Journal of Respiratory Medicine and Lung Disease

Research Article
Published: 28 Sep, 2017

A Study of 19 Pulmonary Cryptococcosis Cases

Feng-Li Zhou, Jia-Xin Zhu, Xiao-Ling Zou, Ding-Yun Feng and Tian-Tuo Zhang*

Department of Respiratory Medicine, Sun Yat-sen University, China

Abstract

Background: Pulmonary cryptococcosis (PC) has recently become more frequent.

Methods: We collected and analyzed data from 19 cryptococcal pneumonia cases that had been treated in our hospital over the past 10 years and analyzed the clinical features, underlying diseases, past medication, and auxiliary examinations of the patients.

Results: The patients demonstrated various symptoms and underlying diseases, but most of the latter involved diseases associated with immune deficiency. More than half of the patients had no history of exposure to bird droppings or soil. Chest computed tomography scans of the 19 patients revealed single or multiple pulmonary nodular shadows in 11 subjects. Initial misdiagnosis was common. Because of the long course of treatment for PC and the high costs of fluconazole and itraconazole, as long as the renal functions in these patients are normal, the more cost-effective drug amphotericin B can be used as an alternative treatment for PC.

Conclusions: This study provides a summary of the characteristics of pulmonary cryptococcosis as a basis for future clinical diagnosis and treatment of pulmonary cryptococcosis.

Keywords: Acquired immune deficiency syndrome; *Cryptococcus neoformans*; Diagnosis; Disseminated cryptococcal infection, Imaging; Pathology; Pulmonary cryptococcosis

Introduction

Pulmonary cryptococcosis (PC) is a subacute or chronic visceral fungal disease caused by *Cryptococcus neoformans* infection [1]. In recent years, the incidence of cryptococcal infection has shown an apparent upward trend [2,3]. Because of the atypical clinical manifestations of pulmonary cryptococcosis, clinical diagnosis is difficult; we have collected 19 cryptococcal pneumonia cases that had been treated in our hospital over the past 10 years, and analyzed the corresponding data. We hope to help clinicians improve their understanding of pulmonary cryptococcosis.

Materials and Methods

Source of subjects

From January 01, 2006 to December 31, 2015, 19 patients in total were diagnosed with cryptococcal pneumonia in our hospital, and pertinent information, including general information, medical history, laboratory data, and treatments were collected, analyzed, and summarized. The study was approved by the Ethics Committee of Clinical Medical Research at The Third Affiliated Hospital, Sun Yat-sen University (Approval No. (2016)2-114). The requirement for obtaining informed consent was waived by the committee due to the retrospective nature of the study.

Diagnostic criteria

The diagnostic criteria [4,5] are outlined below.

Pathogenic examination: Traditional microscopy and culture methods were used to identify *C. neoformans* from the patients' phlegm, broncho-alveolar lavage fluid (BALF), or cerebrospinal fluid (CSF). The strains of *C. neoformans* involved were identified using the Rap ID Yeast Plus System [6].

Pathological diagnosis: The pathological tissues of the patient were collected in different ways, such as by lung needle biopsy or fine-needle aspiration, or by bronchoscopic-protected brushing. *C. neoformans* was then identified by different staining techniques.

Cryptococcal antigens: The latex agglutination test was used to detect the capsular polysaccharide antigen of *C. neoformans* from patients' serum, BALF, or excessive pleural fluid.

OPEN ACCESS

Correspondence:
Feng-Li Zhou, Department of Respiratory Medicine, The Third Affiliated Hospital, Sun Yat-sen University, No. 600 Tianhe Road, Guangzhou, China, Tel: +86-13922165519;
E-mail: zhoufenglizfl@126.com

Received Date: *21 Aug 2017*
Accepted Date: *21 Sep 2017*
Published Date: *28 Sep 2017*

Citation:
Zhou F-L, Zhu J-X, Zou X-L, Feng D-Y, Zhang T-T. A Study of 19 Pulmonary Cryptococcosis Cases. J Respir Med Lung Dis. 2017; 2(4): 1024.
ISSN: 2475-5761

Table 1: Values for CD4+ and CD8+ T lymphocytes in the 19 patients.

