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Case Report

Microbiology Section

Cerebral Phaeohyphomycosis due to *Cladophialophora bantiana* – A Case Report and Review of Literature from India

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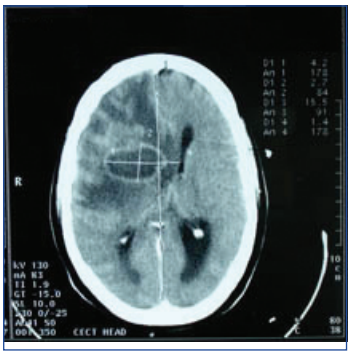
ABSTRACT

Cerebral phaeohyphomycosis is a rare disease caused by dematiaceous fungi. It has poor prognosis irrespective of the immune status of the patient. *Cladophialophora bantiana* is the most commonly isolated species. We report a case of multiple brain abscesses caused by *C. bantiana* in an immune competent patient. The diagnosis was based on CT scan of head, direct examination and culture of the aspirate from the abscess. Despite complete surgical resection of the abscesses and antifungal therapy with amphotericin B and voriconazole the patient could not be saved. All the cases of cerebral phaeohyphomycosis due to this rare neurotropic fungus reported from India between 1962 and 2009 have also been reviewed.

Keywords: Cerebral phaeohyphomycosis, *Cladophialophora bantiana*, Multiple brain abscesses, Voriconazole

CASE REPORT

A 65-year-old male, resident of rural area and farmer by occupation was admitted in a tertiary care hospital with one day history of altered sensorium and six days history of hemiparesis of the left side of the body with slurred speech. There was no history of fever, headache, vomiting or seizures. There was no significant past history of organ transplant, tuberculosis, diabetes mellitus or any chronic immunosuppressive therapy.

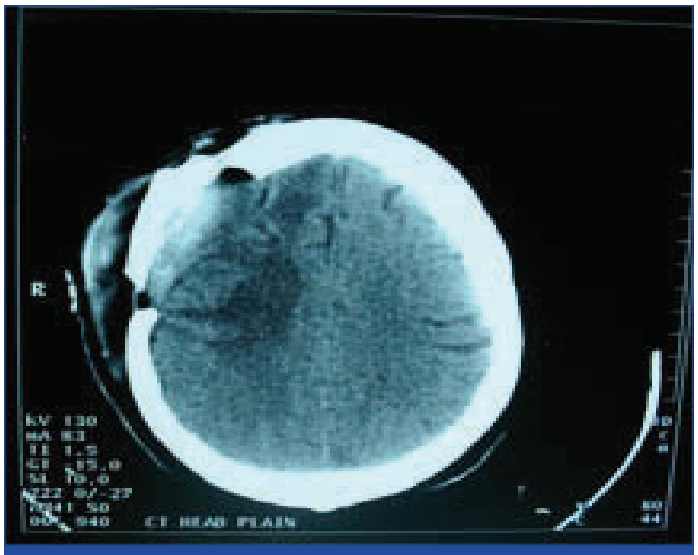


[Table/Fig-1]: CT Scan of head showing multiple ring enhancing lesions with midline shift and diffuse edema
[Table/Fig-2]: KOH mount of the aspirate showing brown, septate fungal hyphae (x400)
[Table/Fig-3]: Sabouraud's Dextrose Agar (SDA) showing olive grey to black velvety growth of C bantiana
[Table/Fig-4]: LCB mount showing brown septate hyphae with unbranched wavy chains of pale brown smooth one celled conidia (x400)

On examination, the patient was unconscious and afebrile. His vital signs were normal. Left leg of the patient showed oedema and ecchymotic patches. Pupils were reactive bilaterally. Neurological examination revealed deficit on left side. The respiratory, cardiovascular and abdominal examinations were within normal limits.

Haematological investigations revealed hemoglobin as – 13.5 g/dl and total leucocyte count as 20,000 cells/ mm^3 (granulocytes- 81.6%, lymphocytes- 17.2 % and monocytes- 1.3%). Serum biochemistry revealed deranged renal function (serum creatinine – 1.9 mg/dl) and slightly deranged serum electrolytes (sodium – 154mmol/l, potassium – 5.1mmol/l, chloride - 108mmol/l and calcium - 8.4mg/l). The liver function tests, lipid profile and blood glucose levels were within normal limits. The patient was seronegative for HIV-1 and HIV-2.

