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The Deferasirox–AmBisome Therapy for Mucormycosis (DEFEAT Mucor) study: a randomized, double-blinded, placebo-controlled trial

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Objectives: Host iron availability is fundamental to mucormycosis pathogenesis. The combination of liposomal amphotericin B (LAmB) and deferasirox iron chelation therapy synergistically improved survival in diabetic mice with mucormycosis. To determine the safety of combination deferasirox plus LAmB therapy for mucormycosis, a multicentred, placebo-controlled, double-blinded clinical trial was conducted.

Methods: Twenty patients with proven or probable mucormycosis were randomized to receive treatment with LAmB plus deferasirox (20 mg/kg/day for 14 days) or LAmB plus placebo (NCT00419770, clinicaltrials.gov). The primary analyses were for safety and exploratory efficacy.

Results: Patients in the deferasirox arm ($n=11$) were more likely than those in the placebo arm ($n=9$) to have active malignancy, neutropenia and corticosteroid therapy, and were less likely to receive concurrent non-study antifungal therapy. Reported adverse events and serious adverse events were similar between the groups. However, death was more frequent in the deferasirox than in the placebo arm at 30 days (45% versus 11%, $P=0.1$) and 90 days (82% versus 22%, $P=0.01$). Global success (alive, clinically stable, radiographically improved) for the deferasirox arm versus the placebo arm at 30 and 90 days, respectively, was 18% (2/11) versus 67% (6/9) ($P=0.06$) and 18% (2/11) versus 56% (5/9) ($P=0.2$).

Conclusions: Patients with mucormycosis treated with deferasirox had a higher mortality rate at 90 days. Population imbalances in this small Phase II study make generalizable conclusions difficult. Nevertheless, these data do not support a role for initial, adjunctive deferasirox therapy for mucormycosis.

Keywords: antifungal, fungal infections, mould infections, combination therapy

Introduction

Mucormycosis is a life-threatening infection caused by fungi of the order Mucorales. Recently, a crucial link between iron availability and the risk of mucormycosis has been described.[1,2] For example, the predisposition of patients with diabetic ketoacidosis (DKA) to mucormycosis is caused in part by increased free serum iron in the setting of acidaemia.[1–4] As well, the predisposition to mucormycosis of patients treated with deferoxamine[5–8] is now known to be due to deferoxamine's role as a siderophore, which specifically delivers iron to the aetiological fungi.[9–11] Indeed, the administration of deferoxamine or free iron worsens the survival of animals with mucormycosis.[10,12–14] In contrast, other iron chelators, which are not used as siderophores by the Mucorales, do not similarly exacerbate mucormycosis infection in animal models.[1,12,15,16]

In 2005, a new iron chelator, deferasirox, was approved by the US FDA for the treatment of chronic iron overload in patients with transfusion-dependent anaemias.[17] Deferasirox chelated iron from and was fungicidal for clinical isolates of Mucorales *in vitro*.[1] In DKA mice with disseminated mucormycosis, deferasirox was as effective as liposomal amphotericin B (LAmB) therapy and combination deferasirox/LAmB therapy synergistically improved survival.[1] Furthermore, deferasirox has been

administered as open-label therapy to patients with mucormycosis with generally favourable results.[18,19] However, aside from these small numbers of published cases, the safety of deferasirox in the treatment of acutely ill patients with mucormycosis is unknown. The Deferasirox–AmBisome Therapy for Mucormycosis (DEFEAT Mucor) study was conducted to define the safety and exploratory efficacy of short-term deferasirox therapy in patients who were acutely infected with mucormycosis, thus providing a foundation for potential future studies.