Subject	CD4+T/T (%)	CD8+T/T (%)	CD4/CD8
1	18.6	25.3	0.74
2	16.8	24.3	0.69
3	21.5	28.3	0.76
4	20.7	38.1	0.54
5	21.6	32.5	0.66
6	19.5	35.2	0.55
7	28.4	32.8	0.87
8	23.6	39.8	0.59
9	14.3	21.8	0.66
10	19.2	29.3	0.66
11	28.1	40.3	0.7
12	21.8	38.4	0.57
13	14.3	25.1	0.57
14	19.6	31.6	0.62
15	25.3	41.7	0.61
16	15.3	28.6	0.53
17	24.7	38.5	0.64
18	21.4	30.2	0.71
19	18.6	25.8	0.72

Normal reference values used in our hospital: CD4+ T lymphocytes/T lymphocytes (T/T) 28% to 58%, CD8+ T/T: 19% to 48%, CD4/CD8: 0.9-2.0.

Results

General information

Of the 19 patients, 11 were male and 8 were female. Patients' ages ranged from 26 to 81 years, with a median age of 51.5 years.

Patients' medical history

Symptoms: Seven individuals were asymptomatic (36.8%), 8 subjects had a cough and expulsion of phlegm (42.1%), 9 had chest pain (47.4%), 3 had fever (15.8%), and 3 had dyspnea and hypoxemia (15.8%). Ten subjects had fatigue and weight loss (52.6%), 4 subjects had hemoptysis (21.1%), 3 had pleural effusion (15.8%), and 1 subject had concurrent cryptococcal meningitis (5.3%).

Underlying diseases: Five patients had no underlying diseases (26.3%), 3 had AIDS (15.8%), 2 had diabetes (10.5%), 4 had tuberculosis (21.1%), 2 had undergone organ transplantation (10.5%), 1 had rheumatic autoimmune disease (5.3%), and 2 had tumors (10.5%).

Exposure history

Two patients (10.5%) had a history of raising chickens, 4 patients (21.1%) had a history of soil exposure, and the remaining patients (68.4%) were not occupationally exposed to bird droppings and/or soil.

Previous medication history

Five subjects without underlying disease and 3 subjects with AIDS had no medication history; the 2 subjects who had undergone organ transplantation had a 0.5- to 3-year history of taking anti-rejection drugs. One subject with rheumatic autoimmune diseases had a history of long-term glucocorticoid intake. Two cancer subjects had received more than 2 courses of chemotherapy.

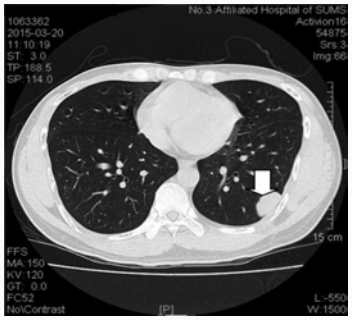


Figure 1: The white arrow indicates a solitary pulmonary nodule.

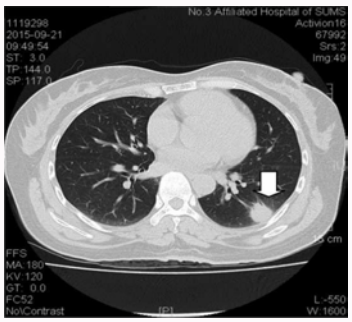


Figure 2: The white arrow indicates a solitary pulmonary nodule with halo signs.

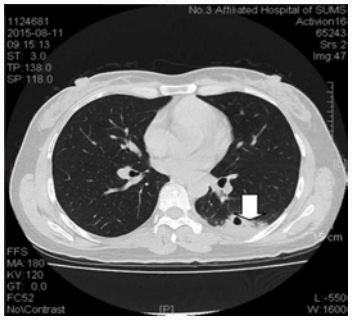


Figure 3: The white arrow indicates a solitary pulmonary nodule with cavities.