The computed tomographic (CT) scan of head was done using contrast which revealed two ring enhancing space occupying lesions in right high parietal region with midline shift and diffuse oedema [Table/Fig-1]. The Arterial and Venous Doppler study of the left lower limb suggested deep vein thrombosis. Chest X-ray was normal.



[Table/Fig-5]: CT scan showing post-operative changes and cerebral oedema

Case No.	Reference & Ref. No.	Age, Sex	Occupation	Clinical Presentation	Risk Factor	Radiological findings and Positive Microbiological/ Pathological findings	Therapy	Outcome
1	Bagchi et al., 1962 Calcutta [3]	54y, M	Not known	Fever-2 mths, Lt* hemiplegia - 6 wks, followed by headache, vomiting, seizures - 3 attacks in 3 wks	None	EEG- SOL†, HPE‡	Total excision	Expired
2	Sandhyamani et al.,1981 Delhi [4]	50y, M	Not known	Headache- 5 yrs, focal seizuresn -3 yrs, Lt hemiparesis – 2 yrs 6 months, Fever- 2 mth, altered sensorium	None	CT- well defined paraventricular mass in Rt§ basal ganglia, Culture, HPE	Partial excision, no antifungal agent given	Not Known
3	Sandhyamani et al., 1981 Delhi [4]	6 months, M	Not known	Seizure and fever-1 wk,	None	CT- multilocular abscess in frontal lobe, Repeat CT after 1 month- hydrocephalous, HPE	Partial excision, VP shunt after 1 month, no antifungal agent given	Survived, on follow up for 10 mths
4	Banerjee et al., 1989 Delhi [5]	28y, M	Rice merchant	Headache - 8mths, seizures - 2 attacks, blurred vision - 1 mth	None	CT-Lt frontal SOL, Gram stain, KOH mount, culture, HPE	Total excision, Amphotericin B,	Survived
5	Goel et al., 1992 Bombay [6]	36y, M	Not Known	Symptoms of raised ICT	None	CT- Rt parietal SOL, HPE, culture	Total excision, Ketoconazole, Amphotericin B	Recurrence after 2 mths, Expired
6	Dar et al., 1993 Delhi [7]	16y, M	Student	Headache & fever-17 days	None	CT- Lt fronto-parietal SOL, KOH mount, culture	Near total excision , Amphotericin B	Recurrence after 5 wks, Not known
7	Nadkarni et al.,1993 Bombay [8]	32y, M	Not known	Convulsions - 1 mth, bitemporal headache, hemiperesis, aphasia and altered sensorium after 1 wk	None	CT- multiple ring enhancing lesions in parieto-occipital region, KOH smear, culture, HPE	Parieto-occipital craniotomy with excision, Amphotericin B (Liposomal)	Expired after 20 days
8	Buxi et al., 1996 Delhi [9]	58y, M	Engineer	Headache & Fever- 10 days	Recurrent allergic rhinitis- taking steroids for 20 yrs	MRI and CT- Multiple conglomerate ring enhancing lesions in Rt and Lt frontal lobe, Abscess in Rt cerebellar hemisphere KOH mount, culture, HPE	Partial excision, Amphotericin B, Fungizone, Fluconazole,	Expired 20 days after surgery (Repeat CT showed no reduction in lesion)
9	Gupta et al., 1997 Chandigarh [10]	35y, M	Not known	3 episodes of seizure in 4 months	Renal allograft recipient	CT- multiple ring enhancing lesions in Rt parietal lobe, Culture	Surgical excision, Amphotericin B	Not known
10	Vyas et al., 2000 Jaipur [11]	35y, M	Farmer	Headache & vomiting - 2 mths, Seizures - 7days	None	CT- Lt frontal lobe SOL, EEG- Lt frontal lobe lesion, HPE	Total excision, Flucytosine	Survived, on follow up for 5 yrs
11	Sood et al., 2000 Delhi [12]	25y, M	Farmer	Seizures - 3 weeks, loss of consciousness – half an hour, speech difficulties, weakness in limbs	None	CT- Lt posterior frontal lesion KOH mount, culture, HPE	Total excision, Fluconazole	Survived
12	Banerjee et al.,2002 Delhi [13]	6 days, M	Neonate born in rural agricultural family	Fever, focal seizures, not accepting feed – 2 days	Unsterile method of cord cutting during home delivery	USG skull - cerebral edema and ventricular narrowing , meningitis CT - bilateral multiple cerebral abscess with exudates in ventricular cavity and CSF cisterns, KOH smear and culture (from CSF & Pus)	Subdural aspiration – 3 times, Amphotericin B, Flucytocine	Not known
13	Raut et al., 2003 Bombay [14]	26y, F	Not known	Facial paresis, sudden onset of Rt hemiparesis	SLE** – on steroids for 2 mths	CT- Lt fronto-parietal ring enhancing lesion Culture, HPE	Stereotactic aspiration, two weeks later - total excision of capsulated lesion, Amphotericin B	Survived, on follow up for 2 yrs, neurological deficit unchanged
14	Deb et al., 2005 Kolkatta [15]	26y, F	Housewife	Headache & vomiting - 4 months, generalized tonic clonic convulsions - 2 episodes, Rt hemiparesis	None	CT- irregular abscess in Lt poserior frontal region, culture, HPE and smear cytology	Total excision, Amphotericin B	Survived, on follow up for 8 months,
15	Vehlo and Ghodaonkar, 2007 Mumbai [16]	40y, M	Not known	Headache, vomiting, Lt hemiparesis – 4 days	None	CT/MRI – multiple ring enhancing lesions in Rt fronto-parietal region, culture, HPE	Subtotal excision, Fluconazole, Follow up scan at 4 wks- increase in abscess size - total excision	Survived on regular follow up

