Cryptococcus gattii Meningitis Developed after Pneumonectomy in A Case of Necrotizing Pneumonia

Chih-Chen Lin1*, Hsiang-Kuang Tseng1,2,3*, Wei-Sheng Wang1,2,3, Yee-Chun Chen4, Tseng-Yu Huang1,2,3, Alice Ying-Jung Wu1,2,3, and Chang-Pan Liu1,2,3

1Section of Infectious Diseases, Department of Internal Medicine, Mackay Memorial Hospital; 2Department of Medicine, Mackay Medical College; 3Mackay Junior College of Medicine, Nursing, and Management; 4Division of Infectious Diseases, Department of Internal Medicine, National Taiwan University Hospital and College of Medicine

Abstract

Cryptococcus gattii infection has rarely been reported in Taiwan. We report a case of 32-year-old immunocompetent male Vietnamese referred for pneumonectomy under the impression of necrotizing pneumonia. Pleural effusion was collected during the pneumonectomy and the culture of pleural effusion yielded Cryptococcus. Although oral fluconazole 200 mg daily was prescribed, meningitis developed 10 days after pneumonectomy. Culture of cerebrospinal fluid (CSF) yielded Cryptococcus. The patient received amphotericin B plus flucytosine for two weeks as antifungal induction therapy, followed by intravenous fluconazole 400 mg per day as consolidation therapy. The molecular typing of Cryptococcus was C. gattii VGII. He was discharged on the 40th day after admission and was prescribed oral fluconazole 200 mg daily for 110 days at outpatient department. No neurological sequela was found at the time of last follow-up. Importantly, Cryptococcus has tendency to infect the central nervous system (CNS), especially the subspecies C. gattii, which could be differentiated from C. neoformans by its activity in the medium containing canavanine, glycine and bromothymol blue (CGB). Patients with necrotizing pneumonia caused by C. gattii should be treated as central nervous system infection. In conclusion, appropriate fungicidal agents to cover CNS infection should be administered for this kind of patient from the time of first disease onset. (J Intern Med Taiwan 2014; 25: 30-35)

Key Words: Cryptococcus, Cryptococcus gattii, Meningitis, Necrotizing pneumonia

Introduction

Cryptococcus infection is life threatening and is related to severe lung and central nervous system complications. It also has unique microbiological, epidemiological, clinical presentations and outcomes1. Among members of Cryptococcus neoformans-Cryptococcus gattii species complex, C. neoformans is distributed worldwide whereas C. gattii is considered to be more prevalent in the subtropical and tropical areas including Taiwan. The two species of Cryptococcus can be differenti-
ated using a solid agar medium containing canavanine, glycine and bromothymol blue (CGB)². Furthermore, by using orotidine monophosphate pyrophosphorylase (URA5) gene restriction fragment length polymorphism analysis, they can be further divided into eight genotypes, including four types of *C. neoformans* (VNI, VNII, VNIII, and VNIV) and four types of *C. gattii* (VGI, VGII, VGIII, and VGIV)³. *C. gattii* VGI molecular type is traditionally considered to be the most prevalent, although *C. gattii* VGII molecular type caused the Vancouver Island outbreak⁴,⁵. The VGIII and VGIV molecular types of *C. gattii* were commonly isolated from HIV-infected patients in Africa and Northern America⁶-⁷.

*C. gattii* was isolated more often than *C. neoformans* in immunocompetent patients⁶. Patients infected with *C. gattii* were younger, more likely to have no underlying conditions and more likely to have meningoencephalitis¹. In common model, rats infected with the *C. gattii* infections result in more cryptococcoma complicated with hemorrhage and neurological sequelae requiring longer treatment duration⁷-⁹. Whether host immune function or different subtypes of *C. gattii* contribute more to poor prognosis is still in debate⁵-⁷.

In 1990, *C. gattii* was isolated from *Eucalyptus camaldulensis*; subsequently, it was believed that *C. gattii* spread through the exportation and flow-er of *Eucalyptus* species¹⁰,¹¹. An environmental study in 2007 also isolated *C. gattii* from a broad range of trees in Canada¹². This implied that plants are probably the niche of *C. gattii*. In 1999, *C. gattii* caused an outbreak in Vancouver Island (British Columbia), Canada. Since then, numerous studies have revealed more information about *C. gattii*; which challenged what we knew about *C. gattii* in the past⁴,⁶. For example, genealogy and phylogenetic analyses revealed that *C. gattii* VGII found in the temperate regions of British Columbia most likely emerged from the tropical Amazon rainforest¹³.