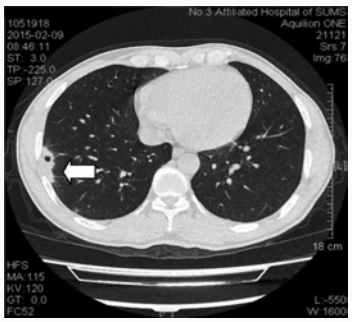


Figure 4: The white arrow indicates a solitary pulmonary nodule with cavities.

Hematology

No patients underwent galactomannan testing, as it was not yet available in our hospital at the time of the study. We detected CD3 and CD4 in the blood of all patients (Table 1).

Computed tomography imaging

Of the 19 patients, CT imaging showed single or multiple

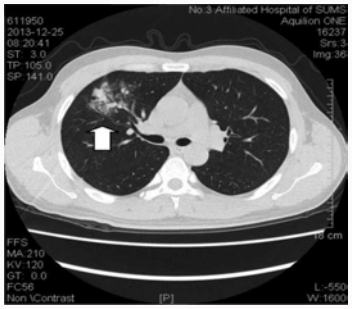


Figure 5: The white arrow indicates a patchy pulmonary opacity with air bronchogram.

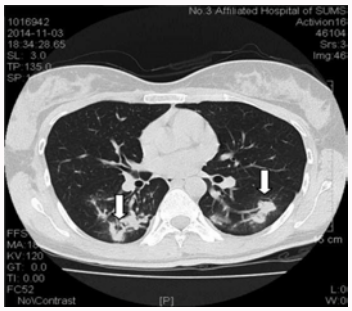


Figure 6: The white arrow indicates a bilateral pulmonary patchy and multiple nodular opacities.

pulmonary nodular shadows in 11 subjects (57.9%), of which 7 cases had cavity formation and 5 subjects had a "halo sign." Lung patchy shadows were found in 5 subjects (26.3%), 2 subjects (10.5%) had diffuse miliary shadows, and 1 subject (5.3%) had acute interstitial pneumonia. Mild mediastinal lymph node enlargement was observed in 12 subjects. Localized foci were generally found in the lower lobes of the lung (Figures 1-6, foci indicated by arrows).

Pathology

The diagnoses of the 19 patients were confirmed by pathological examinations. *C. neoformans* was found in 15 subjects using CT-guided percutaneous needle biopsy of the lung, in 3 subjects by using thoracoscopic lung biopsy, and in 1 subject using bronchoscopic lung biopsy. Granuloma formation (Figure 7) or jelly-like lesions (Figure 8) were observed under low magnification. *C. neoformans* with capsules could be observed inside macrophages under high magnification indicated by the arrows in Figures 9-13 [7].

Etiology

Culture and smear: Among the 19 patients, *C. neoformans* was isolated from only 6 subjects. One subject had a positive CSF direct ink smear, while BALF culture and culture of the secretions collected using bronchoscopic-protected brushing were positive in the other 4 and 1 subjects, respectively. The smear and culture results of *C. neoformans* are shown in Figures 14-19.

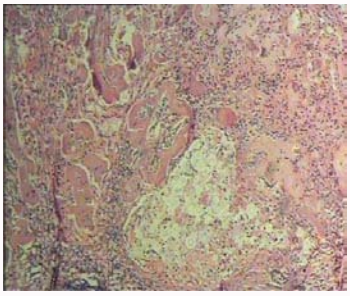


Figure 7: Granulomatous lesions (10 × 10).

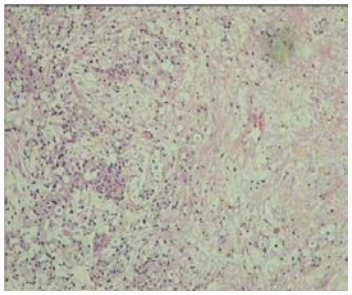


Figure 8: Mucinous lesions (10 × 10).

