Cryptococcemia. An analysis of 28 cases with emphasis on the clinical outcome and its etiologic agent

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Summary Clinical protocols of 28 cases of cryptococcemia studied between April 1995 and November 2002 were reviewed. The varieties of *Cryptococcus neorformans*, the underlying disease, and the severity and outcome of the disease were emphasized. Most patients were immunossupressed (89.3% with AIDS) and *Cryptococcus neoformans* var. *grubii* was the main recovered variety (92.8%). Regardless of antifungal treatment, in-hospital mortality was 41% strongly associated with APACHE II score, >14 (p<0.01).

Key words Cryptococcemia, *Cryptococcus neoformans* varieties *gattii* and *grubii,* AIDS

Criptococemia. Un análisis de 28 casos, con énfasis en la evolución clínica y el agente etiológico

Resumen Se revisan 28 casos de criptococcemia observados durante un periodo de siete años (1995-2002). Se destacan las variedades de *Cryptococcus neoformans*, las condiciones predisponentes y la gravedad y pronóstico de la enfermedad. La mayoría de los pacientes eran inmunodeficientes (89,3% con sida) y la variedad *grubii* fue la principal variedad aislada (92,8%). La mortalidad intrahospitalaria (41% de los pacientes) estuvo asociada con el resultado del APACHE II, principalmente si este era > 14 (p<0,001).

Palabras clave Criptococcemia, *Cryptococcus neoformans* variedades *gattii* y *grubii,* Sida

As previously reported, *Cryptococcus neoformans* has been isolated from blood cultures in around 10 to 30% of patients with cryptococcosis [2,10]. In contrast with other clinical features, cryptococcemia is usually associated with a poor prognosis in patients with cryptococcal meningitis [1,2].

There are only two clinical studies on cryptococcemia [4,6]. However, these studies did not address what varieties of *C. neoformans* were isolated from patients. The purpose of this study was to report the underlying diseases, the variety of etiologic agent involved, severity and outcome of the disease of patients with cryptococcal fungemia.

Patients and methods

Between April 1995 and November 2002, we retrospectively studied all patients with at least one positive blood culture (Isolator®) for *C. neoformans* at the Mycology Laboratory, Santa Casa Complexo Hospitalar. The clinical records of these patients were reviewed, and we were primarily interested in collecting data such as: sex, age, sites/organs infected; underlying diseases; CD4 cells count; titres of cryptococcal antigens in their sera and cerebrospinal fluid (CSF); antifungal treatment; time elapsed between blood culture and beginning of treatment and death; and the variety of *C. neoformans* recovered on culture. Mental status at time of diagnosis and cerebrospinal fluid characteristics were not recorded due to inconsistencies in patient files.

Clinical outcome was based on the report of hospital discharge or death of the patient while hospitalized. Disease severity was estimated using Acute Physiology and Chronic Health Evaluation (APACHE) II score. The observed mortality was compared with estimated mortality by the score, based in a study of 5,815 critically ill patients in intensive care units [5]. Analysis of the data was done with Epi-Info 6.0 software, and P values below 0.05 were statistical significance.

Results

During April 1995 and November 2002, 28 patients were included in this study. The patients were mainly white (89.3%) and male (78.6%), and the mean age was 36 years old (standard deviation, SD ± 13, range, 8-64 years). Tables 1 and 2 show a summary of the 28 patient's clinical data. With the exception of one immunocompetent

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Table 1. General characteristics of the 28 patients with cryptococcemia.

