A black mould death: A case of fatal cerebral phaeohyphomycosis caused by *Cladophialophora bantiana*

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A B S T R A C T

*Cladophialophora bantiana* is a neurotropic mould and primary cause of cerebral phaeohyphomycoses, which presents with brain abscesses in both immunocompromised and immunocompetent individuals. It is associated with high mortality due to delay in diagnosis and absence of standardised therapy. We present a case of fatal cerebral phaeohyphomycosis in a 67-year-old Caucasian man. Diagnosis was achieved by histopathological examination of brain tissue followed by conventional culture and molecular identification. We highlight diagnostic and treatment challenges involved.

1. Introduction

*Cladophialophora bantiana* is a melanised mould and is the commonest cause of cerebral phaeohyphomycosis. Among human fungal pathogens, it is recognised for its neurotropism and ability to cause infections in immunocompetent hosts. Most reported cases are from the Indian subcontinent [1,2]. Mammalian cases acquired in the UK are extremely rare [3], although imported cases are referred to the National Mycology Reference Laboratory (MRL) Public Health England.

2. Case

A 67-year-old Caucasian man was admitted to a District General Hospital in June 2017, having suffered a generalised tonic-clonic seizure on a flight from the Philippines. Past medical history included hypertension, asthma, coronary artery bypass and untreated quiescent sarcoidosis. Further investigation revealed a two-month history of headache, dizziness, weight loss and diplopia, which was initially investigated at his local hospital with a CT head, which was reportedly normal. Following this, he visited his partner in a rural area of the Philippines, which he did regularly. While overseas his symptoms worsened. A CT head performed in the Philippines showed multifocal lesions in the left cerebral hemisphere. He was given a provisional diagnosis of glioblastoma multiforme and returned to the UK for further investigation and treatment.

He was initially treated with levetiracetam and dexamethasone for seizure control, and co-amoxiclav for presumed aspiration pneumonia. His admission CT head revealed multiple ring enhancing intra-axial lesions within the left frontal lobe, left cerebellar hemisphere and pons with surrounding vasogenic oedema. An MRI confirmed the presence of a chain of well-defined round lesions within the left frontal lobe corona radiata, of low T1 and high T2 signal intensity up to 15mm in axial dimension demonstrating rim enhancement (Fig. 1a, b). Further areas of enhancement were demonstrated in the left cerebellar peduncle and pons with surrounding meningeal enhancement. Surrounding parenchymal oedema was noted with surrounding mass effect with approximately 5mm of midline shift (Fig. 1c, d). He was transferred to our centre for further treatment (day 0).

On admission, he was febrile (38.6°C) with a Glasgow Coma Score (GCS) of 10/15 (E3V2M5), deteriorating to 8/15 (E1V2M5), requiring emergency intubation and transfer to the intensive therapy Unit (ITU). A repeat CT head showed no radiological deterioration. A CT thorax demonstrated bilateral upper lobe consolidation. High dose meropenem (2g TDS) and anti-tuberculous therapy (rifampicin 600mg BD, isoniazid 300mg TDS, ethambutol 1500mg TDS) was commenced.
moxifloxacin 400mg BD, isoniazid 300mg OD and pyrazinamide 2g OD with dexamethasone) were initiated to cover potential intracranial bacterial abscesses and disseminated tuberculosis respectively (Table 1).

He was extubated on day 3 but GCS remained 11/15. He had brisk reflexes bilaterally and an absent vestibulo-ocular reflex. Due to fever spikes, gentamicin was added for 48 hours on day 4 to cover Gram-negative bacteria. Given the diagnostic uncertainty a left frontal lobe biopsy was performed on day 6. His GCS deteriorated post-operatively and he required re-intubation. Despite negative TB PCR from bronchoscopy samples, anti-TB medications were continued given high clinical suspicion. He remained febrile and on day 10, vancomycin was added to cover resistant staphylococci. Moxifloxacin was changed to ethambutol owing to concern regarding lowering of the seizure threshold. Liposomal Amphotericin B (5mg/kg) was added to cover possible fungal infection.

