

Title:

Clinical features of pulmonary cryptococcosis in non-HIV patients in Japan

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Abstract

Objective: To clarify the clinical features of pulmonary cryptococcosis in Japanese non-HIV population.

Methods: Retrospective investigation of 151 pulmonary cryptococcosis cases between 1977 and 2012 was executed. The underlying disease (UDs), aggravating factors, radiological characteristics, and treatment were examined.

Results: Sixty-seven patients (44.4%) had no UD. The common UD were diabetes (32.1%) followed by hematologic disease (22.6%), and collagen disease (22.6%).

Peripherally distributed pulmonary nodules/masses were most commonly seen. Lesions in the right middle lobe ($p = 0.01$) and air bronchogram ($P=0.05$) were significantly more frequent, respectively, in patients with UD than patients without them. Azoles were mainly selected for the patients without meningoencephalitis. Mean treatment duration for patients with and without UD was 6.64 and 2.87 months, respectively. Patients whose pulmonary nodules improved after treatment continued to experience gradual reduction of cryptococcosis antigen titers, even if antigen titers were positive at the time of treatment cessation. The average time for antigen titers to become negative after treatment cessation was 13.1 and 10.7 months for patients with and without UD, respectively. When groups were compared according to the presence of meningoencephalitis complications, deaths, and survivals, factors contributing to cryptococcosis prognosis included higher age, hypoproteinemia, hypoalbuminemia, steroid use, high C-reactive protein levels, and meningoencephalitis complications.

Conclusions: It is crucial to consider the presence of UD and meningoencephalitis for

the choice of antifungals and treatment duration for cryptococcosis in non-HIV patients.

Three- and six months- administration of azoles for pulmonary cryptococcosis with or without UDs, respectively is reasonable.

Key words: pulmonary cryptococcosis, computed tomography, non-HIV patient, immune status, cryptococcal antigen, meningoencephalitis

Introduction

Cryptococcus neoformans is a nonmycelial, budding encapsulated yeast-like fungus found in soil contaminated with pigeon and chicken excreta [1-4]. Inhalation of cryptococcal particles from contaminated soil into the lung is considered the usual route of human infection [2, 3]. The organism may cause isolated pulmonary infection or hematogenous dissemination involving the central nervous system (CNS), bones, and skin, mostly depending on the host immunity [2, 3]. Although cryptococcal infection can occur in individuals with normal immunity, it most commonly occurs in immunocompromised hosts. Predisposing factors are acquired immune deficiency syndrome (AIDS) and other causes of impaired T cell-mediated immunity, e.g., transplant-related immunosuppression, hematological malignancies, corticosteroid administration, and diabetes mellitus [4-6].

Although the clinical characteristics and natural history of cryptococcosis in HIV patients have been described elsewhere due to its large number, those in non-HIV patients have rarely been reported [7]. To date, few studies have reported comparative data regarding the clinical manifestations, laboratory findings, radiographic findings and

survival of patients with pulmonary cryptococcosis in Japan [8]. Additionally, very few research comparing clinical manifestation of cryptococcosis between HIV and non-HIV patients [9-11].

In Japan, the number of HIV/AIDS patients is relatively lower compared to those of other countries. However, it is increasing recently and over 20,000 of the cumulative patients are registered in Japanese government database to date (<http://www.nih.go.jp/niid/ja/aids-m/aids-iasrd/2274-kj3888.html>). Hence, the study of clinical manifestation of cryptococcal diseases in non-HIV background possess high impact. We reviewed 151 cryptococcal cases among non-HIV background and investigated the clinical features, including clinical manifestations, underlying conditions, laboratory findings, radiological features, treatment, survival, and outcomes.

Materials and methods

Patients

A retrospective cohort study was conducted by reviewing the medical records of patients who had been diagnosed with pulmonary cryptococcosis at Nagasaki University Hospital and its affiliated hospitals during the 35-year period between 1977 and 2012. The patients were grouped into 2 populations based on positivity of underlying diseases. Definite case of pulmonary cryptococcosis requires isolation or detection of *Cryptococcus* by lung specimen culture and/or by histopathological examination, and only definite cases are recruited in this study. This retrospective study including analysis and release of clinical data was approved by the ethical committee of Nagasaki University Hospital.

Clinical data

All available patient records were reviewed from the time of cryptococcal diagnosis until the patients died or were lost to follow up.

The data included clinical manifestations, underlying conditions, laboratory findings

(age, lymphocyte count, neutrophil count, immunoglobulin, serum protein, serum albumin, CD4/8 ratio, CD4 count, C-reactive protein [CRP], cryptococcal serum antigen titers) at the timing of diagnosis, radiological findings, treatment, survival, and outcome were recorded.

