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CASE REPORT – OPEN ACCESS

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Rhino-Orbito-Cerebral Mucormycosis Resistant to Amphotericin B: Two Diabetic Cases

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ABSTRACT

Mucormycosis is a rare, life-threatening opportunistic infection caused by Mucorales. The most common organisms that cause mucormycosis in humans are *Rhizopus*, *Mucor*, *Rhizomucor*, *Lichtheimia (Absidia)*, and *Cunninghamella*. Risk factors for the disease include diabetes mellitus, metabolic acidosis, hematologic malignancies, prolonged severe neutropenia, use of corticosteroids, and human immunodeficiency virus infection. Diagnosis and treatment of infection are difficult.

Keywords: Mucormycosis, *rhizopus*, amphotericin resistant, diabetes mellitus

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INTRODUCTION

Mucormycosis is a severe infection with high mortality rate due to the angioinvasive character and rapid progression of disease. Patients with diabetes mellitus (DM) have markedly increased incidence of invasive mucormycosis. Despite the advancement of new antifungal drugs and combination therapies, the management of mucormycosis is still challenging (1, 2). Here we presented the two cases of mucormycosis with a history of diabetic ketoacidosis. Both cases were infected by *Rhizopus* spp. and resistant to amphotericin. Unfortunately, one patient died despite early surgical debridement and medical treatment.

CASE REPORTS

Case 1

A 58-year-old female patient was transferred from another hospital's endocrinology clinic, where she was hospitalized due to diabetic ketoacidosis due to a swelling and redness on the left area of her face. She was living in a rural area. Her medical history showed that she had uncontrolled DM for 10 years. Initially, she was treated with beta-lactam group of antibiotics for preseptal cellulitis. However, lesion width and C-reactive protein values continued to increase at 48 h of treatment. The antibiotic therapy was changed to meropenem 6 g/day. Despite the treatment, the patient's fever was increasing. The patient was then transferred to our clinic. At admission, she had a body temperature of 38.7°C, and her Glasgow Coma Scale was 12–13. She had redness and edema on the left maxillary region and eyelid. Results of laboratory investigations are shown in Table 1. Paranasal sinus tomography was performed and revealed only maxillary sinusitis on the left side. As necrotic black lesions on the upper palate (Fig. 1) appeared on endoscopic sinus examination, liposomal amphotericin B (5 mg/kg/day intravenous) was added to the treatment. Surgical debridement was urgently performed for necrotic areas, and tissue biopsy material was sent for direct microscopic examination to the mycology laboratory. The biopsy material was stained with Gram and Giemsa stains and cultured in Sabouraud dextrose agar (SDA) medium with and without antibiotics (cycloheximide and chloramphenicol). Then, it was incubated at 37°C and 25°C. KOH mount and staining prepared from the tissue revealed broad aseptate hyphae. Fast-growing hyphae covered the agar surface quickly with dense, cotton candy-like growth. At first white and then gray colonies were grown on SDA except SDA with cycloheximide. Slide culture on potato dextrose agar showed broad hyphae without septa, the presence of hyaline stolons, brown pigmented root-like hyphae (rhizoids), and sporangiophores that were long and terminate with a dark, round sporangium. According to the criteria based on slide culture and colony color, the mold was identified as *Rhizopus* sp. The identification was confirmed as *Rhizopus oryzae* by the internal transcribed spacer region (including 5.8S rRNA gene nucleotide sequencing and sequence similarity searching using BLAST) in the National Center for Biotechnology Information database (3). An antifungal susceptibility test was performed for amphotericin B, voriconazole, and itraconazole by the microdilution method according to the Clinical and Laboratory Standards Institute document M38-A2 (4) and for caspofungin, anidulafungin, and posaconazole by the E test method (5). The minimum inhibitory concentration (MIC) values are shown in Table 1. On bacterial culture, the tissue culture specimens obtained from surgical debridement yielded Pseudomonas aeruginosa. Posaconazole

Table 1. Results of laboratory investigations

			Minimum Inhibitory Concentration (MIC) for antifungals (μg/mL)				
Cases	**WBC**	**CRP**	**Amphotericin B**	**Itraconazole**	**Caspofungin**	**Anidulafungin**	**Posaconazole**
Case 1	20,2	203	32	32	>32	>32	1
Case 2	41.5	440	8	32	>32	>32	2

MIC: Minimum inhibitory concentration; WBC: White blood cells; CRP: C-reactive protein