Methods

Study design

The DEFEAT Mucor study (NCT00419770, clinicaltrials.gov) was a double-blinded (providers, patients and all study personnel except for study pharmacists, the latter of whom did not participate in any patient evaluations), randomized, placebo-controlled, Phase II trial of adjunctive deferasirox therapy for mucormycosis. Patients were enrolled between January 2008 and October 2009 at the University of California San Francisco (UCSF), the University of Miami Jackson Memorial Medical Center, Duke University Medical Center, the MD Anderson Cancer Center, the Fred Hutchinson Cancer Research Center at the University of Washington, City of Hope Medical Center, Summa Health Systems in Ohio and MedStar Research Institute in Washington DC. Patients were randomized one to one to receive adjunctive deferasirox or placebo therapy, in addition to LAmB. Randomization was accomplished by an automated web-based system established by the Contract Research Organization (Axiom Real-Time Metrics), and randomization was stratified by receipt of haematopoietic/solid organ transplantation. Patients ≥2 years old were eligible if they had proven or probable mucormycosis by modified European Organization for the Research and Treatment of Cancer/Mycoses Study Group criteria.[20] In brief, proven mucormycosis was defined as: (i) histopathological or cytopathological examination showing broad-based, aseptate, ribbon-like hyphae consistent with Mucorales from needle aspiration or biopsy specimen, with evidence of associated tissue damage; OR (ii) a positive culture from a sample obtained from a sterile and clinically or radiologically abnormal site. Probable mucormycosis was defined as: (i) an at-risk host (an absolute neutrophil count <500 cells/μL within 60 days, diabetes, receipt of corticosteroids within 60 days, status post haematopoietic cell or solid organ transplantation, or active graft-versus-host disease); AND (ii) positive culture, cytology or PCR from sputum, bronchoalveolar lavage (BAL), endoscopy/colonoscopy or sinus aspirate/biopsy; AND (iii) clinical signs or symptoms of active infection.

Subjects were excluded if they had a high likelihood of death within 48 h or death due to underlying disease within 30 days, were unable to receive enteral medications, had infection limited to suprafascial skin or had received >14 days of polyene antifungal therapy. Enrolment was discouraged for patients with serum creatinine levels of ≥3 mg/dL or a creatinine clearance of <30 mL/min by the Cockcroft–Gault formula, and those with both an aspartate aminotransferase (AST) or alanine aminotransferase (ALT) >10× the upper limit of normal and a direct bilirubin >5× normal; waivers were issued to allow enrolment of such patients if there were no other exclusion criteria and the subjects met all inclusion criteria.

The study was conducted in accordance with the Declaration of Helsinki and guidelines for studies involving human subjects. An independent Data Safety Monitoring Board oversaw the study conduct and conducted an unblinded interim safety review after six patients were enrolled. The interim safety evaluation results were not shared with the study investigators, aside from recommending that the study continue. The protocol was approved by the institutional review board at each site, as well as at the Los Angeles Biomedical Research Institute at Harbor-University of California Los Angeles Medical Center, which was the sponsor of the study, and all enrolled subjects signed an informed consent form.

Treatment

LAmB dosing was determined by the site investigators, but the protocol specified a minimum dose of ≥5 mg/kg to be given at least thrice weekly during the period of blinded study medication administration. Deferasirox was dosed at 20 mg/kg/day, administered enterally for up to 14 days. Deferasirox and placebo tablets were prepared by unblinded pharmacists by dissolving the tablets in water, orange juice or apple juice. Placebo and deferasirox slurries were not distinguishable after the tablets were dissolved. Although other antifungal therapy was discouraged during the 14 day period of blinded study medications, due to strong resistance from primary physicians waivers were granted if the patient was an otherwise appropriate candidate and the treating physicians refused to enrol the patient unless such therapy was allowed. The type and duration of antifungal therapy after the 14 day period of study medications and the surgical management of such patients were at the discretion of the treating physicians.

Study assessments and endpoint definitions

Clinical signs and symptoms were evaluated prior to enrolment, at days 7 and 14 of study medication administration, and at 30 or 90 days of follow-up. Per the protocol, CT or MRI scans of the affected areas were obtained within 4 days before or after enrolment and again within 4 days after the end of the blinded study medication administration; any other CTs or MRIs obtained for clinical care were also captured. Blood was obtained every other day for safety parameters during the period of blinded study medication administration.