Eiken Latex® (Eiken Kagaku Co., Tokyo, Japan) was used for the qualitative and semi-quantitative detection of the *C. neoformans* capsular polysaccharide antigen in serum and CSF according to the manufacturer's instructions.

Interpretation of chest CT scans

The findings of chest CT scans were assessed for 1) the presence and distribution of parenchymal lesions, including nodules, masses, and consolidation; 2) the characteristics of nodules and masses; and 3) related thoracic abnormalities such as pleural effusion and lymphadenopathy according to previous reports [12]. Based on the predominant parenchymal findings from the CT scans, the morphological characteristics were classified as solitary nodule/mass (type I), multiple nodules/masses (type II), and consolidation (type III). In addition, type II was subdivided into distribution in a single

lobe (type IIa) and distribution in multiple lobes (type IIb).

Statistical analysis

We used FREQ, NPAR1WAY, and ANOVA in SAS. The chi-square test was used to compare the frequency of categorical variables (e.g., underlying disease, steroid usage).

Wilcoxon's test was used to compare age, lymphocyte count, neutrophil count, serum protein, serum albumin, CD4/8 ratio, CD4 count, CRP, and cryptococcal serum antigen titers. The Eiken Latex® latex agglutination test was used to detect cryptococcal polysaccharide. Antigen titers were transformed to the logarithm to the base 2 ($\text{Log}_2[\text{Ag} + 1]$). Ag (cryptococcal antigen titer) is expressed as 0, 1, 2, 4, ... as powers of 2 and $\text{Ag} + 1$ was expressed as $\text{Log}_2(0 + 1) = 0$.

For radiographic analysis, a chi-square test was employed to compare the presence and distribution of parenchymal lesions, nodule and mass characteristics except their number, and related thoracic abnormalities between the 2 groups. A Cochran-Armitage test was used to analyze the differences among 4 groups based on the number of nodules and masses, and among 4 morphological types based on the CT classification

between the 2 patient populations. For all statistical tests, $p < 0.05$ indicated a significant difference.

Results

Patients

One hundred fifty-one patients were diagnosed with pulmonary cryptococcosis during the 35-year period between 1977 and 2012. Sixty-seven (44.4%) occurred in the patients without underlying diseases. Forty-two were men and 25 were women.

Eighty-four cases (56.6%) were the patients with underlying disease. Thirty-eight were men and 46 were women.

Underlying diseases

Among 84 patients with underlying diseases, diabetes mellitus was most dominant (32.1%) followed by hematological diseases including human T-cell leukemia virus type-I carrier (22.6%), collagen disease including systemic lupus erythematosus, rheumatoid arthritis and others (22.6%), renal failure (16.7%), solid tumor (13.1%), chronic lung diseases including bronchiectasis, sequel pulmonary tuberculosis, and interstitial pneumonia (13.1%), liver disease including cirrhosis or hepatitis (9.5%), renal transplantation (2.4%) , and other diseases (9.5%). Treatment with glucocorticoids

(5–40 mg/day or pulse therapy) were recorded in 31 (37.0%) patients. Total of 5 patients were administrated glucocorticoids concomitantly with immunosuppressant such as cyclosporine and azathioprine.

Clinical symptoms

In 67 patients without underlying diseases, 43 (64.2%) patients were asymptomatic and detected accidentally by mass screening examination. Others had pulmonary symptoms such as cough (n=15; 22.3%), sputum (n=4; 6.0%), chest pain (n=7; 10.4%), fever (n=2; 3.0%), and others. In 84 patients with underlying disease, 39 patients (46.4%) were asymptomatic and found by abnormal chest radiograph findings taken as during routine examination of underlying diseases. Others had pulmonary symptoms such as cough (n=15; 17.6%), sputum (n=15; 17.6%), chest pain (n=3; 3.6%), fever (n=20; 23.8%), and other symptoms (n=19; 22.6%).

Laboratory findings

The laboratory findings of the patients with and without underlying disease at the timing

of diagnosis are shown in Table 1. The patients without underlying disease are statistically younger, better nutrition status reflected by higher total protein and albumin value, compared to those with underlying diseases. Serum antigen titers ($\text{Log}_2[\text{Antigen titer}+1]$) were not different statistically in both arms.

Compared to steroid non-usage patients ($n = 114$), steroid usage patients ($n= 36$) were statistically significantly older ($p < 0.0001$), had lower lymphocyte count ($p = 0.03$), higher neutrophil count ($p = 0.02$), lower blood serum protein ($p = 0.0002$), lower blood serum albumin ($p < 0.0001$), and higher CRP ($p = 0.001$). There was no significant difference in IgG, IgA, or IgM between the two groups.

