Case Series

Selection of Resistant Fungi in Liver Transplant Recipients During Use of Newer Antifungal Agents — A Report of Two Cases

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Abstract

Introduction: Because invasive fungal infections cause significant morbidity and mortality in liver transplant recipients, the use of antifungal prophylaxis, and the early empirical use of antifungal agents, is widespread on liver transplant units. The new-generation azoles such as voriconazole and the echinocandins have been welcome additions to the antifungal armamentarium. These agents have become the leading options for prophylaxis in liver transplant units, despite the absence of strong data for their efficacy in this setting. Clinical picture: We report two recipients of living-donor liver transplants who became infected/colonised with fungi resistant to an echinocandin and the azoles after exposure to these agents. One patient developed trichosporonosis while on caspofungin and the other became infected/ colonised with *Candida glabrata* that was resistant to voriconazole and posaconazole. Conclusion: We report these to highlight some of the consequences of using the newer antifungal agents.

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Introduction

Invasive fungal infections (IFI) are associated with a high mortality in liver transplant recipients.[1] The incidence of fungal infections after liver transplantation ranges from 7% to 42%, with *Candida sp.* and *Aspergillus sp.* being the commonest pathogens.[2] A prophylactic strategy against fungi is therefore attractive, and is practised in many liver transplant units.[3] In a meta-analysis, prophylaxis reduced the total number of proven fungal infections, and mortality attributable to fungal infections, but it did not improve overall mortality.[4]

Prophylaxis, however, may not be the best strategy, and may have an ecological impact on the selection of resistant fungi.[3] A meta-analysis of antifungal prophylaxis in liver transplant recipients noted an increase in *Candida glabrata* infections among those who received prophylaxis.[4] Outside the field of transplantation multiple studies have demonstrated the link between the increased use of fluconazole (FCZ) and the development of FCZ-resistant *Candida*. The link between voriconazole (VCZ), a new-generation azole, and the development of resistance, though less well-reported, is unsurprising.[5]

Caspofungin (CAS) was the first commercially-available echinocandin, a new class of anti-fungal agent. It has very good activity against almost all *Candida* species and has also demonstrated activity against *Aspergillus*.[6] It has a good side effect profile. Despite the absence of evidence documenting efficacy of the echinocandins as antifungal prophylaxis in liver transplant recipients, Singh et al[3] found, from a survey of 59 liver transplant programs, that the echinocandins were the top choice for mold-active antifungal prophylaxis. More recently, though, CAS was shown to be efficacious and well-tolerated when used for antifungal prophylaxis in liver transplant recipients.[7] Its somewhat narrow spectrum of antifungal activity, however, implies that super-infection with a fungus intrinsically resistant to it is possible. In particular, it has no activity against *Trichosporon*.

Given the popularity of VCZ and CAS in liver transplant units, we report 2 patients in whom the use of these newer

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antifungal agents was associated with the isolation of fungi resistant to them.

Case Report

The first patient, a 38-year-old man, was transferred to the Medical Intensive Care Unit of our hospital from another country for the management of acute hepatic failure, likely to be drug-induced. He had received multiple antibiotics before arrival in our hospital. He received a right-lobe graft after 6 days of transfer. Fluconazole was used for antifungal prophylaxis, in accordance with departmental guidelines, which recommended it for patients undergoing transplant for fulminant hepatic failure. The transplant surgery was complicated by the need for a colostomy, as the colon had been found to be grossly dilated, with a few dusky areas. He received standard immunosuppressant's basiliximab, mycophenolate mofetil and hydrocortisone. Post-transplant, vancomycin and meropenem were started for suspected bacterial sepsis. On the fourth post-transplant day (PTD), caspofungin (CAS) was added for suspected candidemia. Blood cultures drawn on that day (and 3 days later) were positive for *Candida lusitaniae*. On the tenth PTD, a feculent discharge was noted from the wound and a laparotomy revealed an ileal perforation. An ileostomy was created. Blood cultures yielded *Stenotrophomonas maltophilia* and the patient was switched to levofloxacin. At this point, Candida was no longer isolated. Three weeks later, while still on CAS, blood cultures grew *Trichosporon asahii*, and the patient was switched to voriconazole (VCZ), which led to clearance of fungemia. The patient's hospitalisation was complicated by development of multiple intra-abdominal abscesses, difficult to clear *Stenotrophomonas maltophilia* bacteraemia. He died about 16 weeks after transplant.

Case 2 was a 56-year-old man with chronic Hepatitis B infection and hepatocellular carcinoma, who underwent an elective living-donor liver transplant with a left-lobe graft. He received standard immunosuppressant's and antifungal prophylaxis with syrup nystatin as per departmental guidelines. In the early post-transplant period, hypodensities developed in segment 3, suggestive of infarction. Persistent fevers led to the placement of a biliary drain. Bile aspirated on the 79th PTD grew *Candida albicans* (FCZ MIC 1 ug/ml), and the patient was given fluconazole (FCZ). The patient had several episodes of fever, and received broad-spectrum antibiotics, including cefepime and meropenem (sequentially). FCZ was continued for 4 weeks in view of intermittent fevers. Two weeks into treatment with fluconazole, biliary fluid culture did not reveal any fungal infection. When patient was readmitted on the 139th PTD due to new onset of fever, cholangiogram led to bile cultures that grew *C. albicans* (FCZ MIC 2 ug/ml) and *C. krusei* which was treated with voriconazole. While on voriconazole, bile aspirated on the 160th PTD grew *C. glabrata* that was resistant to FCZ (MIC >256 ug/ml), VCZ (MIC > 4 ug/ml), itraconazole (ITC) (MIC >32 ug/ml) and posaconazole (MIC >32 ug/ml). Following the susceptibility results, voriconazole was changed to caspofungin and treated for 2 weeks. As the patient did well, bile was not recultured. Other significant post transplant issues include several admissions for bacterial sepsis related to line related/hepatobiliary system infections. The patient died suddenly at home 1 year after transplant, possibly due to a cardiac event; the last 3 months of life were free of medical complications.