Safety was assessed by defining the frequency, type and severity of adverse events during the on-therapy period. The primary exploratory efficacy endpoint was the global response rate (composite of clinical and radiographic response) at the end of the study drug administration. A blinded, independent Endpoint Adjudication Committee determined whether clinical signs/symptoms and radiographic images were improved, stable or worse at each follow-up visit, based on data provided by the site investigators, and adjudicated global success or failure for each patient based on protocol-specified criteria. Global response for the primary efficacy endpoint was dichotomized as success (patient was alive AND had stable or improved clinical assessment AND had improved radiographic assessment) or failure (patient was dead OR progressed clinically OR had stable or progressed radiographic assessment).

Deferasirox serum concentrations were obtained at 1–3, 4–8, 10–12 and 20–24 h after the seventh study dose. After protein precipitation, serum deferasirox levels were analysed by liquid chromatography–mass spectrometry (WuXi AppTec Co., Ltd, Shanghai, China). Deferasirox MICs for the available fungal strains were determined as previously described.[1]

Statistical analysis

This was a descriptive, early phase study, designed to elucidate safety and important trial aspects for the purpose of planning a potential future, larger efficacy study. The number of subjects to be enrolled ($n=20$) was believed sufficient to identify any common, unanticipated safety concerns in this target population given the extensive antecedent experience with deferasirox in animal and clinical studies. All safety analyses and the primary exploratory efficacy analysis were conducted in the intention-to-treat (ITT) population (i.e. all subjects enrolled). Pre-specified per-protocol (PP) analyses (i.e. enrolled subjects who received at least four doses of study medication) were also conducted. Proportions were

Table 1. Demographics and baseline characteristics of enrolled patients

Characteristic	Deferasirox ($n=11$)	Placebo ($n=9$)
Sex, *n* (%)		
male	9 (82)	6 (67)
female	2 (18)	3 (33)
Median age, years (range)	59 (30–71)	47 (40–75)
≥65 years old, *n* (%)	2 (18)	1 (11)
Race/ethnicity, *n* (%)		
Caucasian, not Hispanic	6 (55)	3 (33)
African American	0	1 (11)
Hispanic	2 (18)	5 (56)
Asian	2 (18)	0
unknown	1 (9)	0
Sites of infection, *n* (%)[a]		
rhino-orbital	5 (45)	6 (67)
lung	6 (55)	2 (22)
hepatic	0	1 (11)
disseminated	1 (with lung) (9)	1 (with hepatic) (11)
Proven infection, *n* (%)[a]	9 (82)	9 (100)
positive histopathology, *n* (%)	8 (73)	7 (78)
positive culture, *n* (%)	7 (64)	7 (78)
Rhizopus spp., *n* (%)	1 (9)	2 (22)
Rhizopus oryzae, *n* (%)	0	1 (11)
Rhizopus microsporus, *n* (%)	3 (27)	1 (11)
Cunninghamella spp., *n* (%)	1 (9)	0
unknown, *n* (%)	2 (18)	3 (33)
Probable infection, *n* (%)	2 (18)	0
positive PCR, *n* (%)	1 (9)	
positive culture, *n* (%)[b]	1 (9)	
Diabetes, *n* (%)	7 (64)	6 (67)
with no other risk factors for mucormycosis, *n* (%)	1 (9)	3 (33)
Active malignancy, *n* (%)	7 (64)	3 (33)
solid, *n* (%)	1 (9)	1 (11)
haematopoietic, *n* (%)	6 (55)	2 (22)
Neutropenia, *n* (%)	4 (36)	1 (11)
Corticosteroids, *n* (%)	7 (64)	4 (44)
History of previous transplant, *n* (%)	5 (45)	4 (44)
solid organ, *n* (%)	2 (18)	2 (22)
haematopoietic, *n* (%)	3 (27)	2 (22)
Graft-versus-host disease, *n* (%)	1 (9)	1 (11)
HIV positive, *n* (%)	1 (9)	0
Antifungal therapy prior to enrolment, *n* (%)[a]	9 (82)	7 (78)
amphotericin B deoxycholate, *n* (%)	0	1 (11)
LAmB, *n* (%)	3 (27)	3 (33)
amphotericin B lipid complex, *n* (%)	1 (9)	1 (11)
posaconazole, *n* (%)	4 (36)	1 (11)
echinocandin, *n* (%)	4 (36)	3 (33)
voriconazole, *n* (%)	3 (27)	2 (22)
fluconazole, *n* (%)	2 (18)	0