CT findings

Table 2 shows the detail of CT findings between patients with or without underlying diseases. The CT findings of 81 of 151 pulmonary cryptococcosis patients were analyzed. Forty-two and 39 patients were without and with underlying diseases, respectively. The frequency of the four CT classification types based on predominant parenchymal findings and lobar distribution of the lesions is listed in Table 2. Type IIb

and type III lesions occurred more frequently in patients with underlying diseases than in those without underlying diseases. The main finding of this study is the presence of peripherally distributed multiple pulmonary nodules or masses with predominant lower lobe involvement in both patients without and with underlying diseases ($P < 0.0001$, Data not shown). Parenchymal lesions in the right middle lobe ($P = 0.01$), masses and more extensive lung involvement such as multiple lobes in distribution ($P = 0.06$) were more common in patients with underlying disease than those without underlying diseases. The number of masses (>30 -mm diameter) ($P = 0.02$) and air bronchogram ($P = 0.05$) was significantly more common in patients with underlying diseases than those without underlying diseases.

Serum cryptococcal antigen titer and radiological findings

The relationship of serum cryptococcal antigen titer and radiological findings were analyzed. Data from patients with meningoencephalitis was excluded. For patients with solitary nodules ($N = 14$), multiple nodules ($N = 34$) or consolidation ($N = 8$), comparison of the cryptococcal antigen titer ($\text{Log}_2[\text{Antigen titer} + 1]$) revealed no

significant correlation between solitary and multiple nodules (N.S.); however, a significant higher cryptococcal antigen titer was observed in consolidation when compared with solitary nodules ($p < 0.01$) and multiple nodules ($p < 0.05$) (Figure 1).

Antifungal treatment

The mainstay of initial treatment in both groups was azoles. Fifty-six (83.6%) patients without underlying diseases were initially treated with azoles, fluconazole (including Fos-fluconazole) ($n=21$, 31.3%), itraconazole ($n=4$, 5.9%), voriconazole (VRCZ) or miconazole (MCZ) ($n=10$, 15.0%), azoles + 5-fluorocytosine (5-FC) ($n=15$, 22.4%), or amphotericin B ($n=3$, 4.5%). To patients with underlying diseases, fluconazole (including Fos-fluconazole) ($n=30$, 35.7%), azoles plus 5-FC ($n=17$, 20.2%), itraconazole ($n=6$, 7.1%), VRCZ or MCZ ($n=10$, 11.9%), and amphotericin B±5-FC ($n=4$, 4.8%) were administered.

The median duration of fluconazole (Fos-fluconazole) treatment was 90 days (range 60–110 days) for patients without underlying disease. Five patients did not receive any

antifungal drugs because they were initially suspected of having lung cancer and underwent pneumonectomy. Three patients without underlying disease were initially observed without any antifungals under informed consent, as the size and number of radiological abnormalities were reduced spontaneously and the serum cryptococcal antigen titers decreased within a few months.

Antifungal agents were administered for 6 months in all patients except three with refractory cryptococcosis among population with underlying diseases.

Transitional serum Cryptococcal antigen titer before and after treatment

Forty definite cases (14 cases with underlying diseases and 26 cases were without underlying disease) which were followed until the serum cryptococcal antigen became negative or up to 45 months after treatment.

Titer changes in the latex agglutination test before and after therapy in patients without underlying disease and with underlying disease are depicted in Figure 2 and 3, respectively. The mean duration of treatment for 14 patients without underlying diseases was 2.87 months. The cryptococcal antigen titer decreased for all cases after

antifungal treatment. The cryptococcal antigen became negative in 13 of 14 cases following administration of antifungal agents. The mean period from treatment cessation to negative antigen observation was 10.7 months.

The mean duration of treatment for 26 patients with underlying diseases was 6.64 months. In the 22 cases where antigen titers became negative after treatment, the mean period from treatment cessation to negative antigen observation was 13.1 months (Figure 3). No significant difference was observed between two groups. The serum antigen titers were decreased after antifungals were discontinued.

Comorbid cryptococcal meningoencephalitis

In 151 of pulmonary cryptococcosis cases, 122 patients were performed lumbar puncture test. Fourteen patients (9.3%) presented CNS involvement. Four and 10 patients were without and with underlying disease, respectively (Table 3).

