monbrison F, Le Strat Y, et al. Epidemiological trends in invasive aspergillosis in France: the SAIF network (2005–2007). Clin Microbiol Infect. 2011;17(12):1882–9.
59. Morgan J, Wannemuehler KA, Marr KA, Hadley S, Kontoyiannis DP, Walsh TJ, et al. Incidence of invasive aspergillosis following hematopoietic stem cell and solid organ transplantation: interim results of a prospective multicenter surveillance program. Med Mycol. 2005;43(Suppl 1):S49–58.
60.• Bonnal C, Leleu C, Brugiere O, Chochillon C, Porcher R, Boelle PY, et al. Relationship between fungal colonisation of the respiratory tract in lung transplant recipients and fungal contamination of the hospital environment. PLoS One. 2015;10(12):e0144044. **This prospective study shows a significant association between fungal contamination of the patient's room and aspergillus respiratory colonization of lung transplant patients.**
61. Iversen M, Burton CM, Vand S, Skovfoged L, Carlsen J, Milman N, et al. Aspergillus infection in lung transplant patients: incidence and prognosis. Eur J Clin Microbiol Infect dis. 2007;26(12):879–86.
62.• Weigt SS, Copeland CA, Derhovanessian A, Shino MY, Davis WA, Snyder LD, et al. Colonization with small conidia Aspergillus species is associated with bronchiolitis obliterans syndrome: a two-center validation study. Am J Transplant. 2013;13(4):919–27. **This study shows that colonization of the lung allograft with small conidia Aspergillus species, but not large conidia species, is associated with an increased risk of bronchiolitis obliterans syndrome and suggests that prospective evaluation of strategies to prevent Aspergillus colonization of the airways is warranted.**
63. Weigt SS, Elashoff RM, Huang C, Ardehali A, Gregson AL, Kubak B, et al. Aspergillus colonization of the lung allograft is a risk factor for bronchiolitis obliterans syndrome. Am J Transplant. 2009;9(8):1903–11.
64. Garantziotis S, Palmer SM. An unwelcome guest: Aspergillus colonization in lung transplantation and its association with bronchiolitis obliterans syndrome. Am J Transplant. 2009;9(8):1705–6.
65. Husain S, Paterson DL, Studer S, Pilewski J, Crespo M, Zaldonis D, et al. Voriconazole prophylaxis in lung transplant recipients. Am J Transplant. 2006;6(12):3008–16.
66. Neofytos D, Fishman JA, Horn D, Anaissie E, Chang CH, Olyaei A, et al. Epidemiology and outcome of invasive fungal infections in solid organ transplant recipients. Transpl Infect dis. 2010;12(3):220–9.
67. Munoz P, Singh N, Bouza E. Treatment of solid organ transplant patients with invasive fungal infections: should a combination of antifungal drugs be used? Curr Opin Infect dis. 2006;19(4):365–70.
68. Munoz P, Ceron I, Valerio M, Palomo J, Villa A, Eworo A, et al. Invasive aspergillosis among heart transplant recipients: a 24-year perspective. J Heart Lung Transplant. 2014;33(3):278–88.
69. Munoz P, Alcala L, Sanchez Conde M, Palomo J, Yanez J, Pelaez T, et al. The isolation of Aspergillus fumigatus from respiratory tract specimens in heart transplant recipients is highly predictive of invasive aspergillosis. Transplantation. 2003;75(3):326–9.
70. Righi E, Ivaldi F, Carnelutti A, Tomkova L, Villa G, Sartor A, et al. Immunologic and pharmacologic aspects in an elderly recipient of liver transplant with pulmonary aspergillosis and multiple comorbidities. Exp Clin Transplant. 2016;14(5):567–70.
71. Singh N, Husain S, Practice ASTIDCo. Aspergillosis in solid organ transplantation. Am J Transplant. 2013;13(Suppl 4):228–41.
72. Fortun J, Martin-Davila P, Moreno S, De Vicente E, Nuno J, Candelas A, et al. Risk factors for invasive aspergillosis in liver transplant recipients. Liver Transpl. 2002;8(11):1065–70.
73. Brown RS Jr, Lake JR, Katzman BA, Ascher NL, Somberg KA, Emond JC, et al. Incidence and significance of Aspergillus cultures following liver and kidney transplantation. Transplantation. 1996;61(4):666–9.