On day 11 post admission, histopathological exam showed branched septate hyphae (Fig. 1f), with surrounding necrotising granulomatus tissue suggestive of cerebral mycoses. On day 14 anti-tuberculous drugs were discontinued, as both pulmonary and cerebral samples were negative for Mycobacterium tuberculosis. Fungal cultures confirmed growth of a dematiaceous mould. Voriconazole (5mg/kg) and caspofungin (70mg OD) were added on day 15 to provide further antifungal cover. A percutaneous tracheostomy was formed on day 15 to facilitate extubation. A CT head on day 18 showed increasing mass effect and enlargement of the lesions into the midbrain. The lesions were deemed too extensive for surgical resection.

Following a delay due to national shortage, flucytosine 2.5g QDS IV was initiated in place of caspofungin on day 21. There was difficulty achieving therapeutic voriconazole levels likely related to the rifampicin and phenytoin received earlier in admission. Voriconazole levels remained sub-therapeutic despite increasing the dose to 400mg BD on day 22. On day 22, GCS continued to fluctuate between 6 and 10/15. Despite a moderate transaminitis (peak ALT of 203 iu/L) he was able to tolerate triple antifungal therapy.

On day 24 identification of C. bantiana was confirmed. Molecular identification was performed by PCR amplification and sequencing of internal transcribed spacer region ITS-1 and the D1-2 fragment of the 28S rDNA gene [4]. The sequence of the D1-2 portion of the 28S rDNA gene was 100% identical over the entire 355 nt amplicon length with sequences from isolates of C. bantiana present in the synchronised public databases (EMBL accession numbers AM168525, AB363799 and KU928135). Antifungal susceptibility testing was performed using E-test method. Minimum inhibitory concentrations (MIC) of 6 antifungal agents were determined as: amphotericin B (1mg/L), flucytosine (2mg/L), isavuconazole (0.06 mg/L), itraconazole (0.06 mg/L), posaconazole (0.06 mg/L), and voriconazole (0.06 mg/L).

His subsequent clinical course was complicated by recurrent aspiration pneumonias requiring broad-spectrum antibiotics. Given the lack of neurological improvement and after discussions with his family the decision was made to continue medical management but not to attempt re-intubation or cardiopulmonary resuscitation should he deteriorate. On day 39 of admission the patient had a respiratory arrest and passed away. The patient was referred for post-mortem. Autopsy showed infection was confined to the brain. No granulomata were identified in the thoracic lymph nodes or lung therefore pre-existing diagnosis of sarcoidosis was not confirmed.

3. Discussion

C. bantiana is found in decaying plant matter in soil worldwide. The pathogenesis of phaeohyphomycosis due to C. bantiana infection is uncertain. While a minority of infections follows traumatic inoculation [1], it has been hypothesised that a subclinical pulmonary infection followed by haematogenous spread might seed infection to the CNS [5].

According to a recent systematic review [1] this is only the fifth published case of this infection in the UK, although MRL regularly receives isolates from cases acquired overseas (unpublished data). There are only two cases described of cerebral C. bantiana in the UK; both of these were in patients whom were significantly immunosuppressed – one due to an underlying immunodeficiency (Duncan’s syndrome) and one on chemotherapy for lymphoma [6,7]. This is in contrast to the
immunocompetence of our patient. In both cases patients were treated with amphotericin B, flucytosine and voriconazole. Despite this treatment strategy both patients died.

The diagnosis of *C. bantiana* infection presented a significant challenge. Its clinical presentation and radiological findings could not be distinguished from bacterial abscesses, primary CNS neoplastic lesions and cerebral metastatic disease. A fungal infection was considered initially unlikely compared to TB by the clinical teams. On admission it had been advised by the microbiology team to test for fungal biomarkers including β-D-glucan and galactomannan, however this was not done until later in admission. It can be postulated that this delayed the decision to cover for fungal infection.