Fever (57.1%; 8/14), headache (35.7%; 5/14), and appetite loss or vomiting (35.7%; 5/14) suggesting CNS infection were observed. However, 2 patients had no CNS symptoms. The radiographic findings in patients with CNS involvement are shown in

Table 3. Solitary or multiple nodules, consolidation, reticular shadow, granular shadow or mixed findings were observed in meningoencephalitis patients. Pulmonary shadows were seen in both lungs in 7 patients. In meningoencephalitis patients, all patients without underlying disease were improved; however, 6 of 10 patients with underlying disease died. One patient did not have underlying disease; the other had a previous history of diabetes.

Pulmonary cryptococcosis patients who died

Overall cryptococcal-related mortality was 9.4% (14/151). Mortality in patients with underlying diseases was 16.7% (14/84). No patients without underlying diseases died of cryptococcal-related disease. Some patients harbored underlying conditions such as hematologic disease and malignant tumors; the cause of death in many cases could be traced to worsening underlying disease.

The radiographic finding of most of the patients who died revealed consolidation (57.1%; 8/14) and in both lungs (85.7%; 12/14), suggesting disseminated cryptococcal infection. Six of the deceased patients were diagnosed by autopsy without any

anti-cryptococcal treatment. The progression of serum antigen titers could not be observed continuously in the patients that died. Only one patient relapsed (Table. 4).

Laboratory data correlated to meningoencephalitis and outcome.

The clinical features including laboratory data in the patients with and without cryptococcal meningoencephalitis, and outcome are compared in Table 5.

Older age, lower lymphocyte counts, higher neutrophil counts, lower serum total protein, lower serum albumin, low CD4/8 ratio, high CRP and higher cryptococcal antigen titer are related to comorbidity of meningoencephalitis. For prognosis, older age, higher neutrophil counts, lower serum total protein, lower serum albumin, and higher CRP are correlated to death. Comorbidity of meningoencephalitis and poor outcome shares same factors.

Discussion

In Japan, the majority of cryptococcosis is seen in non-HIV patients. In the present study, we reviewed the clinical features of 151 pulmonary cryptococcosis in non-HIV patients in Nagasaki, Japan.

Roughly half of the cryptococcosis patients did not have underlying diseases and almost half of patients (82/151, 54.3%) without respiratory symptoms were found accidentally by mass screening examination or routine chest X-ray check. In Japan, there is a unique medical insurance system which allowed people easy and cheap access to medical examination or annual medical check. This background may cause the potential bias in this study.

It is generally recommended that HIV-negative patients with cryptococcal pneumonia undergo routine lumbar puncture to attempt to identify asymptomatic or subclinical CNS involvement that may require more potent or aggressive therapy [13]. However, 2010 updated IDSA guidelines commented as follows, “In nonimmunocompromised patients with pulmonary cryptococcosis, consider a lumbar puncture to rule out asymptomatic CNS involvement. However, for normal hosts with asymptomatic

pulmonary nodule or infiltrate, no CNS symptoms, and negative or very low serum cryptococcal antigen, a lumbar puncture can be avoided (B-II)” [14]. Of the 14 patients with cryptococcal meningoencephalitis in this study, 4 did not have underlying disease. Six patients with meningoencephalitis were dead (eight were survived) and the correlation between comorbidity of meningoencephalitis and poor outcome shares same clinical factors. However, the possibility of existence of meningoencephalitis links significant poor prognosis was not evaluated due to the low number of cases. Of those, 1 patient had solitary nodules, 1× negativity for cryptococcosis antigens, and no CNS disorders. We believe that the necessity of CSF examinations should be debated thoroughly. There has been ongoing discussion regarding the need for lumbar punctures in patients without CNS symptoms. The 2007 Guidelines [15] also recommend lumbar puncture to identify asymptomatic CNS involvement.

To our knowledge, this review of pulmonary cryptococcosis constitutes the largest report to date describing and comparing chest CT findings in non-HIV patients both with and without underlying diseases. Additionally, since many cases were diagnosed

from mass screening check-up, it is important to investigate the unique features of radiological findings. Similar to recent studies of immunocompetent hosts [16-19] and non-AIDS individuals [20-22], the most common CT feature was the presence of peripherally distributed multiple pulmonary nodules or masses with predominant lower lobe involvement in both patients without and with underlying diseases. Although the number of nodules or masses in previous reports has varied, there was no significant difference in the frequency between multiple nodules or masses (type II) and single lesions (type I) in our series. Multiple nodules or masses distributed in multiple lobes (type IIb) also tended to occur more frequently in patients with underlying diseases than in patients without underlying diseases.

Cryptococcal antigen is widely recognized to have both diagnostic and prognostic value for cryptococcosis. Lu et al. reported on the CSF titer change in the latex agglutination test before and after therapy in non-HIV cryptococcal meningitis. The cryptococcal antigen titer in CSF decreased after therapy for every case and correlated with fungal clearance; however, cryptococcal antigen can remain at low titers for long periods after therapy, even when fungal smear and/or culture become negative. Previous

study suggested that the cryptococcal antigen test may not be used as an index of cure [23].