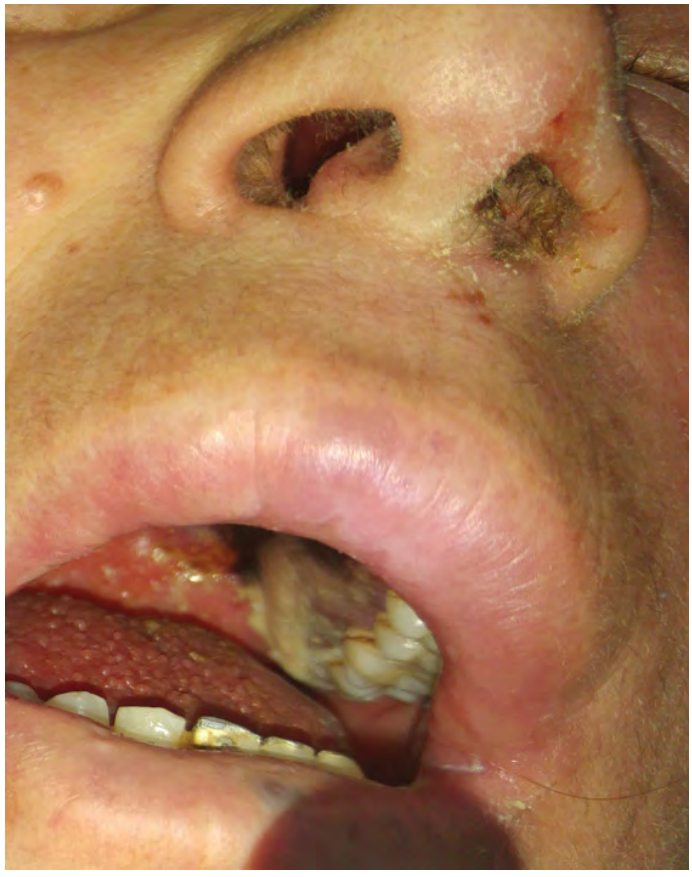

Figure 1. The necrotic lesion on upper palate

tablet (loading dose of 300 mg twice on day 1 and then 300 mg/day) and ciprofloxacin (400 mg/day intravenous) were added to the treatment according to the susceptibility results. The blood glucose level of the patient was closely monitored, and intensive insulin therapy was given. Control cranial tomography revealed inflammation of the left eye and optic nerve. A second surgery was performed as left eye enucleation and extensive debridement. After 3 weeks of treatment, she suffered from headache. Cranial magnetic resonance imaging scan revealed septic emboli in the left cerebellar hemisphere and temporal lobe (Fig. 2). Anticoagulant therapy was added. After 8 weeks of intravenous treatment, she was discharged with oral posaconazole treatment. At 6 months of therapy, control tomography was normal with clean surgical margins.

Case 2

A 55-year-old male patient presented in the emergency department with complaints of pain and swelling on the right maxillary area for 5 days and loss of vision on his right eye for 2 days. He

