# Disseminated cerebral aspergillosis complicated by thrombotic microangiopathy

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## ARTICLE INFO

## ABSTRACT

Invasive aspergillosis (IA) is a serious condition that can affect almost any organ. Cerebral aspergillosis itself is rapidly fatal without treatment. We report a case of disseminated cerebral IA in a patient exposed to cyclophosphamide, rituximab and prednisone. This case is unique because: 1) disseminated IA has not been described in anti-glomerular basement membrane glomerulonephritis; 2) IA led to thrombotic microangiopathy with normal ADAMTS13 and 3) voriconazole toxicity necessitated use of isavuconazole for IA treatment.

## 1. Introduction

Invasive aspergillosis (IA) is a rare fungal infection that most often occurs in immunocompromised patients. Mortality rates range up to 88% in those with disseminated disease or CNS involvement [1]. It is difficult to diagnose and detection relies on radiological findings and analysis of deep tissue samples. The European Organization for Research and Treatment of Cancer/Invasive Fungal Infections Cooperative Group and the National Institute of Allergies and Infectious Disease Mycoses Study Group (EORTC/MSG) require that the fungi be isolated from deep tissue cultures or be seen on histopathology for a proven diagnosis of IA [2]. The 2016 Infectious Diseases Society of America guidelines for the diagnosis of Aspergillosis agree with this statement [3]. Unfortunately, tissue samples are often difficult to obtain in critically ill patients and the yield is poor [4]. We present a case that highlights the diagnostic and management challenges associated with disseminated IA.

## 2. Case

A 54-year-old woman with atypical anti-glomerular basement membrane (anti-GBM) glomerulonephritis on treatment with prednisone, cyclophosphamide and rituximab presented to hospital on day 0 with four weeks of abdominal pain in the context of a recent scald burn to her forearm. She was alert, oriented, and afebrile. Her hemoglobin was 94g/L, platelets were normal at $273 \times 10^9$/L, and her

creatinine was at her baseline of 300μmol/L (Fig. 1). A chest radiograph showed free air under the diaphragm. She empirically received piperacillin-tazobactam 2.25g IV q8h, stress dose steroids, and underwent emergent laparotomy for presumed perforation of abdominal viscus. Despite an unrevealing laparotomy, the patient was continued on piperacillin-tazobactam as she remained unwell in hospital. In the week after surgery, she developed delirium and had a tonic-clonic seizure. An MRI brain on day +7 revealed a 1.1 cm left hemisphere rim-enhancing lesion with surrounding vasogenic edema consistent with an abscess (Fig. 2). Platelets steadily dropped post-operatively to $55 \times 10^9$/L. Her antibiotics were changed to meningitic doses of ceftriaxone 2g IV q12h and metronidazole 500mg q8h. Schistocytes, low haptoglobin and high LDH now accompanied a progressive bicytopenia with hemoglobin and platelets reaching nadirs of 55g/L and $8 \times 10^9$/L respectively over the three weeks of hospitalization consistent with microangiopathic hemolytic anemia (MAHA). Repeat MRI of the brain on day +16 showed increasing size of her abscess to 1.2 cm despite fifteen days of broad-spectrum antibiotics (Fig. 1).

CT scan of her chest, abdomen and pelvis at this time revealed multiple ill-defined centrilobular nodules in both upper lung lobes along with a 2.5cmx1.3cm cavitary lesion with an air fluid level in the left upper lobe. The liver showed multiple hypodense areas suspicious for abscesses. Left lung bronchial washings from a bronchoscopy on day +42 grew Candida parapsilosis and a Geotrichum spp. Serum and bronchial fluid galactomannan antigen testing were negative. Fungal cultures of two separate ultrasound-guided liver biopsies of the
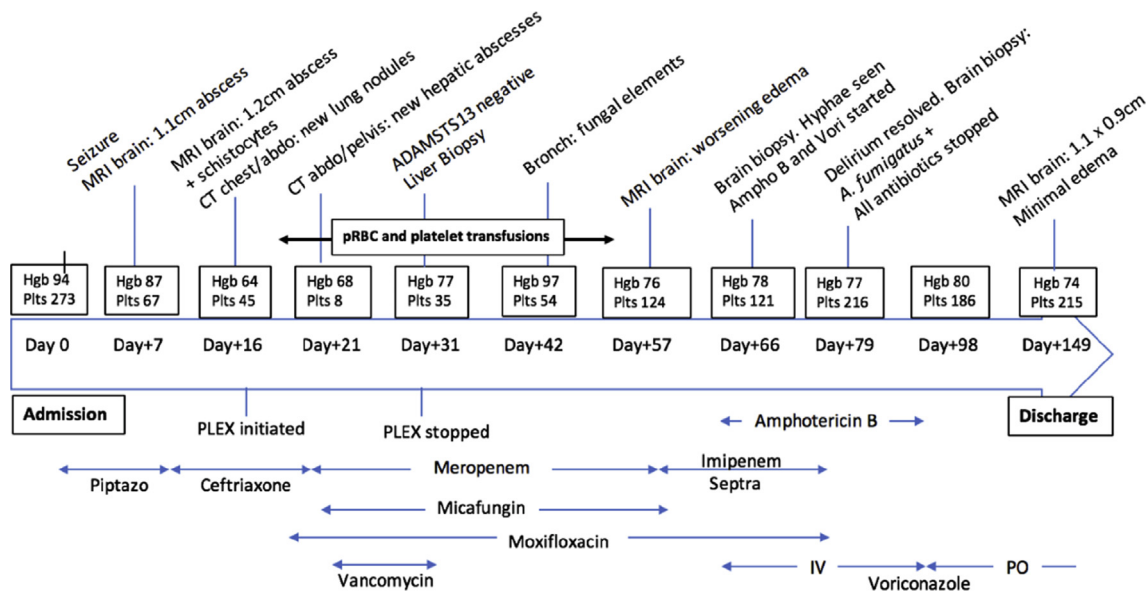
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**Fig. 1.** Timeline of events, antibiotics and antifungals in hospital until first discharge to rehab. Hbg: hemoglobin; Plts: platelets; Piptazo: piperacillin-tazo-bactam; Ampho B: amphotericin B; Vori: voriconazole.