In this study, serum cryptococcal antigen in pulmonary cryptococcosis patients can remain at low titers for long periods after therapy. However, the titers continuously decrease after effective therapy. Our data demonstrate that the cryptococcal antigens in pulmonary cryptococcosis remain detectable even months following successful therapy, suggesting that the cryptococcal antigen test may not be used as an index of cure or decision of discontinuation of treatment.

The first line antifungal drugs were selected because the facilities that reported these cases were conducting clinical trials for antifungal drug development. Moreover, because the guidelines for cryptococcosis management had not yet been presented before 2007, azole-type drugs + 5-FC were used as pulmonary cryptococcosis treatment for a certain period even in patients without meningoencephalitis.

After the 2007 Guidelines [15] was published, treatment was conducted according to these guidelines' recommendations.

The IDSA guideline for pulmonary cryptococcosis in non-HIV, non-immunosuppressed patients states that for mild-to-moderate symptoms, fluconazole (400 mg/day orally) should be administered for 6–12 months; persistently positive serum cryptococcal antigen titers are not criteria for continuance of therapy (B-II) [14].

Conversely, the 2007 Guidelines recommend administering 400 mg/day oral fluconazole for 3 months for immunocompetent pulmonary cryptococcosis patients, and for 6 months for patients with underlying disease [15].

In our experience, with the exception of some severe cases, the duration of treatments as recommended in the 2007 Guidelines [15] appears appropriate.

In the management of pulmonary cryptococcosis in non-HIV patients, it is important to confirm the presence of underlying disease and encephalomeningitis complications, and a careful treatment plan for patients with severe underlying diseases.

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Table 1. Characteristics of patients with cryptococcosis with or without underlying disease

		State of underlying conditions						
		Patients without underlying diseases			Patients with underlying diseases			Wilcoxon test
criteria	n	median	IQR	n	median	IQR	p value	
1	Age	67	41	31	84	63	18.5	<0.0001
2	lymphocyte counts	54	1985.5	573.0	74	1429.0	1218.0	0.03
3	Neutrophil counts	55	3245.0	2403.0	75	4680.0	4273.0	0.02
4	IgG	32	1262.0	435.0	37	1343.0	891.0	0.31
5	IgM	32	142.0	71.3	37	145.0	96.0	0.49
6	IgA	31	249.0	142.0	37	275.0	169.0	0.39
7	Total protein	48	6.90	0.70	65	6.40	1.40	0.0002
8	Serum albumin	42	4.39	0.54	60	3.60	1.29	<0.0001
9	CD4/CD8	31	1.50	0.79	37	1.42	0.90	0.53
10	CRP	40	0.21	0.33	49	0.84	3.60	0.001
11	Cryptococcal antigen	56	16.00	124.00	63	32.00	252.00	0.35
12	CD4 counts	17	874.80	282.20	19	637.00	915.20	0.99

IQR : Inter Quartile Range, CRP, C-reactive protein.

Table 2. Comparison of CT findings of cases with pulmonary cryptococcosis with or without underlying disease

	Patients without underlying diseases		Patients with underlying diseases		p value
	n=42	%	n=39	%	
Mean of Age (range)	47.4(15-80)		61.4(19-79)		
Sex(men: women)	26:16:00		18:21		
Presence of parenchymal lesions					
Nodule and masses	42	100.0	39	100	
Consolidation	3	7.1	7	18.0	0.14
solitary nodules/mass(type I)	14	33.3	9	23.1	0.30
multiple nodules	25	59.5	29	74.4	0.15
single lobe limited (type IIa)	10	23.8	5	12.8	0.06
multiple lobe limited (type IIb)	15	35.7	24	61.5	
consolidation (type III)	3	7.1	7	17.9	0.14
Distribution of parenchymal lesions					
Lobar distribution					
Right upper lobe	13	31.0	16	41.0	0.34
Right middle lobe	6	14.3	15	38.5	0.01
Right lower lobe	26	61.9	28	71.8	0.34
Left upper lobe	10	23.8	6	15.4	0.34

