

CLINICAL AND EPIDEMIOLOGICAL FEATURES OF 123 CASES OF CRYPTOCOCCOSIS IN MATO GROSSO DO SUL, BRAZIL

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SUMMARY

To identify the clinical and epidemiological profile of cryptococcosis diagnosed at the University Hospital of the Federal University of Mato Grosso do Sul, Brazil, medical records of 123 patients admitted from January 1995 to December 2005 were analyzed. One hundred and four cases (84.5%) had HIV infection, six (4.9%) had other predisposing conditions and 13 (10.6%) were immunocompetent. Male patients predominated (68.3%) and their age ranged from 19 to 69 years (mean: 35.9). Most patients (73.2%) were born and lived lifelong in the state of Mato Grosso do Sul. Involvement of the central nervous system occurred in 103 patients (83.7%) and headache and vomiting were the most frequent symptoms. In 77 cases it was possible to identify the *Cryptococcus* species: 69 (89.6%) *C. neoformans* and eight (10.4%) *C. gattii*. Amphotericin B was the drug of choice for treatment (106/123), followed by fluconazole in 60% of cases. The overall lethality rate was 49.6%, being 51% among the HIV infected patients and 41.2% among the non-HIV infected ($p > 0.05$). Although cryptococcosis exhibited in our region a similar behavior to that described in the literature, the detection of an important rate of immunocompetent individuals and five *C. gattii* cryptococcosis in HIV-infected patients is noteworthy.

KEYWORDS: Epidemiology; Cryptococcosis; Brazil; *Cryptococcus gattii*; *Cryptococcus neoformans*.

INTRODUCTION

Cryptococcosis is an important opportunistic systemic mycosis affecting mainly patients with the acquired immunodeficiency syndrome (AIDS). The disease is caused by two species of the *Cryptococcus neoformans* complex, *C. neoformans* (serotypes A, D, and AD) and *C. gattii* (serotypes B and C). The latter, previously regarded as a variety of *C. neoformans*, has now gained full species status^{1,2}. Usually present in urban pigeon droppings, *C. neoformans* has a worldwide distribution and is a common opportunistic infection, whereas *C. gattii* can be found in hollow trees, usually causes the disease in non-immunocompromised individuals and behaves as a primary pathogen^{3,4}. The mycosis is acquired by inhalation of viable propagules of this agent, causing a pulmonary infection from which it can spread to other organs, particularly the central nervous system^{5,6}. The paucity of data on human cryptococcosis in the Brazilian central-western state of Mato Grosso do Sul encouraged us to carry out the present study.

MATERIALS AND METHODS

The study was performed at the University Hospital of the Federal University of Mato Grosso do Sul, a reference center for infectious diseases, analyzing the records of patients diagnosed with

cryptococcosis from January 1995 to December 2005.

The diagnosis of cryptococcosis was based on cerebrospinal fluid (CSF) direct microscopy with India ink and/or isolation of the fungus in culture. The *C. neoformans* complex was identified by the observation of dark brown yeast colonies after cultivating the suspected yeast on Niger seed agar (NSA) medium and the species was identified in canavanine-glycine-bromothymol blue (CGB) medium¹¹.

Demographic, clinical, and laboratory data were recorded in a specific data bank and the analysis was performed with the Epi Info 3.32 program. Student's *t* test was applied to identify differences between means and the chi-squared test was used for differences between proportions, with Fisher's test being applied when the expected value was less than 5. A significance level of 0.05 was adopted.

The study was approved by the Research Ethics Committee of UFMS.

RESULTS

During the eleven years period (1995-2005), 123 cases of cryptococcosis were recorded. Out of them, 104 (84.5%) were HIV

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Table 1
Clinical manifestations of 123 cases of cryptococcosis, according to HIV co-infection. University Hospital, UFMS, Campo Grande, MS, Brazil (Jan. 1995-Dec. 2005)

Clinical manifestation	HIV-positive		HIV-negative		Total		p
	n = 104	%	n = 19	%	n = 123	%	
General manifestations							
Fever	73	70.9	14	73.7	87	71.3	NS
Weight loss	67	65.7	7	36.8	74	61.2	0.01
Neurological manifestations							
Headache	82	79.6	11	57.9	93	76.2	0.03
Mental confusion	24	23.5	5	26.3	29	24.0	NS
Vomiting	63	61.2	10	52.6	73	59.8	NS
Motor deficit	6	5.8	0	0	6	4.9	NS
Visual alterations	24	23.1	5	26.3	29	23.6	NS
Neck stiffness	50	48.5	6	31.6	56	45.9	NS
Cutaneous manifestations							
Skin lesions	1	1.0	0	0	1	0.8	NS
Respiratory manifestations							
Cough	52	50.5	7	36.8	59	48.4	NS

NS: nonsignificant difference

infected. Of the 19 (15.4%) patients without HIV infection, six had other predisposing conditions: systemic lupus erythematosus (two cases), neoplasia (two cases), kidney transplant (one case), and tuberculous pericarditis treated with prednisone (one case). Fig. 1 shows the distribution of the cases per year according to HIV infection.