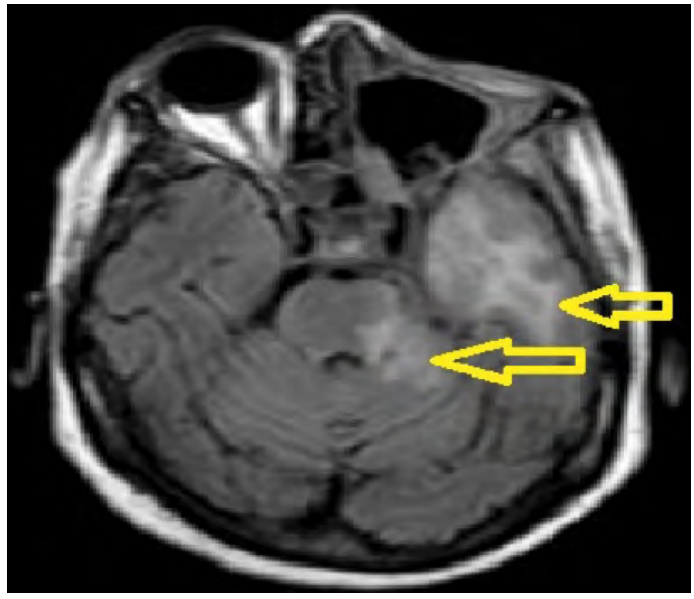

Figure 2. Septic emboli in the left cerebellar hemisphere and temporal lobe

was taking oral amoxicillin clavulanate (3 g/day) treatment for 2 days with a diagnosis of cellulite. Despite this treatment, the pain on his face became worse. He was a known case of type 2 DM for a few months with poorly controlled blood glucose. On physical examination at the time of admission, his vital signs were stable, and Glasgow Coma Scale was 15. Results of laboratory investigations are shown in Table 1. On endoscopic examination, there was hyperemia in the upper palate. Liposomal amphotericin B (5 mg/kg/day intravenous) was urgently initiated. Brain computer tomography showed sinusitis on the right maxillary, frontal, ethmoid, and sphenoid sinus; right preseptal cellulitis; and inflammation on the right optic nerve. Optic nerve and orbital decompression were performed urgently. Tissue culture was sent to the microbiology laboratory. KOH mount and staining prepared from the tissue revealed broad aseptate hyphae. The procedures described in the first case were performed in the same order. Cultures obtained from tissue samples yielded *Rhizopus* spp. According to antifungal susceptibility testing, it was resistant to amphotericin B, itraconazole, and echinocandins and dose-dependent sensitive to posaconazole. The MIC values are shown in Table 1. Posaconazole intravenous (loading dose of 300 mg twice on day 1 and then 300 mg/day) was subsequently added to the therapy. Blood glucose level was closely monitored, and intensive insulin therapy was given. The patient was re-operated on day 3 of the treatment due to the progression of former lesions. He had an enucleation in his right eye and also underwent right maxillectomy. On day 10 of mechanical venti-

Table 2. Epidemiological characteristics, predisposing factors, treatment and outcome of patients with infected by Amphotericin B-resistant rhinocerebral mucormycosis

Author, year	Age	Gender	Underlying disease	Antifungal treatment	Surgery	Prognosis
Barnert et al., 1985 (14)	51	Male	Diabetic ketoacidosis	Miconazole, ketoconazole	Yes	Survived
Vyzantiadis et al., 2009 (15)	78	Male	Idiopathic thrombocytopenic purpura treated with methylprednisolone	Liposomal amphotericin B, posaconazole	Yes	Died
Biswas et al., 2015 (16)	45	Female	Diabetes mellitus	No treatment	Yes	Died
Present case 1	58	Female	Diabetic ketoacidosis	Liposomal amphotericin B, Posaconazole	Yes	Survived
Present case 2	55	Male	Diabetic ketoacidosis	Liposomal amphotericin B, Posaconazole	Yes	Died

lation, ventilator-associated pneumonia developed due to Acinetobacter baumannii and Pseudomonas aeruginosa. Colistin (300 mg/day) and cefepime (6 g/day) combination therapies were also added. Control paranasal tomography revealed the persistence of sinusitis on the left maxillary and sphenoid sinus. He could not be operated due to septic shock. The patient died on day 12 of the treatment.

DISCUSSION

Rhinocerebral mucormycosis is the most common form of mucormycosis for patients with DM or ketoacidosis (1). The diagnosis of the disease is crucial because the symptoms are nonspecific and silent. The most common symptoms are unilateral periorbital pain, eyelid edema, headache, and acute vision loss. In the differential diagnosis, periorbital or facial cellulitis and zona or herpes zoster ophthalmicus should be considered (6).

Multidisciplinary approach in the treatment of mucormycosis control of underlying conditions, surgical, and medical treatment is reduced mortality. Surgical debridement is recommended at AII level. Liposomal amphotericin B is recommended for first-line therapy at BII level and posaconazole as a salvage treatment or as an alternative treatment for refractory agents (7). Amphotericin B is known to be the most active drug against *Mucorales* and suggested as backbone therapy for mucormycosis. Unfortunately, the sensitivity of amphotericin B is variable among *Mucorales* spp. Generally, *Rhizopus* spp. have higher MICs against amphotericin B (8). On the other hand, it is known that azoles used in agriculture cause cross-resistance between other medical azoles (9). The development of cross-resistance in some fungi, such as Candida spp., Cryptococcus neoformans, and Fusarium spp., is discussed in the literature (10–12). There is no evidence for *Mucor* strains. However, the patient presented in the first case report was a farmer and was using antifungal agents for apples. This suggests that cross-resistance between fungicides used in agriculture and amphotericin B resistance is developed.

The second recommended agents include echinocandins, posaconazole, and deferasirox (7, 13). Although there are some studies suggesting that combination therapies are superior, large-scale studies are still needed. The limited number of cases with amphotericin B-resistant mucormycosis in the literature is shown in Table 2 (14–16). In these cases, the azole group of antifungals was used according to sensitivity, but there is no recommendation in the current guidelines. These reported cases may be helpful for the selection of combination therapy. Additionally, surgical debridement is becoming more important.

Another problem in patients with mucormycosis is bacterial superinfections. Microorganisms that colonize the respiratory tract may cause secondary bacterial infections, or nosocomial infections may develop as most patients require long stay in intensive care. Secondary bacterial infections can lead to mortality even if mucormycosis treatment is managed successfully.