hypoattenuating lesions were negative. Fungal culture of a skin biopsy of her forearm burn wound grew *Candida parapsilosis* and *Aspergillus fumigatus* on day +73. These organisms were identified using standard culture and identification methods including MALDI-TOF MS analysis for Candida parapsilosis (bioMerieux, Ste. Laurent, Quebec).

Antibiotics were changed to meropenem 2g IV q12h for presumed disseminated *Nocardia* but MAHA persisted and delirium worsened. On day +16, the patient was initiated on Plasma Exchange (PLEX) therapy for possible thrombotic thrombocytopenic purpura (TTP) but it was discontinued after nine sessions when ADAMTS13 came back normal. Antibiotics were further broadened while she was receiving PLEX to include moxifloxacin 400mg IV q24h (improved activity against *Nocardia*) and vancomycin 750mg IV q24h (broader coverage of hospital-acquired infections). Micafungin 100mg IV q24h was also started for possible fungal infection (Fig. 1). After six days of the higher dose, meropenem was continued at a reduced dose of 1g IV q12h for another four weeks. Moxifloxacin, micafungin and vancomycin were continued at the same doses for six weeks, one month and twelve days, respectively.

Despite broad spectrum antimicrobials, the patient remained delirious with worsening MAHA. Repeat brain MRIs showed increasing abscess size with worsening edema (Fig. 2). Neurosurgery performed an urgent brain abscess aspiration on day +66. While an initial KOH stain showed only yeast, the subsequent Calcifluor white stain showed yeast, pseudohyphae and hyphae. Empiric antifungal therapy was started with liposomal amphotericin B 10mg/kg/day (600mg) IV q24 h × 3 days (subsequently decreased to 5mg/kg/day (300mg) IV q24 h × 1 month) and two loading doses of voriconazole 600mg IV q12h (continued at 400mg IV q12 h × 1 month). Two days later, the fungal culture of the brain aspirate showed *Aspergillus fumigatus* using standard culture and identification methods. All broad-spectrum antibiotics were stopped and she was continued on both antifungal agents.

Her delirium resolved within two days and platelet count increased to 227 × 10$^9$/L within two weeks. Repeat MRI after three weeks of this antifungal therapy showed reduction in the size of the brain abscess with decreased regional edema. After one month of IV antifungal therapy, amphotericin B was stopped and voriconazole was changed to an oral regimen of 400mg PO q12h. The patient was discharged to a rehabilitation unit on day +149 and remained on the same dose of oral voriconazole. Follow-up monthly MRI scans up to this point showed progressive resolution of the brain abscess (Fig. 2).

One month later (day +187), she was transferred back to hospital for a small bowel obstruction requiring surgery. Her mobility deteriorated in hospital and pelvic X-rays revealed extensive periosteal reaction suspicious for voriconazole-induced periostitis. A follow-up MRI brain on day +303 revealed worsening vasogenic edema with increased abscess wall thickness. The patient required a switch back to amphotericin B 350mg IV q24h with resultant radiographic improvement. She was then transitioned to isavuconazole with an initial load of six doses of 200mg PO q8h followed by 200mg PO daily. The patient is currently at home and doing well on isavuconazole.