16	George et al., 2008 Vellore [17]	20y, M	Not known	Fever, headache & vomiting- 1 month, drowsy	None	CT - Multiple ring enhancing lesions in Rt frontal lobe, KOH smear, culture, HPE	Total excision	Expired after 15 days
17	Garg et al., 2008, Bangalore [18]	37y, M	7 farmers, 1 engineer, 1 mill worker 1 not known	Rt hemiparesis -1 month, altered sensorium- 2 days	None	LCA†† - parietal mass	Burr hole biopsy No antifungal agent given (diagnosed postmortem)	Expired
18	-do- [18]	32y, M		Psychotic behavior – 6 months, Lt hemiparesis – 2 days	None	RCA‡‡ - suprasylvian mass culture	Burr hole aspiration, No antifungal agent given (diagnosed postmortem)	Expired
19	-do- [18]	42y, M		Lt hemiparesis – 8 months, headache and fever – 4 months	None	Extensive Rt parieto-occipital hypodense areas with mild hydrocephalus, chronic meningitis KOH mount, culture	No antifungal agent given (diagnosed postmortem)	Expired
20	-do- [18]	20y, M		Raised ICT – 4 months , altered sensorium- 8 days	None	Multiple Rt frontal hypodense ring-enhancing lesions KOH mount, culture	Total excision, Amphotericin B	Expired
21	-do- [18]	16y, M		Raised ICT – 5 months	None	Multiple coalescing Rt frontal ring enhancing lesion KOH mount, culture	Total excision, Amphotericin B, 5-Flucytocine	Survived, 24 months follow up
22	-do- [18]	20y, M		Rt hemiparesis – 10 days	None	Lt posterior frontal hypodense ring enhancing lesion, KOH mount, culture	Total excision, Amphotericin B, 5-Flucytocine	Survived, 5 months follow up
23	-do- [18]	49y, M		Rt hemiparesis – 2 months, raised ICP – 1 month	None	Two cystic ring enhancing lesions in left frontoparietal Region, KOH mount, culture	Excision, Amphtericin B, Fluconazole	Survived, 3 months follow up
24	-do- and Jayakeerti, 2004 [18 and 19]	22y, M		Rt hemiparesis – 2 months, raised ICP – 1 month	None	Ring-enhancing lesion left frontal lobe, KOH mount, culture	Total excision, Amphotericin B, 5-Flucytocine	Expired
25	-do- [18]	25y, M		Raised ICT – 1 month, Rt hemiparesis – 5 days	None	Right frontal ring enhancing lesions, KOH mount, culture	Excision, 5-Flucytocine, Itraconazole	Survived, 3 year follow up
26	-do- [18]	3y, M		Right hemiparesis – 5 months	None	Left posterior frontal multiple coalescing ring enhancing lesions, KOH mount, culture	Total excision, Voriconazole	Survived 3 month follow up
27	Borkar et al., 2008 Delhi [20]	55y, M	Not known	Bifrontal headache, personality change, increasing forgetfulness- 3 mths, unsteady gait	None	MRI - ring enhancing lesion in periventricular region extending to Lt frontal region, KOH smear, culture, HPE	Total excision, Amphotericin B, Fluconazole	Recurrence after 6 months, Total follow up – 10 months, Expired
28	Lakshmi et al., 2008 Vellore [21]	23y, M	Botany student	Headache - 1 wk, Rt sided hemiparesis, facial palsy, diplopia	None	CT - ring enhancing lesion in Lt capsuloganglionic region, KOH smear, culture, HPE	Burr hole aspiration, Amphotericin B	Survived, Good cure
29	Present case, 2010 Ludhiana	65y, M	Farmer	Lt hemiparesis, slurring of speech - 6 days, altered sensorium-1 day	None	CT - two ring enhancing SOL in Rt high parietal region, KOH mount, culture	Total excision, Amphotericin B, Voriconazole	Expired

