Background. Posttransplant renal replacement therapy has been shown to be an independently significant risk factor for invasive fungal infections after liver transplantation. We assessed the efficacy of a lipid preparation of amphotericin B as prophylaxis for invasive fungal infections, directed only toward liver transplant recipients requiring renal replacement therapy.

Methods. A total of 148 patients transplanted between 1990 and 1997 received no antifungal prophylaxis. Since 1997, 38 patients underwent liver transplantation; antifungal prophylaxis with a lipid preparation of amphotericin B was used in patients requiring renal replacement therapy.

Results. Fifteen percent (22 of 148) of the patients transplanted before 1997 required renal replacement therapy. In this cohort, the incidence of invasive fungal infections (36% vs. 7%, \( P=0.0007 \)) and invasive aspergillosis (14% vs. 2%, \( P=0.02 \)) was significantly higher in patients who required renal replacement therapy compared with those who did not. Since 1997, 29% (11 of 38) of the patients required renal replacement therapy and received antifungal prophylaxis. Invasive fungal infections occurred in 36% (8 of 22) of the patients who received no prophylaxis (patients before 1997), and 0% (0 of 11, \( P=0.03 \)) in those who received antifungal prophylaxis (since 1997). Antifungal prophylaxis was independently associated with protection from fungal infection (\( P=0.017 \)). No reduction in mortality with antifungal prophylaxis was documented.

Conclusion. Prophylaxis with a lipid preparation of amphotericin B was associated with a significant reduction in invasive fungal infections in high-risk liver transplant recipients, i.e., those requiring renal replacement therapy. However, no beneficial effect on survival could be documented.

INTRODUCTION

Invasive fungal infection remains amongst the most significant infectious complication in organ transplant recipients. Overall, invasive fungal infections have been reported in 5% to 42% of the liver transplant recipients with mortality rate of 25% to 56% (1–5). Although a vast majority of the invasive fungal infections are due to Candida, the unique vulnerability of liver transplant recipients to invasive aspergillosis is well recognized (6, 7). Liver transplant recipients are notably predisposed to dissemination of Aspergillus infections beyond the lungs (8, 9). These patients also compose a subgroup of organ transplant recipients in whom mortality rate due to invasive aspergillosis exceeds 90%; an estimated 16.9% of all deaths in liver transplant recipients have been due to Aspergillus infections (6).

Renal dysfunction, particularly the requirement of renal replacement therapy has been shown to be an independently significant risk factor for invasive fungal infections, including aspergillosis in liver transplant recipients (2–4, 7, 10). We have previously shown that the risk for developing invasive fungal infections in dialyzed patients was 21-fold higher than in other patients (10). In the study herein, we assessed the efficacy of a lipid preparation of amphotericin B as prophylaxis for invasive fungal infections, directed only toward the high-risk liver transplant recipients, i.e., those requiring renal replacement therapy (hemodialysis or continuous venovenous hemodialysis).

The study population comprised 186 consecutive liver transplant recipients transplanted between January of 1990 and January of 2000. All patients received tacrolimus and low-dose prednisone as immunosuppressive therapy as previously reported (3, 11). Rejection episodes were treated with 1 g of methylprednisolone bolus with or without steroid cycles (methylprednisolone given intravenously in four divided doses daily, tapering the dose from 200 mg to 20 mg per day over 6 days). OKT3 was used for steroid-resistant rejection. Since October of 1995, mycophenolate mofetil (1000 mg twice daily orally) was added to the above-mentioned immunosuppressive regimen of all patients undergoing liver transplantation, if neurotoxicity or nephrotoxicity suspected to be due to tacrolimus developed (12). Perioperative prophylaxis consisted of ampicillin andcefotaxime for 24 hr. Trimethoprim (80 mg) and sulfamethoxazole (400 mg) once daily was used indefinitely as Pneumocystis carinii prophylaxis. Upon detection of CMV infection, preemptive ganciclovir was used as prophylaxis for CMV disease as previously described (13).

Invasive candidiasis required histopathologic evidence of tissue invasion by biopsy, or on autopsy, or isolation of Candida species in one or more blood cultures, or isolation of Candida in normally sterile body fluid or sites, with samples collected intraoperatively or by percutaneous needle aspirate (3, 4, 14). Yeast colonization at isolated sites, e.g., oral cavity,
sputum, upper respiratory tract, stool or urine, in the absence of tissue invasion was considered insufficient evidence of invasive candidal infection. Diagnosis of Cryptococcus neoformans was established by isolation in culture. Invasive aspergillosis required evidence of tissue invasion on biopsy or autopsy plus isolation of Aspergillus species in culture (6, 15).