We introduce a 32 year-old Vietnamese lumberman who had necrotizing pneumonia related to *Cryptococcus gattii* VGI. It was interesting that

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![Figure 1](image_url)

**Figure 1.** (A) The chest X-ray 2 weeks before referral presents right lower lobe opacity; (B) The chest X-ray on referral day presents stationary right lower lobe lesion compared with previous film.
his meningitis developed 10 days after pneumonectomy. Even though the diagnosis was made late and the surgeon did not use fungicidal agents initially, the patient recovered without neurologic sequelae. Because *C. gattii* present with unique characteristics and clinical manifestations, it is important to discriminate it from *C. neoformans*.

**Case report**

A 32 year-old male Vietnamese came to Taitung, Taiwan and worked as a lumberman for five years. He suffered from intermittent fever and persistent cough with yellow sputum for two weeks. He had generalized malaise and poor appetite. He denied body weight loss, chest tightness, gastrointestinal symptoms or genitourinary symptoms. Then he went to clinic for further evaluation. The chest radiograph showed right lower lobe opacity (Figure 1A). Oral amoxicillin/clavulanate and roxythromycin were empirically prescribed. Initially neither bacteria nor mycobacterium was found in the sputum Gram stain and acid fast stain. Fever subsided but he had sustained productive cough. After two weeks of oral antibiotics treatment, infiltrates persisted on the chest radiograph (Figure 1B) and clinical symptoms progressed. Then he was admitted for assessment. He had history of *Blastocystis hominis* infection which was diagnosed through immigrant health examination, for which he received complete treatment with metronidazole. He smoked about one pack per day for 15 years. The blood examination showed white blood cell count 12,400 with 81.9% neutrophils but the rest of the data were in normal range. Contrast-enhanced computed tomography (CT) of the chest revealed heterogeneous lesions on superior segment of the right lower lung, suspicious of necrotizing pneumonia (Figure 2). Empiric piperacillin/tazobactam and levofloxacin were prescribed. Bronchoscopy showed a protruding hypervascular lesion situated at RB 6 orifice (Figure 3), which is the superior segment of the right lower lobe. The sputum cultures for bacteria and *Mycobacterium tuberculosis* yielded no growth and cytology showed no malignant cell. Under the tentative diagnosis of necrotizing pneumonia which failed medical treatment, he was referred to a medical center for surgical intervention. Pneumonectomy was done on the 4th day after referral. Pathology of the lung tissue showed cryptococcosis with mucin pools surrounded by prominent inflammation. Culture
of the pleural effusion showed Cryptococcus. Oral fluconazole 200 mg daily was prescribed. However, fever flared up along with headache, nausea and vomiting on the 10th day after surgery. Brain CT showed no specific finding. Meningitis was suspected and lumbar puncture was done. Cerebrospinal fluid (CSF) study revealed protein 53 mg/dL (10~45 mg/dL), glucose 62 mg/dL (45~75 mg/dL), WBC 84/ml with lymphocyte 66/ml and neutrophil 14/ml. India ink was negative but the Cryptococcus antigen titer in both CSF and serum were positive and were 1:8 and 1:64 respectively. Amphotericin B and fluucytosine were prescribed as antifungal induction therapy. Both pleural effusion and CSF culture yielded Cryptococcus species. Because the yeast turned CGB medium blue, it was identified as Cryptococcus gattii. Molecular typing with URA 5 identified the species as VGII. Antifungal susceptibility obtained by a commercially prepared, dried colorimetric microdilution panel (Sensititre YeastOne, Thermo-Fisher Scientific, West Sussex, UK) was shown in Table 1. No yeast was identified in the blood culture bottle. After two weeks of induction therapy with amphotericin B 50 mg daily and fluucytosine 2 gm every six hours, the patient felt better. We switched to fluconazole 400 mg daily as consolidation therapy. He was discharged on the 40th day after referral. He received oral fluconazole 200 mg daily in the outpatient clinic of a local hospital in Taitung. No neurological sequela was found after a follow-up period of 110 days.

