Clinical microbiological case: fever and headache in a heavy consumer of eucalyptus extract

C. Tascini, S. Ferranti, G. Gemignani, F. Messina and F. Menichetti

U. O. Malattie Infettive, Azienda Ospedaliera Pisana, Via Paradisa 2, 56100 Pisa, Italy

Accepted 9 January 2002

Please refer to the article on page x of this issue to view the questions to which these answers refer.

CLINICAL OUTCOME

Latex antigen testing of the cerebrospinal fluid (CSF) and blood were positive for *Cryptococcus neoformans* with a titre of 1 : 4096 and the organism grew on Sabouraud agar. Subculture on L-lahavanine glycine bromothymol blue (CGB) agar yielded blue colonies, identifying a *C. neoformans* var. *gattii*. This yeast was found to be susceptible, using the E test method, to amphotericin B, itraconazole, fluconazole and 5-flucytosine.

A diagnosis of cryptococcal meningitis was made and a search for risk factors showed that: human immunodeficiency virus (HIV) serology was negative, CD4+ lymphocyte levels were normal in both percentage (30%) and absolute number (700 cells/mL) and total haemolytic complement levels were within normal limits. No cancers were found, nor was there any history of use of immunosuppressive drugs. A contrast-enhanced computerized tomographic scan of the brain and sinuses was unremarkable.

Fluconazole, at the dose of 1200 mg, given intravenously, was started but 1 week later, because of the persistence of fever and headache, treatment was changed to liposomal amphotericin B (5 mg/kg/day). After 4 weeks it was changed again to itraconazole and after 2 more weeks of treatment the patient flew back to the USA, completely recovered from his symptoms.

Corresponding author and reprint requests: C. Tascini, U. O. Malattie Infettive, Azienda Ospedaliera Pisana, Ospedale Cisanello, Via Paradisa 2, 56100 Pisa, Italy
Tel: +39 050 996735
Fax: +39 050 571021
E-mail: carlotascini@katamail.com

The cytokine levels (measured using enzyme-linked immunosorbent assay) on CSF and serum at the beginning of the disease and after 4 weeks of antifungal therapy are reported in Table 1. A remarkable increase of interleukin-10 (IL-10) in the CSF was noted at the beginning of the disease, this cytokine decreased after effective antifungal therapy. Interferon-g (IFN-g) was never stimulated during the disease.

DIAGNOSIS

The diagnosis was meningitis due to *Cryptococcus neoformans* var. *gattii*.

DISCUSSION

Cryptococcus neoformans var. *gattii* meningitis is rare among immunodepressed people and affects especially immunocompetent patients exposed to the flowering of the Australian tree *Eucalyptus camaldulensis* [1,2]. This meningitis is rare in Europe [3–5]. The patient described here had travelled in many countries but had never travelled across Australia. The only risk factor for this patient appeared to be the improper and heavy use of eucalyptus extract.

Eucalyptus extracts could contain *C. neoformans* var. *gattii* yeasts derived from the eucalyptus trees, but the culture of the gel used recently by the patient was negative for bacteria and fungi. However, this finding might not exclude the possibility that the patient was exposed to *C. neoformans* var. *gattii* through eucalyptus extract.

In relation to cytokine levels in patients with cryptococcosis, if we take as reference values in CSF those found by Chaka et al. [6], we observed that at the beginning of the disease in our patient, there were high levels of IL-10 and tumour necrosis fator-a (TNF-a) and low levels of IL-1b, IFN-g and IL-12. In the serum we observed low levels of IL-10, IL-1b, IFN-g, IL-12, but in contrast, a moderate level of TNF-a was observed. It is notable that IL-10 was almost undetectable in the serum; this could mean that the production of IL-10 was not regulated by the presence of *C. neoformans* var. *gattii* capsular antigen, because the cryptococcal antigen latex titre was the same in the CSF and the serum. After 4 weeks of therapy there was a reduction of the production of the anti-inflammatory cytokine IL-10 in the CSF, but on the other hand,

Table 1 Cryptococcal antigen titres and cytokine levels in CSF and serum of a patient with *C. neoformans* var. *gattii* meningitis, at the beginning of the disease and after 4 weeks of effective therapy

	Time		
	0	4 weeks	Normal values
Latex titre			
CSF	1 : 4096	1 : 1024	
Serum	1 : 4096	1 : 1024	
IL-10 (pg/mL)			
CSF	146.8	35.9	
Serum	0.2	0.1	<8.9
TNF-α (pg/mL)			
CSF	156	160	
Serum	21	18	<14
IL-1β (pg/mL)			
CSF	35	29	
Serum	21	23	<21
IFN-γ (UI/mL)			
CSF	0.7	0.5	
Serum	0	0.3	<1.2
IL-12 (pg/mL)			
CSF	6	16	
Serum	27	60	80

there was an increased production of the pro-inflammatory cytokine IL-12, both in the serum and in the CSF.