Of 186 consecutive study patients, 98% (182 of 186) were male. Their mean age was 48 years. The demographic characteristics of the study patients are outlined in Table 1. A total of 148 patients transplanted between January 1990 and April 1997 received no antifungal prophylaxis. Of these 15% (22 of 148) required renal replacement therapy. In 59% (13 of 22) of these patients, renal replacement therapy was required in the early posttransplant period (within 3 months of transplantation) and in 41% (9 of 22), in the late posttransplant period (>3 months after transplantation). Fifty-four percent (12 of 22) of the patients requiring early renal replacement therapy were either in the ICU or were receiving pretransplant renal replacement therapy. The overall incidence of invasive fungal infections (36%, 8 of 22 vs. 7%, 9 of 126, \( P = 0.0007 \)) and, in particular, invasive aspergillosis (14%, 3 of 22 vs. 2%, 2 of 126, \( P = 0.02 \)) was significantly higher in patients who required renal replacement therapy as compared with those who did not. Invasive fungal infections in these patients included, C. albicans (n=2), C. glabrata (n=2), A. fumigatus (n=3), and C. neoformans (n=1).

In logistic regression analysis (with age, CMV infection, increased immunosuppression and renal replacement therapy in the model), only age (\( P = 0.04 \)) and renal replacement therapy (\( P = 0.003 \)) were significantly associated with invasive fungal infections. Thirty-eight percent (5 of 13) of the patients with early-onset and 33% (3 of 9) of those patients with late-onset renal replacement therapy developed an invasive fungal infection (\( P = 0.8 \)). The median time to onset of fungal infection after renal replacement therapy was 13 days and ranged between 2 and 191 days.

Since April of 1997, 38 consecutive patients underwent liver transplantation. Of these, 29% (11 of 38) required renal replacement therapy and received a lipid preparation of amphotericin B as prophylaxis. The lipid preparation of amphotericin B used was 5 mg/kg per day of either amphotericin B lipid complex (Abelcet) in nine patients or liposomal amphotericin B (AmBisome) in two patients. Antifungal prophylaxis was continued until death, discharge, or discontinuation of renal replacement therapy. Invasive fungal infections occurred in 36% (8 of 22) of the patients who received no prophylaxis (before 1997) and in 0% (0 of 11, \( P = 0.03 \)) of those who received antifungal prophylaxis (since 1997) (Table 2).

In logistic regression analysis (with age, CMV infection, increased immunosuppression, and use of antifungal prophylaxis in the model), only antifungal prophylaxis was independently associated with protection from fungal infection (\( P = 0.017 \)).

One-year mortality rate was similar in two cohorts of patients requiring renal replacement therapy; 55% (12 of 22) of the patients who did not receive antifungal prophylaxis died as did 55% (6 of 11) of those who received antifungal prophylaxis (Table 2). Whereas, invasive fungal infections (all due to Aspergillus) accounted for 25% (3 of 12) of the deaths in the patients who did not receive antifungal prophylaxis, none (0 of 6) of the deaths in those who received antifungal prophylaxis were due to fungal infections. Other causes of death in the patients requiring renal replacement therapy in the cohort before 1997, were bacterial infections (\( E. \) coli \( n=1, \) E. cloacae \( n=1 \), and methicillin-resistant \( S. \) aureus \( n=1 \)) and multiorgan system failure in 6 cases. The death in patients requiring renal replacement therapy since 1997 was considered attributable to methicillin-resistant \( S. \) aureus bacteremia in three cases, perforated small bowel with polymicrobial bacteremia in one case, and multiorgan system failure in two cases.

Given the relatively low incidence of invasive fungal infections, the expense, toxicity, and potential for the emergence of resistance, employment of antifungal prophylaxis for all patients is neither justifiable nor feasible (6). Instead, prophylaxis targeted toward high-risk patients has been proposed as a more rationale strategy (6). However, the types of patients to receive prophylaxis, the choice of antifungal agent, and the timing of prophylaxis remains controversial.

In the current era of conservative utilization of OKT3, and a declining incidence of CMV, renal dysfunction has emerged as the foremost predictor of invasive aspergillosis in liver transplant recipients (15). Institution of renal replacement therapy is a readily discernible clinical event that identifies liver transplant recipients at risk for fungal infections. Furthermore, requirement of renal replacement therapy, regardless of the time of institution of renal replacement therapy (whether in the early or late posttransplant period), portends a higher risk for invasive fungal infections. Therefore, we chose to target this subset of high-risk liver transplant recipients for antifungal prophylaxis.

The rationale for using a lipid preparation of amphotericin B and for selecting a 5 mg/kg per day dosage, deserves mention. Invasive aspergillosis is an early occurring infection after liver transplantation; 81% to 100% of the patients developing invasive aspergillosis are still in the intensive care unit after transplant surgery (6). Invasive infection due to Aspergillus has been shown to ensue rapidly in these critically ill patients. Indeed, the median time from renal replacement therapy to onset of invasive aspergillosis in our patients was 13 days. Thus, we reasoned that an antifungal agent used as prophylaxis in this setting must be potent and