74. Altiparmak MR, Apaydin S, Trablus S, Serdengecti K, Ataman R, Ozturk R, et al. Systemic fungal infections after renal transplantation. Scand J Infect dis. 2002;34(4):284–8.
75. Balloy V, Huerre M, Latge JP, Chignard M. Differences in patterns of infection and inflammation for corticosteroid treatment and chemotherapy in experimental invasive pulmonary aspergillosis. Infect Immun. 2005;73(1):494–503.
76. Stergiopoulou T, Meletiadis J, Roilides E, Kleiner DE, Schaufele R, Roden M, et al. Host-dependent patterns of tissue injury in invasive pulmonary aspergillosis. Am J Clin Pathol. 2007;127(3):349–55.
77. Berenguer J, Allende MC, Lee JW, Garrett K, Lyman C, Ali NM, et al. Pathogenesis of pulmonary aspergillosis. Granulocytopenia versus cyclosporine and methylprednisolone-induced immunosuppression. Am J Respir Crit Care med. 1995;152(3):1079–86.
78. Duong M, Ouellet N, Simard M, Bergeron Y, Olivier M, Bergeron MG. Kinetic study of host defense and inflammatory response to Aspergillus fumigatus in steroid-induced immunosuppressed mice. J Infect dis. 1998;178(5):1472–82.
79. Greene RE, Schlamm HT, Oestmann JW, Stark P, Durand C, Lortholary O, et al. Imaging findings in acute invasive pulmonary aspergillosis: clinical significance of the halo sign. Clin Infect dis. 2007;44(3):373–9.
80. Georgiadou SP, Sipsas NV, Marom EM, Kontoyiannis DP. The diagnostic value of halo and reversed halo signs for invasive mold infections in compromised hosts. Clin Infect dis. 2011;52(9):1144–55.
81. Park YS, Seo JB, Lee YK, Do KH, Lee JS, Song JW, et al. Radiological and clinical findings of pulmonary aspergillosis following solid organ transplant. Clin Radiol. 2008;63(6):673–80.
82. Gaspar BL, Agarwal R, Gupta K, Shivaprakash MR. Aspergillus pseudomembranous tracheobronchitis in an immunocompetent individual: a diagnostic conundrum with therapeutic challenge. Lung India. 2016;33(5):550–2.
83. Krenke R, Grabczak EM. Tracheobronchial manifestations of Aspergillus infections. ScientificWorldJournal. 2011;11:2310–29.

84. Geltner C, Lass-Florl C. Invasive pulmonary aspergillosis in organ transplants—focus on lung transplants. Respir Investig. 2016;54(2):76–84.
85. Fernandez-Ruiz M, Silva JT, San-Juan R, de Dios B, Garcia-Lujan R, Lopez-Medrano F, et al. Aspergillus tracheobronchitis: report of 8 cases and review of the literature. Medicine (Baltimore). 2012;91(5):261–73.
86.• Haidar G, Crespo M, Maximous S, Lendermon E, Clancy CJ, Nguyen MH. Invasive tracheobronchial aspergillosis in a lung transplant recipient receiving belatacept as salvage maintenance immunosuppression: a case report. Transplant Proc. 2016;48(1): 275–8. **This is an interesting case report describing belatacept as a potential risk factor for invasive aspergillosis.**
87. Fitzsimons EJ, Aris R, Patterson R. Recurrence of allergic bronchopulmonary aspergillosis in the posttransplant lungs of a cystic fibrosis patient. Chest. 1997;112(1):281–2.
88. Casey P, Garrett J, Eaton T. Allergic bronchopulmonary aspergillosis in a lung transplant patient successfully treated with nebulized amphotericin. J Heart Lung Transplant. 2002;21(11):1237–41.
89. Cerceo E, Kotloff RM, Hadjiliadis D, Ahya VN, Pochettino A, Gillespie C, et al. Central airways obstruction due to Aspergillus fumigatus after lung transplantation. J Heart Lung Transplant. 2009;28(5):515–9.
90. Verleden GM, Dupont LJ, Van Raemdonck DE. ABPA: does it really exist after lung transplantation? J Heart Lung Transplant. 2009;28(11):1239.