Discussion

Caspofungin was the first in a new class of antifungal agents, the echinocandins, which also includes micafungin and anidulafungin. CAS has a relatively narrow spectrum of antifungal activity, covering *Candida sp.* and *Aspergillus sp.*[6] One consequence of this relatively narrow spectrum is illustrated by Case 1 above. Several authors have reported breakthrough Trichosporonosis in patients receiving CAS, though none in a liver transplant recipient. Goodman et al[8] described a patient with refractory leukemia who underwent an allogeneic haematopoietic stem cell transplant (HSCT) while receiving CAS for a presumed fungal pneumonia. Wrist pain led to aspiration of tenosynovial sheaths, and cultures grew *Trichosporon beigelii*. Bayramoglu et al[9] described a patient with acute leukemia who developed fungemia with *T. asahii* while receiving CAS as empirical therapy for febrile neutropenia. Although trichosporonosis has been reported in liver transplant recipients, none

Table 1. Demographic Characteristics and Outcomes of Patients with Invasive *Trichosporon* Infection after Liver Transplantation

Patient	Age	Sex	Reason for Transplant	Time from Transplant	Outcome	Reference
1	25	Male	Acute Hepatitis B virus infection	2 days	Death	Ness et al[10]
2	39	Male	Primary Biliary Cirrhosis	7 days	Death	Finkelstein et al[11]
3	51	Female	Primary Biliary Cirrhosis	4 years 10 months	Death	Abdala et al[12]
4	31	Female	NA	NA	Improved	da silva Rodrigus et al[13]
5	23	Female	Cryptogenic Hepatitis	3rd week	Death	Biasoli et al[14]
6	51	Male	Chronic Hepatitis C virus infection	5 months	Death	Lacasse et al[15]

NA: information not available

were receiving an echinocandin prior to the development of the infection.[10-15] The 6 case reports published so far are summarised in Table 1. Hsin et al[16] characterised breakthrough invasive mycoses in patients (not all of whom were immunocompromised) receiving echinocandins. They found that breakthrough mycoses caused by *Trichosporon* were less frequent than those caused by *Candida* and *Aspergillus*.[16]

Trichosporon species are found ubiquitously in nature. Invasive trichosporonosis which are life threatening, have been increasingly recognised in the compromised hosts. Review of literature reveals these infections to be less common in solid organ transplant recipients and in acquired immunodeficiency syndrome, compared to patients with haematological or oncological malignancies and in those with profound neutropenia.

This case is reported to highlight what might become a new clinical syndrome – that of Trichonosporonosis in liver transplant patients on CAS. This is not a reason to avoid an echinocandin — as with antibacterials, narrow-spectrum agents should be preferred.

Case 2 highlights the relationship between the use of the azoles and the isolation of azole-resistant *Candida*. This phenomenon was seen primarily in patients infected with the human immunodeficiency virus (HIV) who were on long-term azoles for prophylaxis against mucosal and esophageal candidiasis. However, liver transplant recipients may be similarly affected. Fortun et al[17] reported 4 liver transplant patients who developed invasive candidiasis with azole-resistant *C. glabrata* while on azoles.

Intriguing data come from Borst et al[18] They found 5 *C. glabrata* isolates from 1917, 1935, 1960, 1973 and 1975. After establishing baseline susceptibility to FCZ, they cultured them in FCZ-containing media, followed by cultures in FCZ-free media. The first isolates with FCZ MIC of 64 ug/ml emerged on Day 2 of incubation. The last isolates had FCZ MIC from 32 to 64 ug/ml. These investigators also showed that VCZ MIC rose after FCZ exposure, from a baseline of 0.125 ug/ml to 2 ug/ml.[18] This study demonstrating the development of cross-resistance to the newer-generation azole (VCZ) is important as liver transplant (and other) physicians embrace VCZ.[3] As liver transplant patients tend to be those who have had multiple admissions, they are likely to have been exposed to FCZ. The study by Borst et al suggests that susceptibility to VCZ in such a setting is not guaranteed.

Although VCZ is effective against a wider range of *Candida sp.* than FCZ, long-term use of VCZ may also be associated with the isolation of resistant *Candida*. Alexander et al[5] reported 5 HSCT patients with breakthrough *C. glabrata* fungemia while on VCZ. These patients had received VCZ before the fungemia for a median of 48 days; all the 5 had received FCZ before VCZ, for a median of 60 days. The breakthrough *C. glabrata* isolates were resistant to FCZ and ITC, and had MICs to VCZ and posaconazole of 2 ug/ml. Panackal et al[19] have also demonstrated that *C. glabrata* isolates can acquire decreased susceptibility to multiple azoles including VCZ, during exposure to both VCZ and the older azoles, FCZ and ITC. These observations have important therapeutic implications, as it suggests *C. glabrata* can exhibit clinically meaningful resistance across different azole drugs. As far as we know, Case 2 is the first liver transplant recipient in which elevated VCZ MIC has been demonstrated after prolonged use.

We report these 2 cases to highlight clinical consequences of using the newer antifungal agents. As we have pointed out, fungal infections are a problem in liver transplant recipients and prophylaxis does bring benefits.[4] To prevent over usage, we advocate a targeted form of prophylaxis rather than a universal prophylaxis. In our unit, as in others, criteria for starting antifungal prophylaxis in liver transplant recipients are spelt out, such as re-transplantation, prolonged intensive care unit stay and need for renal replacement therapy.[20]

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