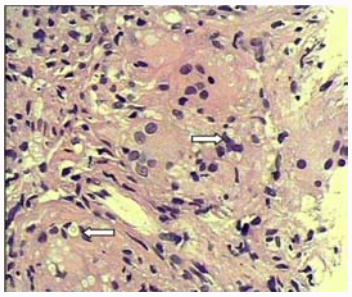


Figure 9: Hematoxylin and eosin stain (20 × 10).

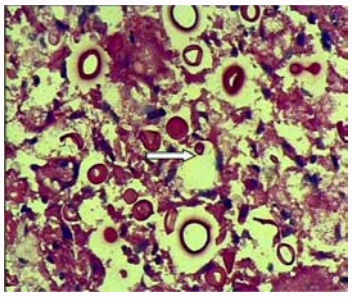


Figure 10: Periodic acid-Schiff stain (40 × 10).

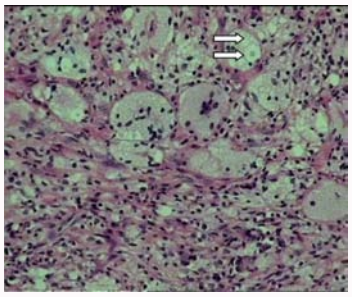


Figure 11: Hematoxylin and eosin stain (10 × 10).

Identification of strains: *C. neoformans* were identified using the Rap ID Yeast Plus System; examples of results are shown in Figures 20-21.

Diagnosis and treatment

All 19 patients were diagnosed with *C. neoformans* infection. All patients had received some form of misdiagnosis. Four patients were

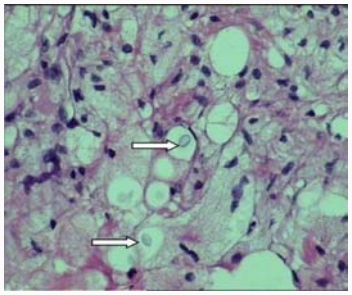


Figure 12: Hematoxylin and eosin stain (40 × 10).

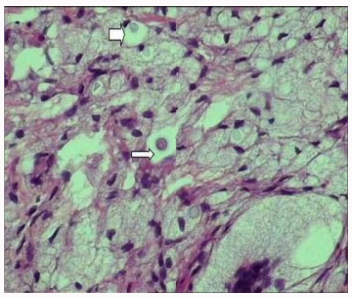


Figure 13: Hematoxylin and eosin stain (40 × 10).

Figure 14: Bronchoalveolar lavage and Sabouraud medium on day 4.

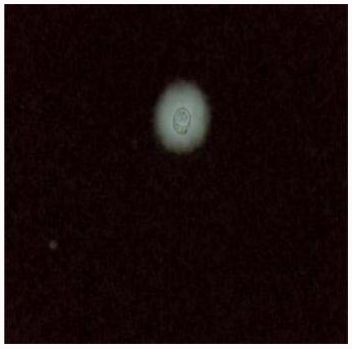


Figure 15: India ink stain of direct cerebrospinal fluid smear, 100 × 10 (thick capsule).

misdiagnosed with tuberculosis, and diagnostic anti-tuberculosis treatment that had been administered for 2-4 weeks was ineffective. Five subjects were misdiagnosed with lung cancer, and were subsequently diagnosed with *C. neoformans* infection based on CT-guided percutaneous needle biopsy of the lung. Three subjects with patchy shadows in their chest CT scans were misdiagnosed with pneumonia, and a 2-week antibiotic treatment was ineffective. Three subjects with underlying diseases (AIDS) were misdiagnosed with viral infection or Pneumocystis carinii infection, because the

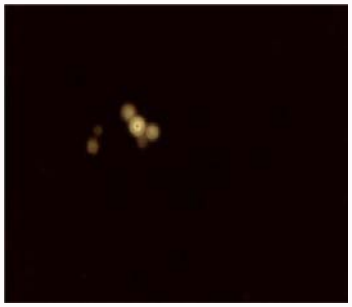


Figure 16: India ink stain of direct cerebrospinal fluid smear, 100 × 10 (very thin capsule).