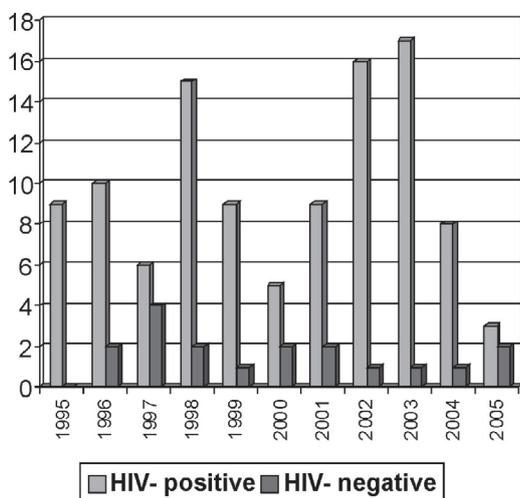


Fig. 1 - Distribution of 123 cases of cryptococcosis, with and without HIV co-infection, by year of diagnosis. UFMS teaching hospital, Campo Grande, MS, Brazil, January 1995 to December 2005.

Eighty-four patients (68.3%) were male, 39 (31.7%) were female, and their ages ranged from 19 to 69 years old, with a mean of 35.9 years. No significant difference in mean age was found between groups with and without HIV infection.

Most patients (88, or 71.5%) were born in counties located in the state of Mato Grosso do Sul and 35 (28.4%) elsewhere, of whom 15 (12.2%) were from the Southeastern region, eight (6.5%) from the

Northeastern region, four (3.3%) from the Southern region, and six (4.9%) came from other Center-Western states. For two patients this information was not available. Most patients (120; 97.6%) had been referred from localities in Mato Grosso do Sul, two (1.6%) from the adjoining state of Mato Grosso, and only one from the neighboring state Rondônia. Ninety patients (73.2%) were born and lived lifelong in Mato Grosso do Sul.

Clinical findings are summarized in Table 1, where the predominance of neurological alterations was observed, followed by general and respiratory manifestations. Cerebrospinal fluid (CSF) exam was performed on 106 patients (90 HIV-infected and 16 noninfected). *C. neoformans* was found in direct microscopy of India ink preparation in 93.4% (85/91) and in cultures of 80.2% (81/101). Histopathology of lung (two cases) and prostate (one case) biopsies diagnosed the remaining cases. The cytometry and biochemistry results are shown in Table 2.

Table 2
Cellularity and biochemical features of cerebrospinal fluid in 106 patients with cryptococcosis, according to HIV co-infection. University Hospital, UFMS, Campo Grande, MS, Brazil (Jan. 1995-Dec. 2005)

	HIV-positive (n = 90)		HIV-negative (n = 16)		p
	Mean	Median	Mean	Median	
Leukocytes (cells/mm ³)	64.3	6.5	207.4	119	0.00
Proteins (mg/dL)	107.0	73.0	247.6	105	0.00
Glucose (mg/dL)	37.5	35.0	27.1	22	0.06

In 77 cases it was possible to identify the *Cryptococcus* species: 69 (89.6%) cases of *C. neoformans* and eight (10.4%) of *C. gattii*. Although five HIV-infected patients had cryptococcosis by *C. gattii*, the infection by *C. neoformans* prevailed in both groups (HIV-infected

(60/65; 92.3%) and HIV-noninfected (9/12; 75%) patients ($p > 0.05$).

Most cases (90.2%) were treated with amphotericin B (106/123) and fluconazole (57/123). 5-Flucytosine was associated with amphotericin B in two cases and with itraconazole in one. The overall lethality rate was 49.6%, reaching 51% among HIV-infected and 41.2% among HIV-noninfected patients ($p > 0.05$).

DISCUSSION

This is the first clinical and epidemiological study of cryptococcosis cases in Mato Grosso do Sul, where it predominates among HIV-infected patients (84.6%), in agreement with the literature⁷⁻⁹. Among the non-AIDS patients with cryptococcosis, nearly 50% had no obvious predisposing factor¹⁰. The association of cryptococcosis with neoplastic and lymphoproliferative disorders, and with the use of immunosuppressive drugs, has been well established^{9,11,12}.

Figure 1 shows that the distribution of cases by year before 2003 does not seem to follow any special patterns. The decreasing trend observed since 2003 is possibly related to the use of combined antiretroviral therapy.

As observed by others^{5,7,13}, male patients predominated in our study. In the recent AIDS era, cryptococcosis accompanies clearly the gender distribution of HIV infection. Thus, the predominance of males reflects the predisposing condition. Given the worldwide trend of feminization of AIDS¹⁴, this situation is subject to changes. Similarly, the predominance of cryptococcosis in the third and fourth decades of life, in accordance with other studies^{5,13}, coincides with the age range where HIV is most prevalent.

