Organ transplant recipients are a growing and increasingly important group of immunocompromised hosts in whom invasive mycoses remain one of the most significant infectious complications.1–3 Although candida infections occur more frequently, the highest mortality rate associated with fungal infections in transplant recipients is for *Aspergillus* spp. infections.4 The last decade has witnessed the emergence of diverse mycelial fungi, including dematiaceous moulds, as significant pathogens in transplant patients.5 Trends in invasive fungal infections in the post-transplant setting have also been notable for a rise in non-*albicans* Candida spp.6 Given the high mortality associated with invasive mycoses in transplant recipients, effective prophylaxis for such infections is a worthy goal: an optimal approach has not, however, been devised. Uncertainty and controversy abound regarding the choice of antifungal agent, mode of drug delivery and types of patients who should receive antifungal prophylaxis. The lipid formulations of amphotericin B represent a significant advance in drug delivery of amphotericin B.7 Although the potential for reduced nephrotoxicity with lipid preparations of amphotericin B has been amply demonstrated, a frequent dilemma pertaining to the use of such agents is whether their high acquisition cost is justifiable in the transplant setting and whether there are data supportive of their superior efficacy as compared with amphotericin B deoxycholate. Finally, a growing body of evidence suggests a potential role for immunomodulatory agents in the treatment of invasive fungal infections.8–10 This review focuses on: (i) optimizing the approach to antifungal prophylaxis in organ transplant recipients; (ii) treatment of invasive mycoses; (iii) the role of adjunctive therapies, e.g. immunomodulation; and (iv) surgery in the management of invasive mycoses in organ transplant recipients.

### Antifungal prophylaxis

Itraconazole, being orally administered and highly active *in vitro* against *Aspergillus* and *Candida* spp., would appear to be an attractive prophylactic agent in transplant recipients. However, the erratic absorption and poor bioavailability of itraconazole capsules make it difficult to achieve adequate serum concentrations. Studies documenting the efficacy of itraconazole capsules as prophylaxis in transplant recipients have been largely uncontrolled, with the incidence of invasive aspergillosis in historical controls being far higher than would now be expected for the type of transplant.2,11,12

Itraconazole cyclodextrin preparation in oral solution has significantly improved bioavailability.13 To date, however, no randomized, controlled trials have demonstrated the efficacy of itraconazole cyclodextrin as prophylaxis against invasive aspergillosis in solid organ transplant recipients. In liver transplant recipients given an oral loading dose of itraconazole (5.0 mg/kg) before transplantation and 2.5 mg/kg following transplantation, fungal infections developed in 4% (1/24) of the patients as compared with 25% (9/37) of those who received placebo ($P = 0.049$).14 The study, although prospective, was insufficiently large to demonstrate the efficacy of prophylaxis against invasive aspergillosis; all fungal infections documented in the patients were due to *Candida*.14 A larger placebo-controlled, double-blind, multicentre European trial in neutropenic patients with haematological malignancies failed to show a reduction in the rate of aspergillus infections with itraconazole oral solution; systemic infections due to *Aspergillus* spp. developed in 2% (4/201) of the patients receiving itraconazole as compared with 0.5% (1/201) of those receiving placebo.15 Mycologically proven aspergillosis occurred in 1.3% (4/293) of the neutropenic...
episodes in a randomized trial in patients receiving 100 mg fluconazole suspension and 0% (0/288) in those receiving itraconazole solution.\textsuperscript{16} When two cases of invasive aspergillosis occurring in the fluconazole group outside the formal study period were included, the difference in the rate of aspergillus infections reached statistical significance. An intravenous cyclodextrin formulation of itraconazole is currently undergoing clinical trials. A concern with iv cyclodextrin is the potential for nephrotoxicity in patients with creatinine clearance $<30$ mL/min. Unlike orally administered cyclodextrin, which is completely retained in the gut lumen, significant accumulation of iv cyclodextrin can occur in the presence of impaired renal function.\textsuperscript{13} The toxicity and efficacy of iv cyclodextrin itraconazole in organ transplant recipients therefore awaits further studies.