Case	Sex	Age	Underlying disease	CD4 cells/mm^3	Other sites of infection			Variety
					CNS	Urine	Other	
1	M	40	AIDS	5	Pos.	NA		*grubii*
2	M	31	AIDS	148	Neg.	NA		*grubii*
3	M	35	AIDS	17	Pos.	NA		*grubii*
4	M	44	AIDS		Pos.	Pos.		*grubii*
5	M	39	AIDS	14	NA	NA		*grubii*
6	F	27	AIDS	12	Pos.	Pos.	BAL	*grubii*
7	M	28	AIDS	30	Pos.	NA		*grubii*
8	M	29	AIDS	14	Pos.	Pos.		*grubii*
9	M	21	AIDS	13	Pos.	NA		*grubii*
10	F	22	AIDS	6	Pos.	Pos.	BAL	*grubii*
11	M	52	AIDS		Pos.	Pos.	BAL	*grubii*
12	M	31	AIDS	50	Pos.	NA		*grubii*
13	M	32	AIDS	87	NA	Pos.	Lung biopsy, prostate	*grubii*
14	F	41	AIDS		Pos.	Neg.		*grubii*
15	M	30	AIDS	56	Pos.	Neg.		*grubii*
16	M	35	AIDS	154	Pos.	Neg.		*grubii*
17	M	40	AIDS	103	Neg.	NA	BAL	*grubii*
18	M	59	Lung transplantation		Pos.	Pos.	BAL, skin	gattii
19	M	34	AIDS		Pos.	Pos.		*grubii*
20	M	25	AIDS		NA	NA		*grubii*
21	F	7	AIDS		Pos.	Neg.		*grubii*
22	M	64	Immunocompetent		Pos.	Neg.	BAL	gattii
23	M	NA	AIDS		Pos.	Neg.	BAL	*grubii*
24	F	59	AIDS		Pos.	Neg.		*grubii*
25	F	27	AIDS		Pos.	Neg.	BAL	*grubii*
26	M	43	AIDS		Pos.	NA	BAL	*grubii*
27	M	34	AIDS	23	Pos.	Pos.		*grubii*
28	M	46	Liver cirrhosis		NA	Pos.		*grubii*

Legend: M = Male; F = Female; NA = Data not available; CNS = Central nervous system; BAL = Bronchoalveolar lavage; Pos.= Positive; Neg.= Negative.

Table 2. Patients with cryptococcemia and their response to antifungal treatment, disease severity (APACHE II score) and clinical outcome.

Case	Antifungal treatment; Maintenance	APACHE II	Outcome
1	Flu 400 mg/day (2 months) followed by AmB 15 mg/day	9	Death
2	AmB 50 mg/day plus 5-FC 2g qid (37 days); Flu	10	Death
3	AmB 50 mg/day plus 5-FC 2g qid (31 days); AmB	7	Discharge
4	AmB 20 mg/day (3 days) followed by Flu 400 mg/day	30	Death
5	AmB 50mg/day	26	Death
6	AmB 50 mg/day (18 days); Flu	14	Discharge
7	AmB 15mg/day	16	Death
8	NA	NA	NA
9	AmB 12.5 mg/day	11	Discharge
10	AmB 50 mg/day (25 days); Flu	10	Discharge
11	AmB 35 mg/day (3 days) followed by Flu 400 mg/day	34	Death
12	AmB 50 mg/day (21 days); Keto	13	Discharge
13	Keto 400 mg/day (5 days) followed by Flu 400 mg/day	7	Discharge
14	NA	NA	NA
15	AmB 50 mg/day (41 days); Flu	7	Discharge
16	AmB 60 mg/day (20 days); Flu	12	Discharge
17	AmB 40 mg/day (27 days); Flu	8	Discharge
18	AmB 40 mg/day plus 5-FC 1.5g qid (31 days); Flu	13	Discharge
19	NA	NA	NA
20	NA	NA	Discharge
21	NA	NA	NA
22	AmB 30mg/day	28	Death
23	NA	NA	NA
24	AmB 50 mg/day	10	Death
25	AmB 50mg/day (28 days); Flu	10	Discharge
26	NA	NA	NA
27	AmB 60 mg/day (45 days); Flu	8	Discharge
28	Flu 100mg/day (5 days) followed by AmB 30mg/day	11	Death

Legend: NA = Not available; Flu = Fluconazole; AmB = Amphotericin B; 5-FC = Flucytosine; Keto = Ketoconazole; APACHE = Acute Physiology and Chronic Health Evaluation.

patient, the remaining patients had the following underlying diseases: AIDS (acquired immunodeficiency syndrome) (89.3%), lung transplantation patient (3.6%) and liver cirrhosis (3.6%). The CD4 cells mean count was 49 cells/mm^3 (range, 5-154, SD ± 51), for the 60% of AIDS patients in those on this examination were performed.

C. neoformans isolated from these patients were identified as variety *grubii* in 26 patients (92.8%) and variety *gattii* in the remaining two patients (7.1%). With one exception (case 28, a patient with liver cirrhosis) *C. neoformans* var. *grubii* infected 25 patients with AIDS ($p<0.01$, relative risk, 3.0, confidence interval, 0.6-14.8). *C. neoformans* variety *gattii* was recovered from an immunocompetent patient and from a lung transplanted patient.