Continued

Table 1. *Continued*

Characteristic	Deferasirox ($n=11$)	Placebo ($n=9$)
Enrolment by site, *n* (%)		
UCSF	4 (36)	3 (33)
University of Miami Jackson Memorial Medical Center	1 (9)	3 (33)
MD Anderson Cancer Center	3 (27)	1 (11)
Duke University Medical Center	1 (9)	2 (22)
Fred Hutchinson Cancer Research Center	2 (18)	0

[a]Patients could be counted more than once.
[b]Positive for *Lichtheimia* from BAL.

compared by Fisher's exact or χ^2 tests, as appropriate. Time-to-event analyses used the log rank test.

Results

Patients

Twenty patients ($n=11$ deferasirox and $n=9$ placebo) were enrolled from five study sites (Table 1). Five of six (83%) patients enrolled at cancer centres were randomized to the deferasirox arm. The patients in each arm had similar demographics, but imbalances were evident in their clinical characteristics (Table 1). Eleven (55%) patients had rhino-orbital infections, 8 (40%) had pulmonary infections and 1 had gastrointestinal (liver) infection; 2 (10%) had disseminated infections, 1 of whom had co-existing pulmonary infection and 1 of whom had the gastrointestinal infection. Of the eight pulmonary infections, six (75%) patients were in the deferasirox arm and two (25%) in the placebo arm.

Eighteen (90%) patients had proven infection and 2 (10%) had probable infection, established in 1 patient each by PCR and by culture from BAL (i.e. non-sterile site). Fourteen (70%) patients had positive cultures (including one probable case). *Rhizopus* was the most common genus isolated, causing eight (57%) of the culture-positive cases; identification of strains was not available for five (36%) culture-positive patients.

Diabetes was the most common risk factor for mucormycosis and was present in 13 (65%) patients (Table 1). However, diabetes was typically accompanied by other risk factors and was the only risk factor for mucormycosis in only four (20%) patients, three of whom were in the placebo arm. Of the 10 (50%) patients with active malignancy, 7 (70%) were in the deferasirox arm and 3 (30%) in the placebo arm. Most (8/10) active malignancies were haematopoietic in origin. Five patients (25%) were neutropenic at baseline, of whom four (80%) were in the deferasirox arm and one (20%) was in the placebo arm. Of the 11 (55%) patients receiving corticosteroid therapy, 7 (64%) were in the deferasirox arm and 4 (36%) were in the placebo arm. Nine of those patients were recipients of transplantation of solid organs ($n=4$) or haematopoietic stem cells ($n=5$).

Study therapy

All randomized patients underwent surgical debridement and all received at least one dose of the blinded study medication. However, only six (55%) patients in the deferasirox arm received at least four doses of the study medication and were included in the pre-specified PP population, versus eight (89%) patients in the placebo arm (Table 2). The median duration of study medication therapy in the deferasirox arm versus the placebo arm was 4 versus 14 days ($P=0.04$). Only four patients (36%) in the deferasirox arm completed 14 days of therapy, versus seven (78%) patients in the placebo arm. All patients were treated with daily LAmB and the daily dose was similar between the two arms (Table 2).

Of the six patients who did not receive at least four doses of the study medication, two withdrew because of inability to tolerate oral medications (one in the deferasirox arm and one in the placebo arm), three had consent withdrawn due to progression of underlying disease and unremitting infection (all in the deferasirox arm) and one patient in the deferasirox arm was withdrawn because of new-onset acute renal failure in the setting of solid organ transplantation, resulting in a change from LAmB to posaconazole-based therapy (Table 3).