This finding led us to postulate that IL-10 is stimulated by the yeast and suppresses the inflammatory response [7], but only in the CSF. In fact, IL-10 was also almost undetectable in the serum at the beginning of the meningitis. On the contrary, IL-12 was produced as the fungus burden decreased after effective therapy, counteracting the effect of IL-10 and stimulating an immune response. IFN-g was never stimulated either in the CSF or the serum; and this could explain why this meningitis had a very long evolution.

This finding agrees with those of Lortholary et al. who found increased levels of CSF IL-10 in patients with cryptococcosis but without HIV infection [8]. IL-10 CSF level might be used, after more extensive studies, as a prognostic factor in *C. neoformans* var. *gatti* meningitis.

We started antifungal therapy with high-dose fluconazole, but changed to liposomal amphotericin B due to persistent fever and headache. Although high-dose fluconazole might be effective from the beginning of cryptococcal meningitis [9], there are some studies that have hypothesized that amphotericin B may be more effective in the first 2 weeks of therapy because of its more fungicidal effect [10].

In this patient there were none of the common risk factors found in cryptococcal meningitis. However, the improper use of eucalyptus extracts, which might be a reservoir for *C. neoformans* var. *gattii*, could be considered a risk factor for this kind of meningitis.

REFERENCES

1. Chen S, Sorrell T, Nimmo G *et al.* Epidemiology and host- and variety-dependent characteristics of infection due to *Cryptococcus neoformans* in Australia and New Zealand. Australasian Cryptococcal Study Group. *Clin Infect Dis* 2000; 31: 99–508.
2. Ellis DH, Pfeiffer TJ. Natural habitat of *Cryptococcus neoformans*, var. g*attii*. *J Clin Microbiol* 1990; 28: 1642–4.
3. Kwon-Chung KJ, Bennett JE. Epidemiologic differences between the two varieties of *Cryptococcus neoformans*. *Am J Epidemiol* 1984; 120: 23–30.
4. Barò T, Torres-Rodriguez JM, De Mendoza MH, Morera Y, Alia C. First identification of autochthonous *Cryptococcus neoformans* var. *gattii* isolated from goats with predominantly severe pulmonary disease in Spain. *J Clin Microbiol* 1998; 36: 458–61.
5. Montagna MT, Tortorano AM, Fiore L, Ingletti AM, Barbuti S. *Cryptococcus neoformans* var. *gattii* en Italie. Not I. Premier cas autochthone de meningite à sèrotype B chez un sujet VIH positif. *J Mycol Med* 1997; 7: 90–2.
6. Chaka W, Heyderman R, Gangaidzo I *et al.* Cytokine profiles in cerebrospinal fluid of human immunodeficiency virus-infected patients with cryptococcal meningitis: No leukocytosis despite high Interleukin-8 levels. *J Infect Dis* 1997; 176: 1633–6.
7. Vecchiarelli A, Retini C, Monari C, Tascini C, Bistoni F, Kozel TR. Purified capsular polysaccharide of *Cryptococcus neoformans* induces interleukin-10 secretion by human monocytes. *Infect Immun* 1996; 64: 2846–9.
8. Lortholary O, Dromer F, Mathoulin-Pelissier S, *et al.* Immune mediators in cerebrospinal fluid during cryptococcosis are influenced by meningeal involvement and human immunodeficiency virus serostatus. *J Infect Dis* 2001; 183: 294–302.
9. Menichetti F, Fiorio M, Tosti A *et al.* High-dose fluconazole therapy for cryptococcal meningitis in patients with AIDS. *Clin Infect Dis* 1996; 22: 838–40.
10. van der Horst CM, Saag MS, Cloud GA *et al.* The National Institute of Allergy and Infectious Diseases Mycoses Study Group and AIDS Clinical Trials Group. Treatment of Cryptococcal Meningitis Associated with the Acquired Immunodeficiency Syndrome. *N Engl J Med* 1997; 337: 15–21.