### Table 1. Demographic characteristics of the study patient

<table>
<thead>
<tr>
<th>Age, mean (years)</th>
<th>48, range 21–72</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex, % male</td>
<td>98% (182/186)</td>
</tr>
<tr>
<td>UNOS status</td>
<td></td>
</tr>
<tr>
<td>2a</td>
<td>25% (46/186)</td>
</tr>
<tr>
<td>2b</td>
<td>73% (136/186)</td>
</tr>
<tr>
<td>3</td>
<td>2% (4/186)</td>
</tr>
<tr>
<td>Underlying liver disease&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>Alcohol</td>
<td>41% (76/186)</td>
</tr>
<tr>
<td>Hepatitis C virus</td>
<td>56% (104/186)</td>
</tr>
<tr>
<td>Hepatitis B virus</td>
<td>10% (18/186)</td>
</tr>
<tr>
<td>Hepatocellular carcinoma</td>
<td>10% (18/186)</td>
</tr>
<tr>
<td>Primary sclerosing cholangitis</td>
<td>9% (17/186)</td>
</tr>
<tr>
<td>Cryptogenic</td>
<td>5% (10/186)</td>
</tr>
<tr>
<td>Other&lt;sup&gt;b&lt;/sup&gt;</td>
<td>5% (9/186)</td>
</tr>
<tr>
<td>Primary biliary cirrhosis</td>
<td>2% (3/186)</td>
</tr>
<tr>
<td>Retransplantation</td>
<td>8% (15/186)</td>
</tr>
</tbody>
</table>

<sup>a</sup> Some patients had more than one underlying liver disease.

<sup>b</sup> Other underlying liver diseases included metabolic, sarcoidosis, polycystic, and autoimmune liver disease.
be rapidly able to achieve systemic drug levels considered adequate for activity against Aspergillus.

Currently available antifungal agents with activity against Aspergillus are limited to itraconazole, amphotericin B deoxycholate, and lipid formulations of amphotericin B (17, 18). A controlled trial has compared itraconazole oral solution with placebo for the prevention of systemic fungal infections in liver transplant recipients. However, the study was insufficiently powered to document an efficacy against Aspergillus infections (19). We have previously demonstrated lack of efficacy of low-dose amphotericin B (0.1 to 0.5 mg/kg per day) as prophylaxis for Aspergillus infections in liver transplant recipients (20). Barring their cost, we considered the lipid formulations of amphotericin B attractive agents for prophylaxis for invasive aspergillosis in our patients. These drugs not only have a good safety profile but also have been documented to be at least equivalent in efficacy against invasive mycelial infections compared with amphotericin B (21–23).

Case-series in liver transplant recipients, however, have documented a failure of prophylaxis for Aspergillus with an AmBisome dosage of 1 mg/kg per day (16, 24). Because an optimal dose of a lipid formulation of amphotericin B for prophylaxis for invasive aspergillosis has not been defined, we feared that our approach toward prophylaxis could prove ineffective merely because a suboptimal dose of the antifungal agent had been used. Therefore, we chose to use a lipid preparation of amphotericin B at a dosage of 5 mg/kg per day as antifungal prophylaxis targeted toward the high-risk liver transplant recipients.

The antifungal prophylaxis was independently associated with protection against invasive fungal infections in our patients; unfortunately, the overall survival was not significantly different between the dialyzed patients who did and those who did not receive prophylaxis. Although fewer deaths related to fungal infections were observed in the dialyzed patients before 1997 compared with those since 1997, the difference was not statistically significant. Bacterial infections, particularly due to multiresistant bacteria, accounted for 40% of the deaths in the dialyzed patients who did not die of fungal infections. Thus, even if invasive fungal infections were effectively prevented, patients requiring renal replacement therapy succumbed to a multiresistant bacterial infection or multiorgan system failure. These data underscore the grave overall prognosis that renal replacement therapy portends after liver transplantation (11, 25).

Our study has limitations that deserve to be acknowledged. Foremost amongst these limitations is the comparison of the efficacy of antifungal prophylaxis in the interventional cohort with a historic cohort. To demonstrate the efficacy of intervention in a prospective, controlled trial, 70 patients on renal replacement therapy would have been required to document a difference in the rate of fungal infection between 36% without intervention, and 5% with intervention at alpha=0.05 and power=80%. Assuming that only 15% of all liver transplant recipients required renal replacement therapy, an estimated 466 patients would have been needed to document the efficacy of intervention in a prospective trial. Accumulating patients to attain this sample size within a reasonable time was deemed logistically unfeasible. Furthermore, given that renal replacement therapy was such a strong predictor of fungal infections in our patients, the conduct of a randomized, controlled trial (without prophylaxis in one arm) was considered unrealistic, if not unethical. Finally, we point out that our analyses controlled for confounding variables that could have influenced the incidence of fungal infections, including CMV infection, retransplantation, and immunosuppression (Table 2); only lack of antifungal prophylaxis was a significant predictor of fungal infections.

In summary, prophylaxis with a lipid preparation of amphotericin B targeted toward high-risk liver transplant recipients requiring renal replacement therapy was associated with a significant reduction in invasive fungal infections. However, a beneficial effect on survival could not be documented in the patients who received antifungal prophylaxis. Indeed, no study to our knowledge has documented a reduction in mortality with the employment of antifungal prophylaxis in liver transplant recipients (26–28). These data are a sobering reminder that unless risk factors that render the liver transplant recipients vulnerable to fungal infections can be modified, antifungal prophylaxis, despite its efficacy, may not lead to a decrease in mortality.

REFERENCES


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