Two cases of polyene-resistant mucormycosis that are rare in the literature are presented here to draw attention to the difficulties in treatment.

Informed Consent: Written informed consent was obtained from the first patient and the second patient's wife.

Peer-review: Externally peer-reviewed.

Author Contributions: Concept – EE, EA; Design – EE, AUK; Supervision – EE, AUK; Resource – EE; Materials – MAA, NK; Data Collection and/or Processing – EE, SMB; Analysis and/or Interpretation – EE, EA; Literature Search – EE, SMB; Writing – EE, AUK; Critical Reviews – AUK, MAA.

Conflict of Interest: The authors have no conflict of interest to declare.

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REFERENCES

1. Riley TT, Muzny CA, Swiatlo E, Legendre DP. Breaking the Mold: A Review of Mucormycosis and Current Pharmacological Treatment Options. Ann Pharmacother 2016; 50(9): 747–57. [CrossRef]
2. Rai S, Yadav S, Kumar D, Kumar V, Rattan V. Management of rhinomaxillary mucormycosis with Posaconazole in immunocompetent patients. J Oral Biol Craniofac Res 2016; 6(Suppl 1): S5–S8. [CrossRef]
3. http://blast.ncbi.nlm.nih.gov/Blast.cgi. Accessed Jul 25, 2019.
4. Document M38-A2. Reference method for broth dilution antifungal susceptibility testing of filamentous fungi; approved standard–second edition. Wayne, PA: CLSI; 2008.
5. Lamoth F, Alexander BD. Comparing Etest and Broth Microdilution for Antifungal Susceptibility Testing of the Most-Relevant Pathogenic

Molds. J Clin Microbiol 2015; 53(10): 3176–81. [CrossRef]
6. Kosmidis C, Denning D.W. Opportunistic and Systemic Fungi. Cohen J, Powderly WG, Opal SM, editors. Infectious Disease; 2017. p. 1696–8. [CrossRef]
7. Tissot F, Agrawal S, Pagano L, Petrikkos G, Groll AH, Skiada A, et al. ECIL-6 guidelines for the treatment of invasive candidiasis, aspergillosis and mucormycosis in leukemia and hematopoietic stem cell transplant patients. Haematologica 2017; 102(3): 433–44. [CrossRef]
8. Dannaoui E. Antifungal resistance in mucorales. Int J Antimicrob Agents 2017; 50(5): 617–21. [CrossRef]
9. Faria-Ramos I, Farinha S, Neves-Maia J, Tavares PR, Miranda IM, Estevinho LM, et al. Development of cross-resistance by Aspergillus fumigatus to clinical azoles following exposure to prochloraz, an agricultural azole. BMC Microbiol 2014; 14: 155. [CrossRef]
10. Kelly SL, Lamb DC, Kelly DE, Manning NJ, Loeffler J, Hebart H, et al. Resistance to fluconazole and cross-resistance to amphotericin B in Candida albicans from AIDS patients caused by defective sterol delta5,6-desaturation. FEBS Lett 1997; 400(1): 80–2. [CrossRef]
11. Joseph-Horne T, Hollomon D, Loeffler RS, Kelly SL. Cross-resistance to polyene and azole drugs in Cryptococcus neoformans. Antimicrob Agents Chemother 1995; 39(7): 1526–9. [CrossRef]
12. Al-Hatmi AM, Meis JF, de Hoog GS. Fusarium: Molecular Diversity and Intrinsic Drug Resistance. PLoS Pathog 2016; 12(4): e1005464.
13. Spellberg B, Ibrahim A, Roilides E, Lewis RE, Lortholary O, Petrikkos G, et al. Combination therapy for mucormycosis: why, what, and how? Clin Infect Dis 2012;54 Suppl 1: S73–8. [CrossRef]
14. Barnert J, Behr W, Reich H. An amphotericin B-resistant case of rhinocerebral mucor mycosis. Infection 1985; 13(3): 134–6. [CrossRef]
15. Vyzantiadis TA, Kioumi A, Papadakis E, Braimi M, Dermitzakis E, Tsitouridis I, et al. Rhino-cerebral zygomycosis resistant to antimycotic treatment: a case report. Mycoses 2009; 52(1): 87–90. [CrossRef]
16. Biswas D, Kotwal A, Kakati B, Ahmad S. Amphotericin B Resistant Apophysomyces elegans Causing Rhino-oculo-Cerebral Mucormycosis in an Immunocompetent Host. J Clin Diagn Res 2015; 9(8): DD01-2.