## 3. Discussion

*A. fumigatus* is the most common causative agent of IA followed by *A.flavus*, *A.niger*, and *A.terreus* [2]. *Aspergillus fumigatus* spores typically enter the body via inhalation [5]. Rarer routes of entry include the gastrointestinal tract and the skin [5]. The fungus then disseminates through the bloodstream to invade other organs. In our patient with disseminated IA, the burn on the skin (which was culture positive for *A.fumigatus*) was most likely the portal of entry. The *C. parapsilosis* and *Geotrichium* spp. isolated from the lungs were thought to be contaminants. This case highlights several important aspects in the diagnosis and management of IA.

Traditionally described risk factors for IA include chronic granulomatous disease, stem cell and solid organ transplant, hematological malignancies, chronic obstructive pulmonary disease and acquired immune deficiency syndrome [4]. However, with the ever-increasing scope of new immune suppressing agents, there are more patients that are at risk of IA. As far as we are aware, this is the first case report documenting disseminated cerebral IA in a patient on treatment for *anti*-GBM glomerulonephritis. Rituximab itself has been associated with an increased risk of IA in stem cell transplant recipients [6]. Glucocorticoids have also been linked to IA when taken in prolonged, high-dose regimens [7]. Steroids inhibit transcription of cytokine genes involved in defense against *Aspergillus* and weaken neutrophils attacking its hyphae [10]. While there are fewer reports of cyclophosphamide causing IA in the literature, this medication is frequently used in mouse models to study IA [8]. Cyclophosphamide was shown in mouse models to reduce expression of Dectin-1, an important receptor involved in immune responses to *Aspergillus* [8]. While these exact molecular mechanisms are still being elucidated, it is likely that the combination of
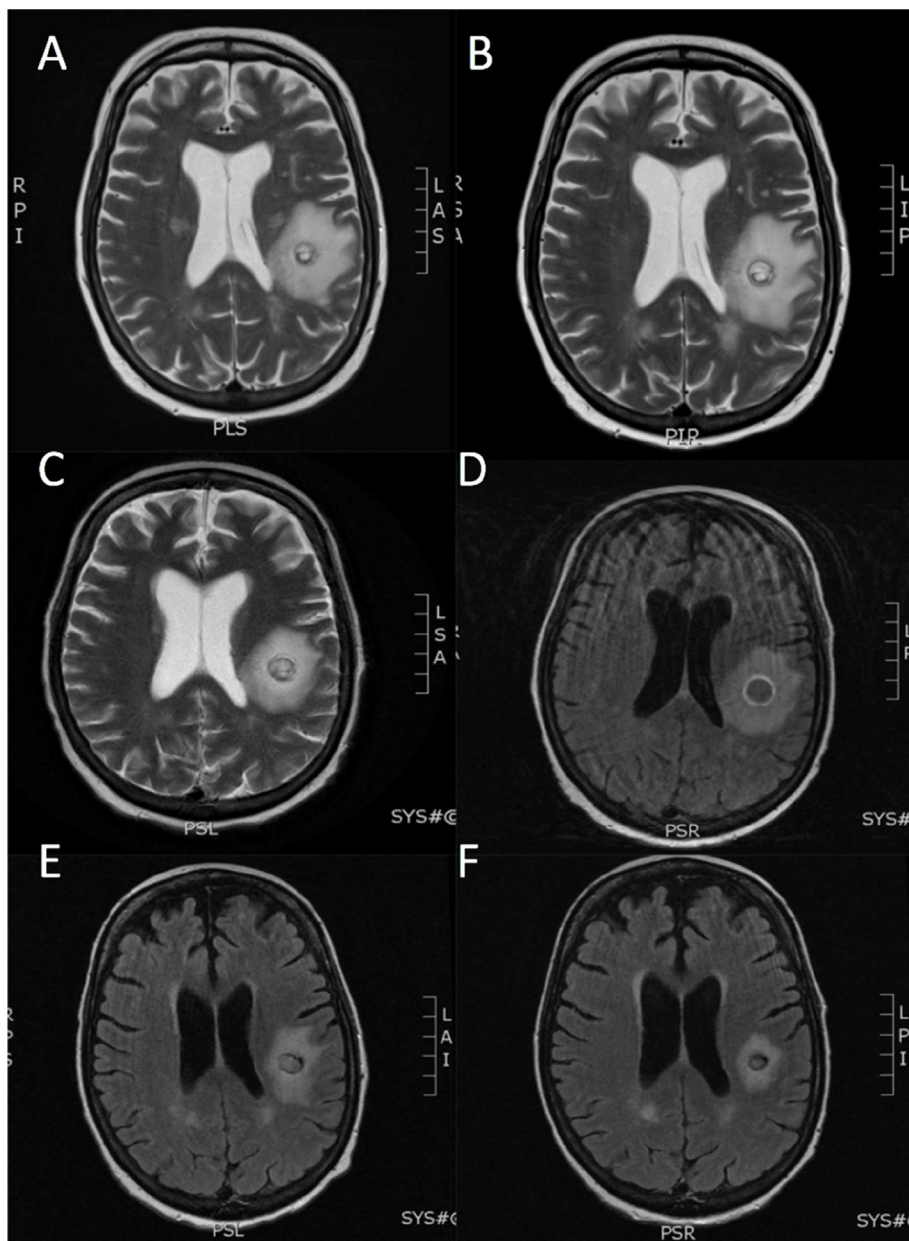
**Fig. 2.** MRI brain at various time points showing progression then improvement once amphotericin B and voriconazole were initiated. **A:** First MRI brain at 1 week post-admission, 1.1cm abscess; **B:** 2 weeks, 1.2cm abscess; **C:** 6 weeks. Size unchanged, progressing edema; **D:** 7 weeks, 1.6cm abscess at time of biopsy; **E:** 12 weeks, 1.2cm abscess. Decreased edema; **F.** 20 weeks, 1.1cm abscess. Significantly decreased edema.