[Table/Fig-6]: Summary of cases reported from India
* Lt=Left, †SOL=Space Occupying Lesion, ‡HPE=Histopathological Examination, § Rt= Right, ¶ ICT= Intra cranial tension, **SLE=Systemic Lupus Erythematosus, ††LCA=Left Carotid Angiogram, ‡‡RCA= Right Carotid Angiogram

The patient was started empirically on broad-spectrum antibiotics. Neurological intervention was done on day 3 of admission using burr hole surgery and right fronto-parietal craniotomy. 50 ml of greenish yellow pus was aspirated. Total excision of the abscesses was performed. The aspirated pus was sent for microbiological examination. The Gram's stain of the exudates showed no microorganisms, Ziehl-Neelsen's stain showed no acid fast bacilli but KOH mount of the aspirate showed brown, septate fungal hyphae [Table/Fig-2]. The pyogenic culture and mycobacteriological culture were sterile. The fungal culture done on Sabouraud's Dextrose Agar (SDA) with and without antibiotics showed olive grey to black velvety growth on the obverse and black pigment on the reverse side [Table/Fig-3]. The lactophenol cotton blue (LCB) mount showed brown, septate hyphae with unbranched wavy chains of pale brown smooth oval one celled conidia without pigmented hila [Table/Fig-4]. Slide culture of the isolate also showed septate hyphae with

long chains of oval conidia. Based on the findings of KOH mount and culture, the fungus was identified as *C. bantiana*. The thermal tolerance test (growth at 42^0 C) was positive - differentiating it from other *Cladophialophora spp*. Antifungal susceptibility testing could not be performed due to non-availability of containment level 3 in our laboratory. The material collected from the skin lesions was also inoculated on SDA but no fungal growth was obtained.

On the basis of KOH mount findings, treatment with amphotericin B (10-20 mg/day) and voriconazole (400 mg stat and 300 mg OD) was started on day 4 of admission. Full dose of amphotericin B could not be given because of impaired renal function. The liposomal formulation of amphotericin B (known to be less nephrotoxic) and full dose of voriconazole could not be added due to cost constraints. Five days post-surgery the patient started responding to verbal commands and was extubated but two days later he developed fever and upper gastrointestinal bleed. Later post-operative period was complicated because of subsequent development of renal failure and pneumonitis. There was sudden deterioration in his sensorium after 14 days of surgery and CT scan of head was repeated but it showed post-operative changes only [Table/Fig-5]. In spite of all possible efforts the patient could not be saved and expired on day 18 of hospitalization due to cardiorespiratory arrest. An autopsy was not permitted.

DISCUSSION

Cerebral phaeohyphomycosis caused by dematiaceous (darkly pigmented) fungi is a rare infection. *Cladophialophora bantiana* (*C. bantiana*) is the most commonly encountered agent causing phaeohyphomycosis of the central nervous system (CNS) [1]. It is a dematiaceous fungus with distinct neurotropism. Earlier it was known as *Cladosporium trichoides*, *Cladosporium bantianum*, *Xylohypha bantiana* and *Xylohypha emmonsii* [2]. A total of 28 cases have been reported from India between 1962 and 2009, results of which (including the present case) have been summarized in [Table/Fig-6] [3-21]. Infection usually occurs in the second to third decade of life (unlike our patient who was 65-year-old). The median age of the patients was 28 years (range- 6 days to 58 years). Most of the patients in the cases reported from India are males (n=26) and male: female ratio in India is about 14:1 which is much higher than the male: female ratio of 3:1 reported worldwide [22]. It has no ethnic or geographic predilection. But an occupational predisposition in agricultural workers, particularly farmers have been reported. Occurrence of this rare infection in a botany student (case 29), an engineer living in an agricultural institute (case 8) and a neonate born in rural agricultural family (case 12) also suggests increased risk with exposure to plants/ vegetation.