The fact that most patients (73.2%) were from and lived lifelong in the state of Mato Grosso do Sul suggests that the AIDS epidemic has already hit the Center-Western Region of Brazil in an important manner.

The most frequent clinical manifestations involved the central nervous system and included headache and vomiting, in addition to general symptoms such as fever and weight loss, are in agreement with others^{7,13}. The occurrence of fever was similar in both groups, but the weight loss was more frequent among HIV carriers ($p < 0.05$), a finding possibly related to the consumptive nature of AIDS. Headache reported by 79.6% of HIV-infected patients was significantly more frequent than the 57.9% reported by patients of the other group. In contrast, visual alterations (blindness, visual impairment and double vision) reported by 23.6% of all patients, were similarly distributed between the groups, a result that agrees with the literature¹⁶.

Cutaneous manifestations were found only in HIV-infected patients, resulting from hematogenous spread and increased immunologic deficit³.

Since CSF cellularity and biochemical alterations are not specific, differential diagnosis should consider principally tuberculous meningoencephalitis¹³. Major CSF alterations were lymphocytic pleocytosis, hyperproteinorrachia and hypoglycorrachia¹³, as observed also in the present study. Significant differences were found in CSF

protein concentration and cellularity, with lower mean leukocyte counts in the HIV-infected group. Of the HIV-infected patients, 45.6% had 5 leukocytes/mm³ or less, a finding that stresses the importance of surveying fungi even when cytological parameters are normal. Other investigators also report decreased inflammatory response in CSF in patients with AIDS¹⁷.

The high sensitivity of direct microscopy of CSF with India ink for detecting *Cryptococcus* spp. is well known⁹. Association with culture on Sabouraud agar medium increases the likelihood of identifying these fungi. Because *C. neoformans* and *C. gattii* usually cause meningoencephalitis and because late diagnosis compromises prognosis, search for capsulated yeasts in India ink and culture on appropriate media should be part of routine procedures for every CSF sample collected from patients suspected of central nervous system inflammatory disease.

In cases of expansive masses, given the clinical and imaging similarities with neoplasia, diagnosis usually is reached by histopathological examination, as it occurred with three patients in the present study. Besides, the importance of medical requests for microbiological examination including surveys by direct microscopy and cultures for fungi should be stressed. Through proper and careful mycological processing of each clinical specimen it is not only possible to confirm diagnosis, but also to identify the species.

In the present cases two rare occurrences were observed: one of prostate mass involvement and one of fungemia concurrent with *Histoplasma capsulatum*, as both fungi have been isolated from the same blood samples.

C. gattii is likely to cause disease in healthy hosts (primary pathogen)¹⁸, whereas *C. neoformans* attains predominantly immunodepressed individuals^{12,19}. Although AIDS associated cryptococcosis by *C. gattii* has not been often reported, five of our eight patients affected by this species also had HIV infection.

Antiretroviral therapy is intended to eliminate viral replication and thus prevent devastation of the immune system, particularly of CD4+ lymphocytes. HIV-infected patients who are given combined antiretroviral therapy have a lower incidence of cryptococcosis¹⁹. Control of this opportunistic mycosis in patients with AIDS depends therefore on the use of adequate antiretroviral treatment, with a resulting improvement in the level of cellular immunodepression¹¹⁻¹².

RESUMO

Características clínicas e epidemiológicas de 123 casos de criptococose observados em Mato Grosso do Sul, Brasil

O perfil clínico-epidemiológico de 123 casos de criptococose diagnosticados no Hospital Universitário da Universidade Federal de Mato Grosso do Sul, no período de janeiro de 1995 até dezembro de 2005, foi estudado retrospectivamente. Cento e quatro (84,9%) casos tinham associação com HIV, seis (4,9%) tinham outra condição predisponente e 13 (10,6%) eram imunocompetentes. Houve predomínio do sexo masculino (68,3%) e a idade variou de 19 a 69 anos (média de 35,9 anos). A maioria (73,2%) era natural e procedente

de Mato Grosso do Sul. O envolvimento do sistema nervoso central ocorreu em 103 (83,7%) pacientes e os sintomas mais frequentes foram cefaléia e vômitos. Em 77 casos foi possível identificar a espécie do agente, sendo 69 (89,6%) *C. neoformans* e oito (10,4%) *C. gattii*. O antifúngico mais utilizado foi anfotericina B (106/123) seguido de fluconazol em aproximadamente 60% dos casos. A taxa de letalidade foi de 49,6%, sendo 51% entre os pacientes infectados pelo HIV e 41,2% entre os não infectados pelo HIV ($p > 0,005$). Apesar da criptococose observada em nossa região apresentar comportamento semelhante ao descrito na literatura, chama a atenção a importante taxa da micose em imunocompetentes e cinco casos de infecção por *C. gattii* em pacientes HIV-positivos.

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