Table 2. Treatment while on study

Characteristic	Deferasirox ($n=11$)	Placebo ($n=9$)	*P* value
ITT, *n*[a]	11	9	
PP, *n* (%)[a]	6 (55)	8 (89)	0.1
Blinded study medication			
median (range) duration, days	4 (1–14)	14 (3–14)	0.04
total receiving 1–3 days, *n* (%)	5 (45)	1 (11)	
total receiving 4–13 days, *n* (%)	2 (18)	1 (11)	
total receiving 14 days, *n* (%)	4 (36)	7 (78)	
LAmB, median (range) dose, mg/kg/day	7.5 (5–20)	8 (4–22)	
Surgical debridement, *n* (%)	11 (100)	9 (100)	
Concomitant antifungal therapy, *n* (%)[b]	6 (55)	7 (78)	0.3
posaconazole, *n* (%)	3 (27)	4 (44)	0.4
echinocandin, *n* (%)	5 (45)	4 (44)	0.7

[a]ITT = all randomized patients and PP = all randomized patients receiving at least four doses of blinded study medication.
[b]Two patients in the deferasirox arm and one patient in the placebo arm were treated with both posaconazole and an echinocandin (i.e. triple therapy).

Table 3. Summary of patients terminating study prior to receipt of four doses of study medication

Study arm	Duration of blinded therapy (days)	Reason for discontinuation
Deferasirox	1	withdrew due to nausea and vomiting
Deferasirox	2	patient's family withdrew consent due to disease progression
Deferasirox	2	patient withdrew consent due to disease progression
Deferasirox	2	patient withdrawn by the investigator due to rising creatinine
Deferasirox	3	patient was changed to comfort care and withdrew from the study
Placebo	2	withdrew due to nausea and vomiting

Concomitant, non-study antifungal therapy was common (55% in the deferasirox arm and 78% in the placebo arm) despite protocol prohibitions on such therapy during the period of study medication administration (Table 2). Of the patients who received non-study antifungal therapy, 4/6 (66%) died in the deferasirox arm while only 2/7 (29%) died in the placebo arm.

Safety analyses

There were no apparent differences in either the number of patients with serious adverse events (SAEs), the total number of SAEs or the types of SAEs reported between the two arms (Table 4). There was also no apparent difference in the number or type of non-SAEs (Table 4). Further, there were no apparent differences in laboratory values between the two arms over time, including creatinine, liver function tests, blood counts and tacrolimus levels (Table S1, available as Supplementary data at *JAC* Online). However, there were more deaths in the deferasirox arm at days 30 and 90 (Table 4), and the time to death was shorter in the deferasirox arm by Kaplan–Meier analysis ($P=0.02$; Figure 1).

Of the four patients who did complete 14 days of deferasirox therapy, three died. Two of these patients had rhino-orbital disease in the setting of diabetes and two had pulmonary disease in the setting of active malignancy or solid organ transplant—none of these patients was neutropenic. Of the two patients in the deferasirox arm who survived through day 90, one had pulmonary disease in the setting of diabetes, corticosteroids and solid organ transplantation, and one had rhino-orbital disease in the setting of diabetes. Of patients without active malignancy or neutropenia, 2/4 (50%) treated with deferasirox survived versus 5/6 (83%) treated with placebo. Of patients with active malignancy, 0/7 (0%) treated with deferasirox survived versus 2/3 (66%) treated with placebo.

Exploratory efficacy analyses

In the ITT population, global success rates were low and similar in both arms at the end of therapy (pre-specified primary efficacy endpoint) (Table 5). Global success rates trended to lower at day 30 in the deferasirox arm. The primary drivers of failure in the deferasirox arm were patient deaths (Table 4) and clinical failure (Table 5). In the PP population, the results were similar, with success rates trending to lower at 30 days in the deferasirox arm (Table 5).