Low-dose amphotericin B has not proven uniformly effective as prophylaxis\textsuperscript{17–19} and may, in fact, be associated with a higher incidence of invasive aspergillosis in liver transplant recipients. At least three studies in liver transplant recipients have reported on the use of lipid preparations of amphotericin B for antifungal prophylaxis.\textsuperscript{20–22} Liposomal amphotericin B (1 mg/kg/day) for 5 days was associated with a significantly lower incidence of invasive fungal infections in a randomized, controlled trial. However, no cases of aspergillosis were documented in either study group.\textsuperscript{21} In another report, 7% (4/58) of the liver transplant recipients developed fungal infections (three invasive aspergillosis and one invasive candidosis) despite liposomal amphotericin B at a dose of 1 mg/kg administered for 7 days after transplantation.\textsuperscript{20} Finally, 21 of 100 liver transplant recipients identified as being at high risk for invasive fungal infections received liposomal amphotericin B (1 mg/kg/day) for 7–10 days.\textsuperscript{22} Invasive aspergillosis developed in 10%, although no cases of candidosis were documented.\textsuperscript{22} Thus, in the dosages and duration employed, liposomal amphotericin B has not been uniformly protective against invasive fungi, particularly aspergillosis. Whether higher dosages or long duration of this or other lipid preparations would be more efficacious, remains to be determined.

Since the portal for entry for Aspergillus spp. is the lungs, aerosolized amphotericin B prophylaxis has been attempted. Aerosolized amphotericin B prophylaxis administered during post-transplant hospital stay was shown to reduce significantly the incidence of fungal infections including aspergillosis, in lung, heart–lung and heart transplant recipients; the benefit was sustained for 12 months after transplantation.\textsuperscript{23} A number of retrospective analyses have also showed a significant reduction in invasive aspergillosis compared with historical controls in the transplant setting when aerosolized amphotericin B was used.\textsuperscript{25–28} On the other hand, a randomized, controlled trial utilizing aerosolized amphotericin B in neutropenic patients with haematological malignancies failed to show a reduction in invasive aspergillosis when compared with placebo.\textsuperscript{29}

Animal studies have suggested that aerosolized lipid preparations of amphotericin B may be more effective as prophylaxis for invasive aspergillosis than amphotericin B deoxycholate. In a rat model of pulmonary aspergillosis, aerosolized amphotericin B lipid complex provided higher and more sustained concentrations of the drug in the lungs and was more effective in delaying mortality than aerosolized amphotericin B deoxycholate.\textsuperscript{30} In lung and heart–lung transplant recipients, nebulized liposomal amphotericin B was administered daily for 4 days after transplantation, followed by four weekly treatments.\textsuperscript{31} Only one of 20 patients (5%) was documented to have an invasive fungal infection. These data, while encouraging, are preliminary and warrant larger comparative trials.\textsuperscript{31}

Given the uncertain clinical benefit of prophylaxis and the fact that the infection is relatively infrequent, universal prophylaxis against invasive aspergillosis is neither advisable nor feasible. Alternatives to prophylaxis include pre-emptive therapy targeted towards high-risk patients (Table).\textsuperscript{2,32} Although an optimal antifungal regimen for such an approach has not been determined, itraconazole

\textbf{Table.} Incidence and transplant-specific risk factors for invasive aspergillosis in organ transplant recipients

<table>
<thead>
<tr>
<th>Type of transplant</th>
<th>Incidence (% of invasive aspergillosis [range, (average)])</th>
<th>Particular risk factors</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lung</td>
<td>3–14 (8)</td>
<td>Aspergillus colonization (except in cystic fibrosis patients), cytomegalovirus infection, obliterative bronchiolitis, allograft rejection</td>
</tr>
<tr>
<td>Heart\textsuperscript{a}</td>
<td>0–11 (6)</td>
<td>No data</td>
</tr>
<tr>
<td>Liver</td>
<td>1–8 (1.7)</td>
<td>Allograft dysfunction, dialysis, OKT3 use</td>
</tr>
<tr>
<td>Renal</td>
<td>0–0.9 (0.7)</td>
<td>Graft failure, augmented immunosuppression</td>
</tr>
<tr>
<td>Small bowel</td>
<td>0–3.6 (2.2)</td>
<td>No data\textsuperscript{a}</td>
</tr>
<tr>
<td>Pancreas</td>
<td>0–2.9 (1.3)</td>
<td>No data\textsuperscript{a}</td>
</tr>
</tbody>
</table>