Neuroimaging with CT or MRI confirms the presence of ring enhancing lesions with associated perilesional oedema and meningeal involvement may yield abnormalities in cerebrospinal fluid (CSF) such as raised white cell count and protein [8]. However in this case concerns about risk of coning meant lumbar puncture was not performed. Definitive diagnosis requires tissue biopsy for fungal culture, histology and panfungal polymerase chain reaction (PCR). In our patient, brain biopsy did not take place until day 6 as it was hoped that the bronchoalveolar lavage (BAL) would confirm tuberculous infection. Despite the relatively low invasiveness of BAL, extra-cerebral disease is rare, as in our case, so brain biopsy should be considered early.

Once a diagnosis is established, there is no defined treatment for cerebral phaeohyphomycosis due to *C. bantiana* and various strategies have been employed in the literature. The joint ESCMID/ECMM guidelines for management of systemic phaeohyphomycosis recommend complete excision of brain abscesses wherever possible along with combination antifungal therapy [9]. The efficacy of antifungal therapy is limited by both sensitivity of the organism and the relatively poor CNS penetration of many antifungals. In the systematic reviews of intracranial *C. bantiana*, no treatment strategy was associated with improved survival except for regimens containing itraconazole [1]. Our patient did not receive this at any point. The selection of an antifungal regimen in this case was complicated by low serum levels of voriconazole, likely due to cytochrome P450 induction from his rifampicin therapy.

| Day | Clinical Progress / Investigations                                                                 | Antimicrobials |
|-----|-----------------------------------------------------------------------------------------------------|----------------|
| 0   | Right hemiplegia, left focal seizures. WCC 21.3 CRP 115.6                                          |                |
| 1   | Intubated, bronchoscopy done. CT CAP bilateral upper lobe consolidation and pneumonia               |                |
| 2   | HIV negative                                                                                       |                |
| 3   | BAL. TB PCR negative                                                                               |                |
| 4   | Brain biopsy taken                                                                                 |                |
| 5   | CTH - worsening oedema, midline shift. Histopathology of brain biopsy suggestive of mycotic infection |                |
|     | Reintubated for low GCS                                                                            |                |
| 10  | Tracheostomy                                                                                      |                |
| 11  | CTH - progression of enhancing lesions with worsening mass effect                                  |                |
| 21  | CTH - ventricles more dilated suggestive of developing hydrocephalus                               |                |
| 22  | Stepped down to ward                                                                               |                |
| 23  | Transaminitis, peak ALT 203                                                                        |                |
| 25  | CTH - stable appearances                                                                          |                |
| 29  | Hospital acquired pneumonia                                                                       |                |
| 37  | Cellular immunology - no suggestion of immunodeficiency                                           |                |
| 38  | Respiratory arrest, RIP                                                                            |                |

Legend of abbreviations: ALT – Alanine Aminotransferase; BAL – Bronchoalveolar Lavage; CRP – C-reactive protein; CT CAP – Computerised Tomography Chest Abdomen Pelvis; CTH – Computerised Tomography Head; GCS – Glasgow Coma Score; HIV – Human Immunodeficiency Virus; PCR – Polymerase Chain Reaction; TB - Tuberculosis; WCC – White Cell Count.
treatment.

Although surgical resection is desirable, in our case brainstem disease at presentation meant that it was not possible. The autopsy report hypothesised that the patient’s final respiratory demise resulted from fungal infection directly affecting his respiratory centres in the brainstem or cerebral oedema causing the same. Had the disease been detected prior to the brainstem being affected it is possible that resection may have impacted on outcome.

Overall, this case demonstrates extensive CNS disease secondary to *C. bantiana* in an immunocompetent host. It highlights the potential diagnostic difficulties and the importance of astute clinical suspicion. Whilst there is currently no optimal therapeutic regimen, a combination of anti-fungal therapy and complete excision of brain abscesses should be employed.

**Conflict of interest**

The authors have no conflicts of interest to declare.

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