Lingura	3	7.1	5	12.8	0.39
Left lower lobe	20	47.6	17	43.6	0.71
Contact with pleura	33	78.6	33	85.0	0.48
<hr/>					
Size(mm)					
1-30	31	73.8	16	41.0	0.02
31-	5	11.9	11	28.0	
Number					
1	15	35.7	11	28.0	0.85
2-4	10	23.8	10	26.0	
5-9	7	16.7	7	18.0	
10-	9	21.4	11	28.0	
Border					
Well-defined / ill-defined	34 / 8	(81/19)	34 / 5	(87/13)	0.44
Margin					
Smooth / irregular / speculated	20 / 21 / 19	(48/50/45)	12 / 27 / 20	(31/69/51)	0.25
Convergence of bronchi and vessel	35	83.3	31	79.0	0.65
Pleural identification	19	45.2	22	56.0	0.32
<hr/>					
Internal characteristics					
Air-bronchogram	21	50.0	28	72.0	0.05

Cavitation	12	28.6	16	41.0	0.24
Calcification	1	2.4	0	0.0	0.33
CT halo sign	25	59.5	18	46.0	0.22
Satellite lesion	27	64.3	26	67.0	0.82

Table 3. Summary of cases with cryptococcal meningoencephalitis

Age	Sex	Symptoms	Radiographic findings	Site of shadow	Underlying diseases	Steroid usage	Cr Ag	Treatment	Prognosis
28	M	headache	solitary nodule	rt. middle robe	(-)	(-)	NA	AMPH-B + 5-FC	improved
59	M	cough, sputum	consolidation	both lungs	(-)	(-)	NA	AMPH-B + 5-FC, MCZ	improved
62	M	headache, cough	multiple nodules	both lungs	(-)	(-)	512	FLCZ + 5-FC	improved
80	M	fever, headache, vomiting, lumbago	consolidation	rt. middle robe	(-)	(-)	2046	FLCZ + 5-FC	improved
48	F	fever, appetite loss	reticular shadow	both lungs	SLE, NS	(+)	NA	(-)	death
75	F	fever, headache, consciousness disorder	granular shadow	both lungs	ATL	(-)	2048	FLCZ+5-FC	death
60	F	no symptom	solitary nodule	rt. lower lobe	DM	(-)	1	MCZ +5-FC, FLCZ	**
73	F	fever, cervical lymph node enlargement	**	**	ATL	**	128	FLCZ+5-FC	death
61	M	headache, change in personality	consolidation	rt. lower lobe	SLE、 APS	(+)	1024	AMPH-B + 5-FC	improved
76	F	fever, vomiting	multiple nodules + consolidation	both lungs	DM, RA	(-)	NA	FLCZ+5-FC	improved

86	M	fever, respiratory discomfort	diffuse GGA	both lungs	CRF	(-)	1024	F-FLCZ	death
74	F	fever	multiple nodules	rt. lower lobe	RA, CRF, amyloidosis	(+)	1024	FLCZ+5-FC+AMPH-B	death
74	M	fever, appetite loss, general fatigue	solitary nodule	lt. lower lobe	Wegener 's granulomatosis	(+)	256	FLCZ+5-FC	improved
60	M	appetite loss, general fatigue	multiple nodules	both lungs	ATL, DM	(+)	NA	FLCZ+5-FC	death

ATL, Adult T cell leukemia; SLE, systemic lupus erythematosus; NS, nephrosis syndrome; DM, diabetes mellitus; RA, rheumatoid arthritis; CRF, chronic renal failure; APS, anti-phospholipid antibody syndrome, rt., right, and lt., left; Cr Ag, Cryptococcal antigen titer; NA, not available. AMPH-B, amphotericin B; 5-FC, flucytosine; FLCZ, fluconazole; F-FLCZ, fos-fluconazole; MCZ, miconazole.

** : Data was missed

Table 4. Summary of patients who died of cryptococcosis

Age	Sex	Radiographic findings	Site of shadow	Underlying condition	Steroid usage	Treatment	Meningoencephalitis	Diagnostic methods
40	M	consolidation	both lungs	ML	(-)	MCZ+5FC, AMPH-B+5FC	(-)	TBLB
57	M	consolidation	both lungs	DM.LC.ATL, HCC	(+)	AMPH-B+5FC , ITZ, FLCZ	(-)	TBLB
64	F	consolidation	both lungs	ATL	(-)	FLCZ	(+)	sputum culture
69	F	consolidation	rt. upper lobe	LK	(+)	FLCZ+5FC	not done	BALF culture
86	M	diffuse GGA	both lungs	CRF	(-)	F-FLCZ	(+)	sputum, CSF, Urine, blood culture
48	F	reticular shadow	both lungs	SLE, NS	(+)	(-)	(+)*	autopsy
75	F	consolidation	both lungs	Bladder tumor	(-)	(-)	(-)	autopsy
62	F	consolidation	both lungs	PN, ARF	(+)	FLCZ, MCZ	(-)	autopsy
75	F	granular shadow	both lungs	ATL	(-)	(-)	(+)*	autopsy
82	F	consolidation	both lungs	RA, miliary TB	(+)	(-)	not done	autopsy