Table 4. Safety of therapy

Characteristic	Deferasirox ($n=11$)	Placebo ($n=9$)	*P* value
SAE, *n* (%)	8 (73)	4 (44)	0.2
SAE, total number	12	7	
progressive infection, *n*	1	0	
progression of underlying disease, *n*	3	0	
respiratory failure/hypoxia, *n*	2	2	
liver failure, *n*	1	0	
non-ST-elevation myocardial infarction, *n*	1	0	
intolerance of study drug/vomiting, *n*	1	1	
renal failure, *n*	1	0	
intracranial bleeding, *n*	1	0	
arrhythmia, *n*	0	1	
alveolar haemorrhage, *n*	0	1	
bacterial pneumonia, *n*	1	1	
fever, *n*	0	1	
Non-SAEs, *n*	8	12	
thrombocytopenia, *n* (%)	0	1 (11)	
exophthalmos, *n* (%)	0	1 (11)	
blurry vision, *n* (%)	0	1 (11)	
diarrhoea, *n* (%)	1 (9)	1 (11)	
vomiting, *n* (%)	0	1 (11)	
hyperbilirubinaemia, *n* (%)	1 (9)	0	
elevated creatinine, *n* (%)	0	1 (11)	
hypoalbuminaemia, *n* (%)	1 (9)	0	
hypokalaemia, *n* (%)	1 (9)	1 (11)	
hypomagnesaemia, *n* (%)	1 (9)	0	
metabolic acidosis, *n* (%)	1 (9)	0	
myoclonus, *n* (%)	0	1 (11)	
somnolence/insomnia, *n* (%)	0	2 (22)	
confusion, *n* (%)	0	1 (11)	
hypoxia, *n* (%)	0	1 (11)	
respiratory distress, *n* (%)	1 (9)	0	
rash, *n* (%)	1 (9)	0	
Death at			
30 days, *n* (%)	5 (45)	1 (11)	0.1
90 days, *n* (%)	9 (82)	2 (22)	0.01

Pharmacokinetic/pharmacodynamic results

Deferasirox MICs for the seven strains available for testing (five from the deferasirox arm and two from the placebo arm) ranged from 0.78 to 12.5 mg/L, consistent with previous results.[1] Due to concerns about oral absorption in acutely ill patients with mucormycosis, exploratory pharmacokinetic

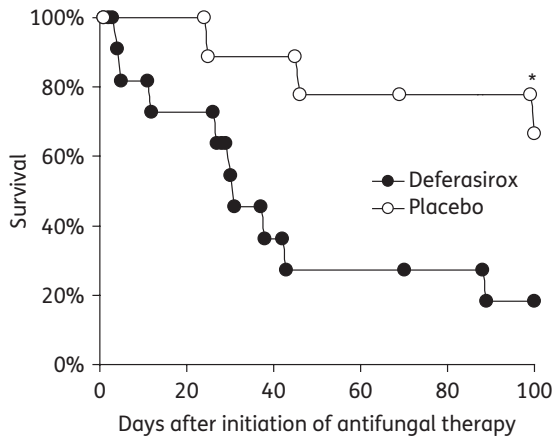

Figure 1. Time to death of patients randomized to deferasirox versus placebo. All patients were followed through 90 days, with one death in the placebo arm captured on follow-up of an SAE at day 100. *$P=0.02$.