Figure 17: Gram-stained culture, 100 × 10.

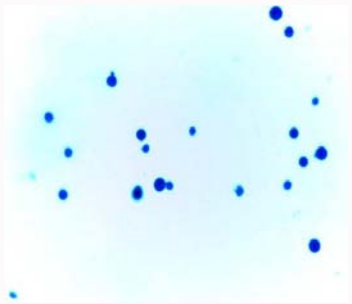


Figure 18: Methylene blue-stained culture, 100 × 10.

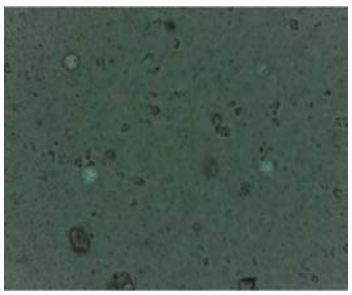
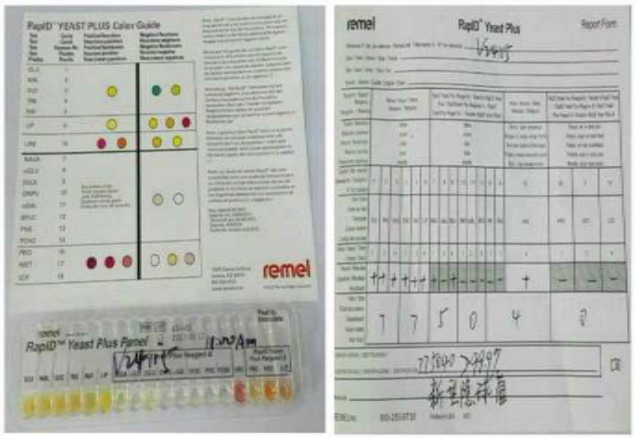


Figure 19: India ink-stained culture, 100 × 10 (very thin capsule).

bilateral lung lesions involved diffuse pulmonary infiltrates, and 2-week ganciclovir or caspofungin therapy was not effective. After receiving the correct diagnosis, 16 patients with localized PC were administered fluconazole (0.4 g/day) intravenously for 2-4 weeks and were then given fluconazole (0.4 g/day) orally for 3-6 consecutive months. One patient who also had central nervous system infection received fluconazole (0.4 g/day) orally for 12 months. The 3 subjects with underlying diseases (AIDS) had disseminated cryptococcosis;

Figure 20: Identification of strains.



Figures 21: Identification of strains.

for these patients, amphotericin B was administered for 6-8 weeks (starting from 5 mg/day, and gradually increasing to the maintenance dose of 0.6 mg/kg per day), co-administered with flucytosine (100 mg/kg per day) for the first 2 weeks. After 8 weeks, fluconazole 0.4 g/day was given orally for 6-12 consecutive months. The treatments for all subjects followed the Clinical Practice Guidelines for the Management of Cryptococcal Disease [8].

Outcome

All 16 non-AIDS patients completed the course of treatment, and the corresponding clinical symptoms disappeared. Chest CT lung scans were performed again, and revealed that the pulmonary foci had been absorbed, and the patients were cured or in a stable condition for more than 3 months. Of the 3 subjects with underlying diseases (AIDS), 1 died after 4 weeks of treatment due to the deteriorating condition associated with AIDS. For another of these subjects, amphotericin B was switched to oral fluconazole due to the impaired renal function, and the subject had a worsening lung infection after 2 weeks and died of respiratory failure. The remaining subject with AIDS was treated for 12 months; the pulmonary foci were absorbed, and the subject was cured of the cryptococcal infection and is still alive.