**Discussion**

Both Vietnam and Taitung, Taiwan are in the tropics, with climate suitable for the growth of *C. gattii*. This Vietnamese patient has been working in Taiwan for 5 years as a lumberman. He was in the risk of exposure to certain kinds of plants which might possess *C. gattii* although there is a lack of environmental survey studies in Taiwan. Since articles report the delayed onset *C. gattii* infection which occurred more than 10 months after traveling to *C. gattii* endemic area, it is unknown whether the patient acquired *C. gattii* from Taiwan or Vietnam. Further genetic analysis such as multi-locus sequence typing to confirm the original source of pathogen was needed.

It is an unusual for cryptococcal meningitis to develop after pneumonectomy. Several possibilities could have contributed to the patient’s meningitis, including fungal load, molecular types, lack of initial application of fungicidal agent, and transient fungemia post pneumonectomy. A rat model was built to explore the pathogenesis of *C. gattii* infection. Rats developed gross lung lesions with dissemination to brain, kidney and spleen only when the yeast inoculated was more than $10^7$ colony forming units (CFUs) per 0.1 ml. Different clinical manifestation and prognosis among *C. gattii* correlated with different subtype and malting type of *Cryptococcus*. The molecular type VGII (Colombia VGIIa- MAT alpha and VGIIb- MAT alpha) has tendency to induce late onset CNS infection. In addition, the lack of fungicidal agents use initially also contributed to the disseminated infection in our case. According to the 2010 IDSA guideline for cryptococcal infection, severe pulmonary cryptococcal infection should be treated as CNS infection. The IDSA guideline suggested two steps therapy. Induction therapy with amphotericin B 0.5~0.8 mg/kg/day intravenous infusion and fluucytosine 25mg/

### Table 1. Antifungal susceptibility of the pleural effusion and CSF *Cryptococcus gattii* isolates by a commercial microdilution panel Sensititre YeastOne

<table>
<thead>
<tr>
<th>Antifungal</th>
<th>Pleural effusion</th>
<th>CSF</th>
</tr>
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<tbody>
<tr>
<td>Amphotericin B</td>
<td>0.25</td>
<td>0.25</td>
</tr>
<tr>
<td>Flucytosine</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Fluconazole</td>
<td>16</td>
<td>16</td>
</tr>
<tr>
<td>Voriconazole</td>
<td>0.12</td>
<td>0.12</td>
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kg oral every six hours is suggested until patients become afebrile and the culture turn negative. Further antifungal consolidation therapy with oral fluconazole 400 mg daily is recommended for 8–10 weeks. Chen et al. studied 86 patients who suffered from C. gattii infection in the lung or in the central nervous system\textsuperscript{18}. In comparison with C. neoformans, it was found that infection with C. gattii required longer treatment duration; additionally, antifungal induction therapy with amphotericin B plus flucytosine led to better outcome in comparison to other antifungal agents\textsuperscript{18}.

The Clinical and Laboratory Standards Institute (CLSI) does not provide clinical breakpoints (CBPs) for Cryptococcus species\textsuperscript{19}; the epidemiologic cutoff values (ECVs) (highest wild type susceptibility endpoint) of antifungal susceptibility for reference was also not available for VGI\textsuperscript{20,21}. ECVs is the minimum inhibition concentration (MIC) values that captured >95% of the observed population in RPMI medium provided in recent studies\textsuperscript{20,21}. While CBPs predict the clinical outcome of therapy, the ECVs could monitor the emergence of strains with reduced susceptibility to the agent being evaluated due to mutation.

Finally, pneumonectomy might have resulted in transient fungemia which subsequently led to meningitis. In a murine inhalation model, C. gattii could be isolated from the brain despite consistent failure to recover infected cells from the blood.\textsuperscript{7} The authors speculate that transient fungemia may be the reason of the dissemination. Several factors may explain this phenomenon. First, C. gattii has ability to suppress the release of inflammatory factors\textsuperscript{7}. Second, C. gattii could accommodate into macrophages, which allows it to cross the blood-brain barrier (Trojan’s horse mechanism)\textsuperscript{7,22,23}. Hence, despite the low fungal load in blood, C. gattii could still effectively cross the BBB and cause CNS infection.

In conclusion, C. gattii infection is rare in Taiwan. Early diagnosis and prompt treatment are important in CNS C. gattii infection. This case reminds us that severe pulmonary cryptococcal infection should be treated as CNS infection. The case also demonstrates that it is important to discriminate C. gattii from C. neoformans by CGB medium in clinical situations.

References

Cryptococcus gattii Meningitis