\textsuperscript{a}Specific risk factors for invasive aspergillosis have not been well defined for these transplant groups.
with or without aerosolized amphotericin B or a lipid preparation of amphotericin B (3–5 mg/kg/day) may be considered. Alternatively, in a high-risk patient, early empirical therapy with a lipid preparation of amphotericin B may be initiated (pending confirmation of invasive disease) based on clinical presentation compatible with invasive aspergillosis.7

A worrying observation, largely paralleling the trends in the non-transplant setting, is the rising incidence of non-albicans Candida spp. and the emergence of azole resistance in candida.8 Although controversial, the use of fluconazole has been proposed as one of the factors accounting for the ecological shift in the relative prevalence of Candida spp.33–35 A recent study in bone marrow transplant recipients also documented a significantly higher incidence of aspergillus/zygomycete infection in patients receiving fluconazole as compared with those who received no antifungal prophylaxis.36

At my institution, antifungal prophylaxis with fluconazole is not employed and, in my opinion, universal prophylaxis with fluconazole is not indicated. Invasive Candida glabrata infection with reduced susceptibility to azoles occurred in 4% (4/101) of the liver transplant recipients receiving fluconazole prophylaxis and was the direct cause of death in one patient.9 Fluconazole (400 mg/day) employed for 10 weeks was shown to be associated with a significantly lower rate of fungal infections as compared with placebo in liver transplant recipients.37 An unusually high rate of proven fungal infections (43%) was documented in this study. However, the incidence of invasive candidosis (7%) was virtually identical to that in many other studies where no systemic antifungal prophylaxis was employed.38,39 In another randomized trial comparing fluconazole with nystatin for the first 28 days after transplantation, a decrease in colonization with Candida spp. and superficial fungal infections was observed.40 However, a difference in the incidence of invasive candidosis could not be shown between the two groups.40 In the light of these data, fluconazole prophylaxis targeted towards high-risk patients only may be a more rational alternative to universal antifungal prophylaxis.

Antifungal therapy

Amphotericin B deoxycholate in dosages ranging between 1.0 and 1.5 mg/kg/day has long been regarded as the standard therapy for invasive aspergillosis. However, amphotericin B in such dosages in organ transplant recipients is frequently complicated by nephrotoxicity, precluding its continuation or necessitating a dosage reduction. In solid organ transplant recipients receiving amphotericin B deoxycholate for aspergillosis, nephrotoxicity developed in 36% and dialysis was required in 18%.41 It is imperative that the need for dialysis be prevented in transplant recipients. Post-transplant dialysis in organ transplant recipients portends a grave outcome and has been shown to be an independent predictor of death in liver transplant recipients.42

In an animal model, liposomal amphotericin B (10 mg/kg/day) was more effective in preventing dissemination of pulmonary aspergillosis than amphotericin B deoxycholate (1 mg/kg/day).43 Both amphotericin B deoxycholate and liposomal amphotericin B increased survival and delayed mortality, but only liposomal amphotericin B reduced the number of fungal cfus and prevented dissemination of pulmonary aspergillosis.45 Although prospective controlled trials are lacking, in a retrospective study of patients, including transplant patients with invasive aspergillosis, clinical response rate (48.8% versus 23.4%) and survival rate (50% versus 28.4%) among patients treated with amphotericin B colloidal dispersion were significantly higher than among amphotericin B deoxycholate-treated patients (P < 0.001 for both comparisons).44 Multivariate analysis showed that treatment group was the best predictor of response.45 In another study, liver transplant recipients with invasive aspergillosis treated with amphotericin B lipid complex between 1995 and 1998 were compared with a retrospective cohort of patients treated between 1992 and 1995 with amphotericin B deoxycholate.46 Invasive aspergillosis-associated mortality was 83% in the patients who received amphotericin B deoxycholate compared with 17% in those who received amphotericin B lipid complex (P = 0.0003).46