Table 5. Outcomes

Characteristic	Deferasirox ($n=11$)	Placebo ($n=9$)	*P* value
ITT population			
Global success at EOT, *n* (%)	3 (27)	3 (33)	1
Global success at 30 days, *n* (%)	2 (18)	6 (67)	0.06
Global success at 90 days, *n* (%)	2 (18)	5 (56)	0.2
Clinical response, *n* (%)			
at EOT	7 (64)	9 (100)	0.08
at 30 days	4 (36)	8 (89)	0.02
at 90 days	2 (18)	7 (78)	0.01
Radiographic response, *n* (%)			
at EOT[a]	4 (36)	3 (33)	0.6
PP population			
Global success, *n* (%)			
at EOT	2/6 (33)	3/8 (38)	0.7
at 30 days	1/6 (17)	6/8 (75)	0.05
at 90 days	1/6 (17)	4/8 (50)	0.2
Survival, *n* (%)			
at EOT	6/6 (100)	8/8 (100)	1
at 30 days	4/6 (67)	8/8 (100)	0.2
at 90 days	1/6 (17)	7/8 (88)	0.02
Clinical response, *n* (%)			
at EOT	5/6 (83)	8/8 (100)	0.5
at 30 days	3/6 (50)	8/8 (100)	0.06
at 90 days	1/6 (17)	7/8 (88)	0.02
Radiographic response, *n* (%)			
at EOT[a]	2/6 (33)	3/8 (38)	0.7

EOT=end of therapy with blinded study medication.
[a]Because the protocol only required a follow-up CT/MRI scan at EOT, very few follow-up scans were available beyond EOT.

studies were conducted. Serum for deferasirox levels was available from only three patients after the seventh dose of study therapy (only four patients received 7 days of deferasirox therapy; pharmacokinetic samples were unavailable for one patient). Peak serum deferasirox concentrations ranged from 35 to 45 mg/L (Figure S1, available as Supplementary data at *JAC* Online). By 20–24 h post-dose, trough levels ranged between 9 and 22 mg/L.

Discussion

The DEFEAT Mucor study was the first randomized trial conducted of any treatment strategy for mucormycosis, or indeed for any non-*Aspergillus* mould infection. Although SAEs and AEs were similar in frequency and severity in patients treated with deferasirox and placebo, by 90 days of follow-up the mortality rate was higher in patients treated with deferasirox than in those treated with placebo. The primary driver of excess mortality appeared to be clinical failure due to progression of infection and progression of underlying disease, rather than any specific toxicity. No subset was identified that had superior outcomes with deferasirox therapy versus placebo, including patients who completed all 14 days of study therapy or those without malignancy or neutropenia.

There are several possible explanations for the discrepancy between the promising efficacy of deferasirox seen in earlier studies and the current results. First, despite its fungicidal effects on Mucorales *in vitro* and substantial efficacy in mice, deferasirox may have no impact on clinical disease or may even worsen the clinical course of mucormycosis in humans. There are no published reports of deferasirox antagonism with other antifungal agents and the drug does not promote fungal growth *in vitro* or in mice.[1] Hence, mechanisms by which the drug could worsen clinical infection are not clear. Second, efficacy of deferasirox was most apparent in mice with DKA, which have increased free iron levels, and efficacy was substantially less apparent in neutropenic mice.[1] Similarly, in the open-label description of favourable clinical outcomes of adjunctive deferasirox therapy (including initial therapy), most patients were diabetic and/or receiving corticosteroids, and no patients were neutropenic.[19] Very few patients in the current study had diabetes as their only risk factor for mucormycosis and it is possible that the results of the current study reflect diminished deferasirox efficacy in specific host populations (e.g. neutropenia, haematological malignancy etc.). As well, data were not available regarding the time from onset of symptoms to the initiation of antifungal therapy, which is now known to be a major predictor of survival from mucormycosis.[21]

Probably due to the small number of enrolled patients spread across five sites with highly heterogeneous patient populations, there were imbalances at the time of randomization in the underlying disease and risk factors in the deferasirox and placebo arms. Most of the patients enrolled at cancer centres were randomized to receive deferasirox therapy. As a result, more patients in the deferasirox arm had malignancy, including haematological malignancy and/or myelodysplastic syndrome and neutropenia. Furthermore, the majority of patients with pulmonary infection were randomized to the deferasirox arm. It is well established that clinical outcomes are worse and mortality higher in patients with pulmonary mucormycosis versus rhino-orbital infection, in patients with malignancy versus diabetes as the predominant risk factor for mucormycosis

and in patients with prolonged bone marrow suppression.[21–25] Thus, imbalances in unmeasured confounders as well as recognized imbalances in underlying diseases and site of infection make it challenging to interpret different survival and treatment success rates in the deferasirox versus placebo arm. The small number of patients enrolled precludes a multifactorial analysis to isolate the contribution of treatment versus baseline factors to outcomes. Nevertheless, future studies of mucormycosis should carefully stratify patients by underlying disease (diabetic and solid organ transplantation versus haematological malignancy, neutropenia and haematopoietic stem cell transplantation) and study site.