immunosuppressive agents in our patient created an additive effect, increasing her risk for IA.

Guidelines are geared towards established disease with histopathologic demonstration of fungi rather than early recognition. Although biomarkers exist to aid in the diagnosis, their utility is variable. Galactomannan antigen has been validated only in bone marrow transplant and hematologic cancer patients [4]. It has a reported sensitivity of 85% in bronchoalveolar lavage fluid [9] and 29–100% in serum [10]. However, numerous factors affect the accuracy including concurrent antifungal therapy [11]. This explains why our patient who had been on concurrent antifungals had a negative level both in serum and bronchoalveolar lavage fluid.

A striking aspect of this case is the marked MAHA that developed. Her complement levels showed elevated C5–C9 suggesting activation of the alternative complement cascade which is consistent with her disseminated fungal infection. This patient likely had a complement-mediated thrombotic microangiopathy (TMA) secondary to her IA.

Indeed, studies of complement-mediated TMA have shown a similar pattern of complement levels [12]. This is supported by the fact that her blood counts improved significantly once appropriate antifungal therapy was initiated. TTP has been reported in several cases of IA, however TMA with normal ADAMTS13 activity is exceedingly rare. This has important clinical implications, as unlike TTP, our patient's MAHA did not respond to PLEX. There have been a few studies investigating transplant-associated TMA (TA-TMA) and IA but these studies either included TTP under the definition of TA-TMA [13] or did not distinguish ADAMTS13 activity altogether [14].

Voriconazole, a second-generation triazole, is the treatment of choice for IA. However, voriconazole has been reported to cause periostitis largely in stem cell transplant recipients [15]. Fluorosis has known toxic effects on bone causing osteoblast stimulation and periostitis deformans. A significant association between blood fluoride levels and periostitis in those taking voriconazole has been shown and cessation of voriconazole reduces skeletal pain from periostitis [16].

Isavuconazole is a new oral triazole antifungal which is non-inferior to voriconazole for the treatment of IA. Side effects including fluorosis and periostitis have been observed significantly less frequently in isavuconazole compared to voriconazole [17].

Two possible explanations for voriconazole failure in our case are inadequate blood levels and immune reconstitution inflammatory syndrome (IRIS) [18]. The minimal therapeutic level for voriconazole is 1–1.5 µg/mL for most invasive fungal infections, but a higher threshold of 2µg/mL has been recommended for CNS infections [19]. Our patient had six therapeutic drug levels drawn within the month after voriconazole initiation consistent with therapeutic drug monitoring (TDM) recommendations. For the remaining nine months of voriconazole therapy, two drug levels were drawn. One of which, drawn at four months was subtherapeutic at 0.7µg/mL and the second level drawn at eight months was 1.2µg/mL. Although frequency of TDM in those on long-term antifungal therapy is largely based on clinical judgement, there are certain circumstances such as critical illness or CNS infection that may warrant regular TDM [19]. It is important to note that though the levels may appear low, they were enough to induce the side effects.

IRIS is another possible reason for voriconazole failure. IRIS occurred in 25% of neutropenic patients with invasive pulmonary aspergillosis in one study and voriconazole appeared to have a propensity to induce IRIS [20]. Three months following voriconazole initiation, our patient became neutropenic for four months. She also had several prednisone tapers over her year-long hospitalization in the context of recurrent AKI and presumed glomerulonephritis flares. Both these factors could have contributed to an IRIS which may have impacted voriconazole effectiveness.

## 4. Conclusion

Disseminated IA is increasing with the increasing numbers of immunosuppressed people. The current diagnostic challenges as well as high associated mortality means that clinicians need to think about IA early and be aware of potential complications including thrombotic microangiopathy. Long-term treatment with voriconazole is associated with uncertainty in monitoring pattern as well as toxicity. Isavuconazole is a reasonable alternative for treatment of IA.

## Conflicts of interest

There are none.

## Acknowledgements

There are none.

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