The higher tissue concentrations achievable with the lipid formulations of amphotericin B are proposed to have led to their greater clinical efficacy. Uptake by the reticuloendothelial system is believed to account for disposition of liposomes.7 Thus, liposomal amphotericin B preparations concentrate primarily in the spleen and liver, organs that harbour reticuloendothelial system phagocytic cells. Drug-laden liposomes are also taken up and transported to the site of inflammation or infection by monocytes and macrophages. In transplant recipients with invasive aspergillosis, concentrations in the lungs achievable with liposomal amphotericin B were 17–78 times higher than those obtained with amphotericin B deoxycholate.47 Additionally, amphotericin B lipid complex has been shown to augment synergistically the antifungal activity of pulmonary alveolar macrophages with, or without, macrophage colony stimulating factor.48 Amphotericin B deoxycholate did so only in the presence of macrophage colony stimulating factor whereas itraconazole had no effect on fungicidal activity of pulmonary alveolar macrophages.49 Thus, low nephrotoxicity, higher achievable tissue levels and possibly greater clinical efficacy support the use of lipid preparations of amphotericin B as therapy of choice for invasive aspergillosis in organ transplant recipients.

The role of itraconazole as initial therapy for invasive aspergillosis is unproven. However, it may be used as follow-up therapy for patients who have experienced initial improvement with amphotericin B.2 Clinical trials are
underway to assess the efficacy of voriconazole for invasive aspergillosis.

Echinocandins, unlike azoles, exhibit fungicidal activity against a number of medically important fungi.49 These drugs inhibit β-(1,3)-D-glucan synthesis. Thus, while active against Candida and Aspergillus spp., and Pneumocystis carinii, they are inactive against most Cryptococcus neoformans, since it possesses little or no β-(1,3)-D-glucan. Owing to the unique mechanism of action, the newer echinocandins, e.g. MK-0991, are also active against candida isolates resistant to other antifungal agents, including azoles.49 Clinical trials to assess the efficacy of echinocandins against invasive aspergillosis are being undertaken. In an animal model, pneumocandicas administered as an aerosol were also effective against invasive pulmonary aspergillosis.50

Amphotericin B is also considered the drug of choice for invasive candidosis. Dosages of amphotericin B deoxycholate (0.5–0.7 mg/kg/day) used for candidosis are likely to be better tolerated in transplant recipients than the higher dosages required for invasive aspergillosis. However, for patients intolerant of amphotericin B deoxycholate or for those with pre-existing renal dysfunction, lipid preparations of amphotericin B may be employed. Use of fluconazole is also reasonable for infections due to Candida albicans.

Based on studies in immunocompromised patients with human immunodeficiency virus (HIV) infection, amphotericin B deoxycholate (0.7 mg/kg/day) is regarded by many as the therapy of choice for cryptococcosis in transplant recipients.52 I prefer the addition of 5-flucytosine (5FC) to amphotericin B for the first 2 weeks, at a dosage of 75 mg/kg/day in divided doses. This dose is better tolerated and less likely to be associated with marrow suppression than a dose of 100 mg/kg/day. Lipid preparations of amphotericin B are an alternative for patients intolerant of amphotericin B deoxycholate. A regimen of oral fluconazole plus oral 5FC has been successfully used in a liver transplant recipient with non-meningeal cryptococcosis.52 Lifelong maintenance, as in the HIV setting, is not necessary in transplant recipients. Itraconazole is the drug of choice for skin and soft tissue infections due to dematiaceous fungi, whereas amphotericin B plus 5FC is preferred for systemic infections caused by these fungi.53