Discussion

C. neoformans can invade various organs. The lungs are the main entrance for *C. neoformans* invasion [9]. Generally, PC patients are asymptomatic or show mild symptoms; the main manifestations are cough, an expulsion of phlegm, and dull chest pain, and there is usually a large amount of bacteria in the phlegm [10,11]. The 19 patients presented in the present study had similar clinical symptoms as those described in the literature. The imaging findings of PC are non-specific [12]. Among the 19 patients presented in this paper, those with solitary or multiple mass shadows or nodular shadows, lung patchy shadows, diffuse miliary shadows, and acute interstitial pneumonia in their chest CT scans were present in 57.9%, 26.3%, 10.5%, and 5.3%, respectively. Localized foci were generally found in the lower lobes of the lung, which was consistent with the report by Ye et al. [13]. The non-specificity in the imaging finding of PC is one of the leading causes for the clinical misdiagnosis of PC.

The pathological features of *C. neoformans* are jelly-like lesions or noncaseating granulomatous lesions. Encapsulated *C. neoformans* can be observed inside macrophages under high magnification [5]. *C. neoformans* was found in the lung biopsy specimens obtained from the 19 patients reported here, and currently, pathological diagnosis is the main approach for the diagnosis of PC. Among the 19 patients described here, *C. neoformans* was cultured from the CSF, BALF, or the secretions collected using bronchoscopic-protected brushing in 31.6% of patients, only 1 patient showed a positive result in the CSF cryptococcal capsular polysaccharide antigen test, and *C. neoformans* could not be isolated from 63.2% of patients. This result is related to minimal or absent phlegm in most patients and the localized nature of the foci. Because of the diverse imaging findings of PC, clinical misdiagnosis is common [14]. Most of the 19 PC cases collected in this study were first misdiagnosed, and the average time from the onset to diagnosis was 21 days. Based on our experience, we suggest that for patients with a lung mass or nodular lesions, the possibility of PC should be considered, and corresponding diagnostic tests should be conducted. For patients with pulmonary inflammation or with disseminated disease, the possibility of *C. neoformans* infection should be considered when antimicrobial therapy is not efficacious, particularly in patients with immunodeficiency or those receiving immunosuppressive therapy. Early percutaneous lung biopsy, transbronchial lung biopsy, or even surgical thoracoscopic lung biopsy is the key to accurate diagnosis of PC. *C. neoformans* infects both AIDS patients [2,15] and non-AIDS patients [16,17], and infects both adults and children [18]. Among the 19 PC cases collected in this study, there were 3 AIDS patients (15.7%), and non-AIDS patients accounted for 84.3%. Among them were 1 patient with rheumatic autoimmune disease and 2 patients who had undergone organ transplantation and had received continuous glucocorticoid or immunosuppressive therapy; these patients all had impaired cellular immune function, which is the root cause for *C. neoformans* infection. Three patients with malignant tumors received earlier induction chemotherapy, and their humoral and cellular immune functions were suppressed, which is also related to *C. neoformans* infection. The primary diseases in the 2 diabetic patients and 4 tuberculosis patients were not well controlled and they had a long-term medication history, and the changes in their own immune function are one of the causes for *C. neoformans* infection. The reason for the *C. neoformans* infection in the other 5 PC patients without underlying diseases was not clear, but the CD4+ and CD8+ T lymphocyte counts were low. This indicated that, despite the absence of underlying disease in these patients, their cellular immune functions decreased, which is probably the main reason for their susceptibility. In this study, 15 non-AIDS patients with localized PC

were treated with fluconazole and were cured, but treatment required 5-10 weeks. For the 3 subjects with underlying AIDS, the pulmonary manifestation involved disseminated cryptococcosis; amphotericin B treatment was administered for 6-8 weeks, and fluconazole was given orally for 6-12 consecutive months. The mortality rate was only 10.5%, and the cure rate was 89.5%. All patients were followed up for 2 years, without recurrence. Our treatment approach can therefore be recommended. As long as the renal functions in these patients are normal, the more cost-effective drug amphotericin B can be used as an alternative treatment for PC.

Acknowledgements

This study was funded as a social development project of the Science and Technology Department of Guangdong province (Grant No. 2014A020212531).