		interstitial shadow						
66	M	(by underlying disease)	both lungs	IP	(+)	(-)	not done	autopsy
73	F	consolidation	both lungs	ATL	**	FLCZ+5-FC	not done	sputum culture
				RA, CRF,				
74	F	multiple nodules	rt. lower lobe	secondary amyloidosis	(+)	FLCZ+5-FC+AMPH-B(+)		BALF, CSF, blood culture
60	M	multiple nodules	both lungs	ATL, DM	(+)	FLCZ+5-FC	(+)	BALF, CSF, blood, prostatic fluid culture

GGA, ground-glass attenuation; rt., right; ML, malignant lymphoma; DM, diabetes mellitus; LC, liver cirrhosis; ATL, adult T cell leukemia; HCC, hepatocellular carcinoma; LK, lung cancer; CRF, chronic renal failure; SLE, systemic lupus erythematosus; NS, nephrosis syndrome; PN, polyarteritis nodosa; ARF, acute renal failure; RA, rheumatoid arthritis; TB, tuberculosis; IP, interstitial pneumonia; TBLB, transbronchial biopsy; BALF, bronchial alveolar lavage fluid, and CSF, cerebrospinal fluid. AMPH-B, amphotericin B; 5-FC, flucytosine; FLCZ, fluconazole; ITZ, itraconazole; F-FLCZ, fos fluconazole; MCZ, miconazole.

** : Data was missed

Table 5. Comparison of clinical characters of patients with or without cryptococcal meningoencephalitis and prognosis

criteria	Comorbid Meningoencephalitis							Prognosis						
	Without Meningoencephalitis			With Meningoencephalitis			Wilcoxon test	improved			Died			Wilcoxon test
	n	median	IQR	n	median	IQR	p value	n	median	IQR	n	median	IQR	p value
1 Age	110	54.5	30.0	14	67.5	15.0	0.004	123	55.0	30.0	13	69.0	13.0	0.004
2 lymphocyte counts	98	1914.0	1006.0	11	1023.0	926.0	0.005	108	1874.0	992.5	11	1370.0	2792.0	0.26
3 Neutrophil counts	99	3355.0	2658.0	11	7626.0	4280.0	0.001	109	3780.0	2780.0	11	7832.0	4908.0	0.003
4 IgG	61	1290.0	661.0	5	1140.0	468.0	0.78	63	1307.0	600.0	5	1343.0	694.0	0.87
5 IgM	61	142.0	79.0	5	103.0	115.3	0.33	63	142.0	81.0	5	157.0	115.3	0.80
6 IgA	60	259.0	179.0	5	328.0	133.0	0.74	62	259.0	167.0	5	454.0	256.0	0.34
7 Total protein	88	6.90	0.90	11	5.90	1.50	0.02	99	6.90	0.90	10	5.30	0.50	0.0003
8 Serum albumin	82	4.10	0.80	9	3.10	1.60	0.01	90	4.10	0.80	8	2.65	1.13	0.0006
9 CD4/CD8	60	1.47	0.74	3	0.96	0.43	0.03	61	1.45	0.72	5	1.34	0.85	0.45
10 CRP	69	0.26	1.02	4	10.59	13.44	0.004	76	0.29	1.08	5	4.12	12.29	0.003
11 Cryptococcal antigen	90	16.00	120.00	7	1024.00	1920.00	0.02	103	16.00	252.00	11	128.00	1016.00	0.15
12 CD4 counts	30	719.50	474.60	1	421.50		0.30	34	734.60	480.80	1	95.00		0.09

IQR : Inter Quartile Range, CRP, C-reactive protein.

Figure legend

Figure 1. The relationship of serum cryptococcal antigen titer and radiological findings. The morphological characteristics were classified as solitary nodule/mass (type I), multiple nodules/masses (type II), and consolidation (type III). Cryptococcal antigen titers were transformed to the logarithm to the base 2 ($\text{Log}_2[\text{Ag} + 1]$). Data from patients with meningoencephalitis was excluded.