Adjuunctive therapies

Role of surgery

Surgery has been utilized as an adjunct to antifungal therapy for a number of invasive mycoses. The purpose of surgical treatment is two-fold: prevention of serious, at times fatal, haemoptysis in cases of angioinvasive fungi, e.g. aspergillosis and zygomycosis, and a reduction in fungal burden. Early thoracic computed tomographic scans, along with aggressive medical and surgical approach to therapy, led to survival rates of 72% among neutropenic haematological patients with invasive aspergillosis.54 Post-surgical survival of 64% was documented in another report in patients with haematological malignancy and invasive aspergillosis who underwent surgery.57 Although anecdotally reported to lead to a successful outcome, the role of surgery in the routine management of invasive aspergillosis in solid organ transplant recipients is less well-defined.55 Surgery, however, is a definite indication for aspergillus lesions in close proximity to pulmonary vessels, an ominous finding with a high risk of rupture and fatal haemoptysis. Thoracic computed tomography has been shown to play a major role in early identification of such lesions.56

Surgery is considered a critical component of the management of another angioinvasive mycosis, zygomycosis.56 The rhinocerebral form accounts for nearly 57% of all cases of zygomycosis in solid organ transplant recipients.56 In a recent review, 45% (10/22) of organ transplant recipients in whom rhinocerebral zygomycosis was diagnosed ante mortem and who received amphotericin B, were cured;56 seven of the 10 patients cured, however, also underwent surgical debridement. Five of the six transplant recipients with pulmonary zygomycosis in whom the infection was diagnosed ante mortem, were cured. Two patients received amphotericin B alone, two underwent lobectomy in addition to amphotericin B and one patient was treated with lobectomy alone.

Surgery is considered the mainstay of therapy for phaeohyphomycosis. The infections caused by dematiaceous or black pigmented fungi may be difficult to eradicate, even with prolonged antifungal therapy.57 Thus, for these fungi, combined surgical resection with antifungal therapy is the recommended therapeutic approach.53 For cutaneous and subcutaneous phaeohyphomycosis, complete, wide and deep margin resection should be performed.57 Surgical resection, when feasible, is also recommended for central nervous system lesions due to phaeohyphomycosis. Complete neurosurgical resection or drainage of the lesion was the most significant variable determining survival in a report of 26 cases of central nervous lesions due to Cladophialaphora bantiana in non-transplant patients.45

Immunomodulatory therapies

The predominant host defences against invasive aspergillosis are polymorphonuclear leucocytes (PMNs) and macrophages. Corticosteroids have been shown to impair monocyte/macrophage function against Aspergillus spp. and cyclosporin and tacrolimus inhibit interferon-γ, a potent activator of macrophages.58 Granulocyte–macrophage colony stimulating factor (GM-CSF) has been documented to increase the phagocytic activity of PMNs and other effector cells.58 GM-CSF augmented the antifungal activity of mononuclear phagocytes against both conidia and hyphae of Aspergillus fumigatus, in part by enhancing the oxidation-dependent mechanisms.58 In vitro data and studies in bone marrow transplant recipients have shown a
promising role for GM-CSF as adjunctive therapy for invasive fungal infections.59,60

Of concern, however, is the fact that in an animal model of invasive aspergillosis, granulocyte colony stimulating factor (G-CSF) not only proved ineffective, but antagonized potent antifungal agents.61 Thus, the precise role of haemopoietic growth factors in the treatment of invasive fungal infections in organ transplant recipients remains undefined, and their use at the current time can only be recommended in the context of clinical trials.

Interferon-γ has been shown to enhance the fungicidal activity and the oxidative burst of PMNs in response to *A. fumigatus* hyphae and prevent the suppression of PMN-induced hyphal damage resulting from corticosteroids.50,57,61 In patients with chronic granulomatous disease, interferon-γ led to a reduction in the risk of serious infections, including pulmonary aspergillosis.62 The combination of interferon-γ and GM-CSF or G-CSF may have an additive effect on the antifungal activity of PMNs and macrophages.63 Aerosolized interferon-γ has also been shown to activate human alveolar macrophages.64 A potential concern with the use of interferon in organ transplant recipients is the augmented risk of allograft rejection. The clinical data on the use of immunomodulatory therapy in organ transplant recipients are limited and must await further studies.

References


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Invasive mycoses: controversies in prophylaxis and management


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