References

1. Aberg JA, Mundy LM, Powderly WG. Pulmonary cryptococcosis in patients without HIV infection. Chest. 1999;115(3):734-40.
2. Beardsley J, Denning DW, Chau NV, Yen NT, Crump JA, Day JN. Estimating the burden of fungal disease in Vietnam. Mycoses. 2015;58:101-6.
3. Nissapatorn V, Lee C, Fatt QK, Abdullah KA. AIDS-related opportunistic infections in Hospital Kuala Lumpur. Jpn J Infect Dis. 2003;56(5):187-92.
4. Visnegarwala F, Graviss EA, Lacke CE, Dural AT, Johnson PC, Atmar RL, et al. Acute respiratory failure associated with cryptococcosis in patients with AIDS: analysis of predictive factors. Clin Infect Dis. 1998;27(5):1231-7.
5. Zuo-Jun XU. Diagnosis and treatment of pulmonary cryptococcal infection. Chin J Tuberculosis Resp Dis. 2006;29:295-6.
6. Smith MB, Dunklee D, Vu H, Woods GL. Comparative performance of the RapID Yeast Plus System and the API 20C AUX Clinical Yeast System. J Clin Microbiol. 1999;37(8):2697-8.
7. Feldmesser M, Tucker S, Casadevall A. Intracellular parasitism of macrophages by Cryptococcus neoformans. Trends Microbiol. 2001;9(6):273-8.
8. Perfect JR, Dismukes WE, Dromer F, Goldman DL, Graybill JR, Hamill RJ, et al. Infectious Diseases Society of America. Clinical practice guidelines for the management of cryptococcal disease: 2010 update by the infectious diseases society of America. Clin Infect Dis. 2010;50:291-322.
9. Miyazato A. [Mechanism of Cryptococcus Meningoencephalitis]. Med Mycol J. 2016;57(1):J27-32.
10. Jan IS, Cheng WC, Lo SC, Weng MH, Lee LN. Cryptococcus neoformans in sputum and lung aspiration cytology smears. J Microbiol Immunol Infect. 2015;48(4):463-4.
11. Wang L, Shi Y, Ding Y, Wang Q, Su X. Pulmonary cryptococcosis: a retrospective analysis of 65 cases. Zhonghua Jie He Hu Xi Za Zhi. 2014;37(10):764-8.
12. Yamakawa H, Yoshida M, Yabe M, Baba E, Okuda K, Fujimoto S, et al. Correlation between clinical characteristics and chest computed tomography findings of pulmonary cryptococcosis. Pulm Med. 2015;2015:703407.
13. Ye F, Xie JX, Zeng QS, Chen GQ, Zhong SQ, Zhong NS. Retrospective analysis of 76 immunocompetent patients with primary pulmonary cryptococcosis. Lung. 2012;190(3):339-46.
14. Kerkering TM, Duma RJ, Shadomy S. The evolution of pulmonary cryptococcosis. Clinical implications from a study of 41 patients with and without compromising host factors. Ann Intern Med. 1981;94(5):611-6.
15. Gunda DW, Bakshi FA, Rambau P, Kilonzo SB. Pulmonary cryptococcosis presenting as acute severe respiratory distress in a newly diagnosed HIV patient in Tanzania: a case report. Clin Case Rep. 2015;3(9):749-52.
16. Tanaka T, Takahagi A, Tao H, Hayashi T, Yoshiyama K, Furukawa M, et al. A case of multiple pleural cryptococcosis without pleural effusion. J Thorac Dis. 2015;7(9):361-4.
17. Nabaei G, Afhami S. Disseminated cryptococcosis and active pulmonary tuberculosis co-infection in an otherwise healthy adult. Iran J Neurol. 2015;14(3):174-6.
18. Luo FL, Tao YH, Wang YM, Li H. Clinical study of 23 pediatric patients with cryptococcosis. Eur Rev Med Pharmacol Sci. 2015;19(20):3801-10.