Peak and trough deferasirox serum levels were similar to previously published results from patients treated for transfusion-related iron overload, in whom mean peak and trough serum levels were 38 and 17 mg/L, respectively.[26] Serum levels were in excess of the deferasirox MICs for the small number of fungal isolates available for testing. Small sample sizes preclude definitive conclusions regarding any pharmacokinetic/pharmacodynamic relationship with clinical outcome.

The DEFEAT Mucor study enrolled slowly, despite broad enrolment criteria. On average, 0.9 patients per month were enrolled, with enrolment open in at least five sites at any one time, resulting in a conservative estimate of an average enrolment rate of 0.15 patients per site per month. The slow enrolment rate underscores the need for any future study of mucormycosis to enrol in a large number of study sites to achieve large sample sizes in reasonable periods of time, including the possible use of sites in Europe, where the disease is rising in incidence,[27] and in India, where some hospitals see ≥50 patients with mucormycosis per year.[28,29]

Concordant with studies in mice,[30] recent retrospective studies have demonstrated a superior survival rate in patients with mucormycosis treated with LAmB versus amphotericin B deoxycholate.[23,24,31,32] Survival in patients treated with LAmB has ranged from 40% to 70% in these studies, with the primary drivers of survival relating to underlying predisposing risk factors (mortality worse with haematopoietic malignancies and prolonged neutropenia). These retrospective comparisons appear validated by the surprisingly high treatment success and survival rates in patients treated with LAmB (i.e. the placebo arm) in the current study. These results are critical for planning future pivotal studies of novel treatment strategies for mucormycosis and support published expert opinion that LAmB is preferred to amphotericin B deoxycholate for the treatment of mucormycosis.[22]

In summary, given the excess mortality of patients treated with adjunctive deferasirox therapy, deferasirox cannot be recommended as part of an initial combination regimen for the treatment of mucormycosis. Although no obvious toxicities were identified, the limited sample size precludes definitive establishment of the safety of deferasirox therapy for mucormycosis. Imbalances in underlying diseases at the time of randomization may have accounted for treatment efficacy differences between the study arms. Only a large, Phase III trial, potentially enrolling only diabetic or corticosteroid-treated patients, and excluding cancer/neutropenia patients, could further elucidate the safety and efficacy of initial, adjunctive deferasirox for the treatment of mucormycosis. Furthermore, these data do not address the potential for deferasirox to be used as part of a combination salvage therapy regimen, as early pre-emptive therapy in patients with less advanced disease identified by novel biomarkers (e.g. PCR) or as prophylaxis in high-risk patients. However, until additional studies are conducted, caution should be used when administering deferasirox for patients with mucormycosis, even in a salvage setting.

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Author contributions

B. S. designed the study, served as the Principal Investigator (PI) and helped draft the manuscript. A. S. I. performed the susceptibility and microbiology testing, and helped draft the manuscript. P. V. C.-H. helped design the study, served as the site PI at UCSF and helped draft the manuscript. D. P. K. helped design the study, served as the site PI at MD Anderson and helped draft the manuscript. M. I. M. helped design the study, served as the site PI at the University of Miami and helped draft the manuscript. J. R. P. helped design the study, served as the site PI at Duke and helped draft the manuscript. D. F. served as the site PI at the Fred Hutchison Cancer Research Center and helped draft

the manuscript. E. P. B. designed the study and helped draft the manuscript.

Supplementary data

Table S1 and Figure S1 are available as Supplementary data at *JAC* Online (http://jac.oxfordjournals.org/).

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