Figure 2. Transitional change of the latex agglutination test after treatment in patients without underlying diseases. Solid line indicates the duration of treatment and dotted line indicates the following time after treatment. The mean duration of treatment for 14 patients without underlying diseases was 2.87 months. The cryptococcal antigen titer decreased for all cases after antifungal treatment. The cryptococcal antigen became negative in 13 of 14 cases following administration of antifungal agents. The mean period from treatment cessation to negative antigen observation was 10.7 months. The patients were followed until the serum antigen become negative or up to 45 months

Figure 3. Transitional change of the latex agglutination test after treatment in patients with underlying diseases. Solid lines and dotted lines denote treatment duration and duration of follow-up after treatment, respectively. The mean duration of treatment for 26 patients with underlying diseases was 6.64 months. In the 22 cases where antigen titers became negative after treatment, the mean period from treatment cessation to negative antigen observation was 13.1 months. The patients were followed until the serum antigen become negative or up to 45 months

Figure 1. The relationship of serum cryptococcal antigen titer and radiological findings.

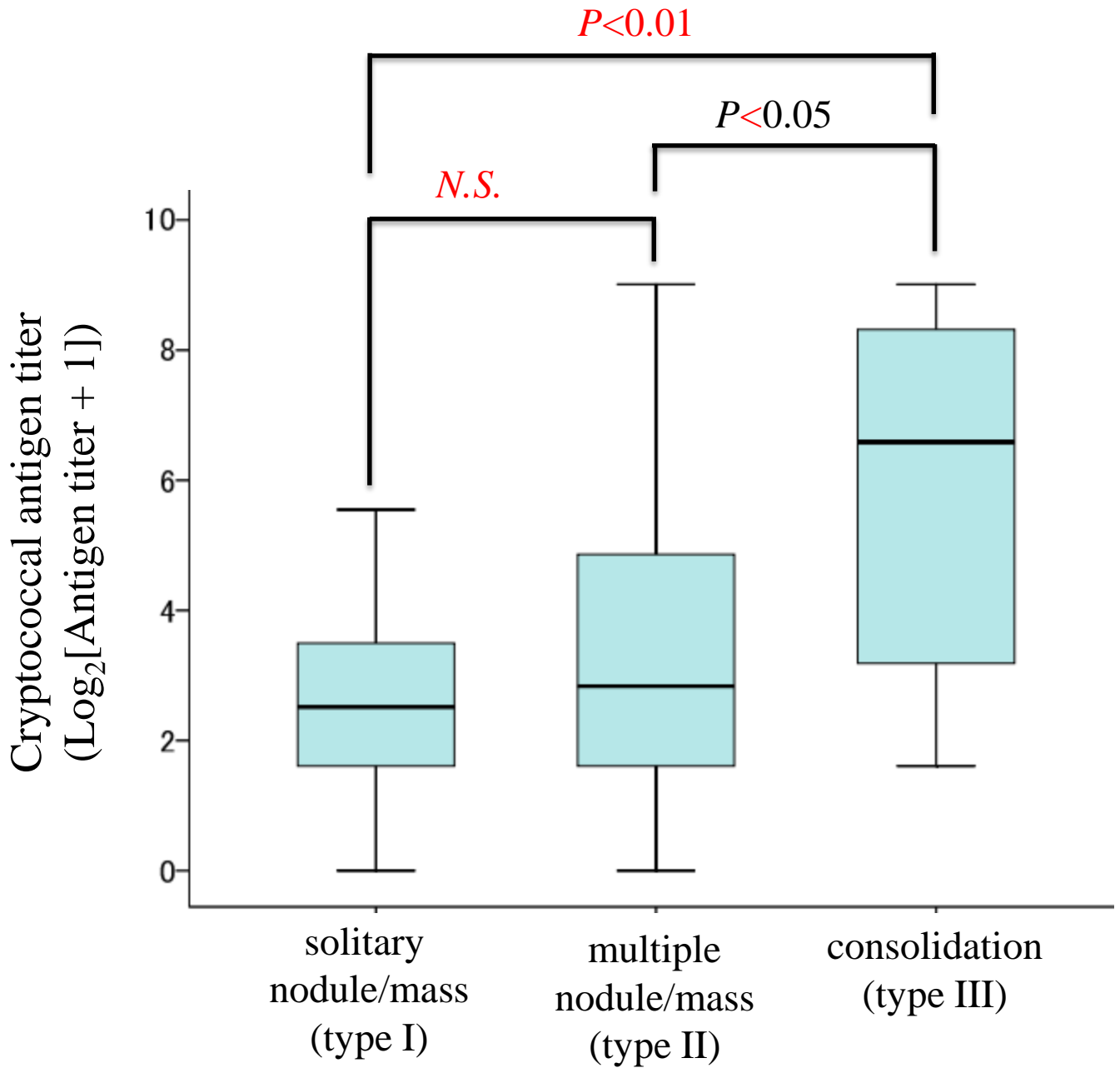


Figure 3. Transitional change of the latex agglutination test after treatment in patients with underlying diseases

(serum antigen titer)