Besides blood cultures, *C. neoformans* was also isolated from the following body sites: CSF (89.3%), respiratory tract (35.7%), urine (35.7%), skin lesions (3.6%) and prostate (3.6%). Only one patient had a negative culture for *C. neoformans* in the CSF. No association between underlying disease and fungal isolation from CSF or other body site was observed. Cryptococcal antigen in the sera of eight patients showed titres ranging from 1:6 to 1:524,288 (median, 1:2,048). All patients with positive CSF in culture had positive CSF cryptococcal antigen with titres ranging from 1:1 to 1:65,536 (median, 1:4,096).

Systemic antifungal therapy was initiated within 24 h after positive blood culture in 95% of the patients. The clinical outcome of six patients could not be obtained. Among the remaining 22 patients the in-hospital mortality was 41%. Mean time for death was 25 days (range, 1-100, SD ± 35), and 44% of deaths occurred within 24 h after positive blood sampling. For patients that discharged, the mean time of hospitalization was 43 days (range, 7-112, SD ± 29). Two patients voluntarily left the hospital before discharge (cases 9 and 15) and one patient asked to be discharged before his evaluation was completed (case 17).

The APACHE II score in seven patients was not obtained. In the remaining 21 patients, the mean value was 14 (SD ± 8). There was no association between APACHE II values and underlying diseases or the variety of *C. neoformans* involved. The mean score for survivors was 10 (SD ± 3), significantly lower than the mean for non-survivors (19, SD ± 10) ($p=0.02$). There were a strong association between APACHE II > 14 and mortality ($p<0.01$). Table 3 compares the estimated mortality on critical patients with the observed mortality stratified according to APACHE II score. There was no association between mortality or duration of hospitalization and the isolated variety of *C. neoformans*, underlying disease, CD4 cells count, antifungal therapy and serum or CSF cryptococcal titres.

Table 3. Comparison of the estimated mortality obtained by APACHE II score and the mortality observed in this study.

APACHE II score	In-hospital mortality (%)	
	Estimated	Observed
0-4	4	*
5-9	6	17
10-14	12	30
15-19	22	100
20-24	40	*
25-29	51	100
30-34	71	100
≥ 35	82	*

Legend: APACHE = Acute Physiology and Chronic Health Evaluation.
* No patient in the range.

Discussion

Based on the immunologic properties of its capsular polysaccharides, *C. neoformans* has been classified in four distinct serotypes: A, B, C, and D. However, using phenotypical, genetical, biochemical and epidemiological differences, these serotypes have been further grouped in three different varieties: *grubii* (serotype A) [3], *gattii* (serotypes B and C) and *neoformans* (serotype D). Variety *grubii* is an opportunistic fungus with a ubiquitous distribution affecting mainly AIDS patients. Whereas, var. *gattii* is found in tropical and subtropical areas as a primary pathogen [9] rarely associated with AIDS, even in the endemic areas [7,8]. Interestingly, variety *neoformans* is usually restricted to North European countries. The majority patients in our study (89.3%) had AIDS, supporting previous epidemiological studies involving variety *grubii* as the etiologic agent.

Only few data are available on the clinical significance of cryptococcemia, most them anecdotal reports. In the first published series of cases, involving 15 patients diagnosed and treated before 1983, Perfect et al [6] concluded that the progress of underlying diseases and the outcome of concomitant infections were more important determinants of survival than cryptococcemia. Jean et al [4], studied clinical features and natural history of 52 patients with cryptococcemia on patients with AIDS between 1981 and 2001. In this study, 46% of the patients were HIV-infected, and 81% presented with sepsis; fourteen-day mortality was 31%, with 68% of deaths occurring 30 days after blood sampling. Antifungal therapy started within 48 h after blood culture improved survival.

The high mortality rate among patients with cryptococcemia in our study (41%) showed strong association with the APACHE II score ($p<0.01$). Jean et al [4] noted that mortality was also significantly higher among patients with cryptococcemia and APACHE II ≥ 20. However, using other parameters, this association was not significant, unlike variables such as liver cirrhosis and severity of sepsis. In our patients, systemic antifungal therapy was initiated early, frequently before or simultaneously to blood culture. In spite of it, death usually occurred within 24 h after blood sampling. The number of AIDS patients (89.3%) could explain the high mortality found in this study.

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