Infection with this emerging neurotropic fungus commonly presents as brain abscess and rarely may it present as meningitis and myelitis. According to an earlier review of 17 cases by Middleton et al., [23] cerebral abscess was present in 13 cases and only four patients had meningitis. In a recent review of culture proven cases of primary CNS phaeohyphomycosis by Revankar et al., [1] almost all cases of *C. bantiana* presented with brain abscess. In the present review of cases reported from India brain abscess was the most common clinical presentation (26 cases, 93%). Solitary SOL was noted in 18 cases (67%), while 9 cases (33%) had multiple lesions. Most common site for abscess formation was the frontal lobe (14 cases, 52%). All the patients had > 2 symptoms at the time of presentation. Hemiparesis and headache were the most common clinical manifestations followed by seizures, symptoms of raised ICT and altered sensorium. Hemiparesis was noted in 15 cases (54%), headache was present in 13 cases (46%) and fever was noted in 11 cases (39%). Duration of symptoms ranged from 2 days to 5 years (median - 2 months).

The portal of entry of this fungus is not clearly understood; most probably it is by inhalation of spores followed by haematogenous dissemination to brain [1]. Other suggested routes are through direct extension from adjacent paranasal sinuses, or by penetrating trauma to the head. In cases reported from India risk factors or underlying predisposing factors were documented in 4 cases only – one each of Systemic Lupus Erythematosus on steroids for 2 months, recurrent allergic rhinitis on steroids for 20 years, solid organ transplant recipient (Renal allograft recipient), and a 6-day-old neonate who was delivered at home and cord was cut by unsterile method; whereas no obvious predisposition was noticed in rest 24 cases. Our patient was a farmer by occupation suggesting soil as the most probable source of infection and inhalation followed by haematogenous seeding of brain as the most probable route of infection. The presence of multiple brain abscesses in our case further support hematogenous source of the abscesses.

Out of 28 cases reported, 25 (89%) were diagnosed during life and 3 cases (11%) were diagnosed on postmortem/ autopsy. CT scan was done in all the patients except 3 cases, out of which angiogram was performed in 2 cases and EEG was performed in one case prior to the availability of CT scan. Out of 28 cases fungal culture was done and was positive for *C. bantiana* in 25 cases, where as no fungus culture was performed in 3 cases. The etiological agent in these 3 cases was identified only on the basis of histopathological findings.

Majority of these patients were in good health when they got the infection. An immunological deficiency was seen in only three of these cases. The duration of symptoms has also been variously reported varying from a few days to many years. In the comprehensive review of 10 cases done by Garg et al., [18] (cases 14-23), CSF analysis has been reported in 5 cases, which shows significant polymorph predominance in 3 cases. In the review by Revankar et al., [1] significant pleocytosis was noted in 70% of the patients with brain abscess and 100% patients with meningitis. CSF analysis was not documented in most of the Indian reports. In the present review, the authors have summarized the findings of all the previous Indian reports and observed that in most of these cases there was no indication of infection with such rare fungus. In fact demonstration/ recovery of this fungus from clinical material obtained during surgery or postmortem clinched the diagnosis. In our case the autopsy was not permitted which could have given important information e.g. disseminated fungal lesions in other organ systems or cerebral haemorrhage.

Mortality rate of the disease is as high as 65% to 70% in patients who undergo aggressive medical and surgical treatment [1] and 100% in those who do not undergo surgery [24]. High mortality is mainly due to delay in surgical resection and treatment with less effective antifungal agents like amphotericin B [25]. In the present review an overall mortality rate of 48% was observed in those patients where outcome was reported (12 out of 25). Only 4 patients had documented cure in the present case review following Revankar et al., definition of cure (patients free of symptoms for >1 year) [1]. Eleven patients survived, 14 expired and the outcome of 3 patients could not be known. Recurrence of the abscess was noted in 7 cases (in 6 cases after 1-2 months and in 1 case after 6 months) in spite of being on antifungal therapy. In our case the patient's poor outcome can be attributed to the development of renal failure and the presence of multiple brain abscesses which is associated with poor prognosis as compared to solitary lesions [26].

There is no standardized therapy for CNS phaeohyphomycosis although the combination of amphotericin B, flucytosine and itraconazole/ voriconazole along with complete surgical excision of the abscess is associated with improved survival rates. Keeping in mind the high mortality associated with this infection an aggressive therapeutic approach is needed.

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