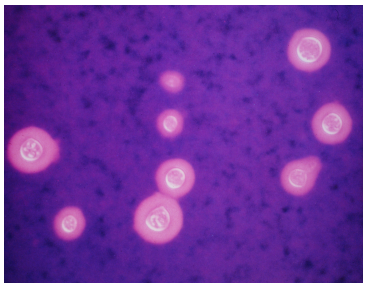
GAFFI Fact Sheet

Cryptococcal meningitis

Introduction

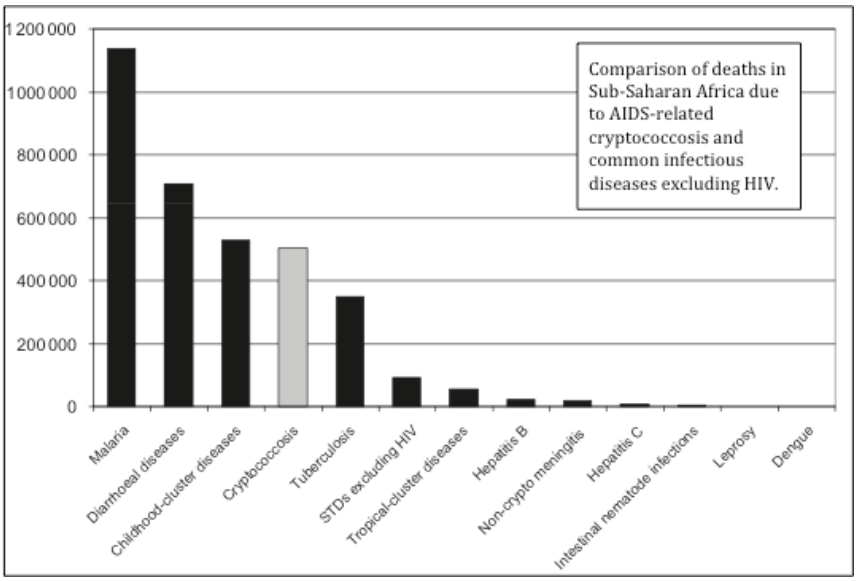
Cryptococcal meningitis is caused by one of the two closely related encapsulated environmental fungi, *Cryptococcus neoformans (serotypes A, D, and AD)* and *C. gattii (serotypes B and C). C. neoformans* has a world-wide distribution, while *C. gattii* is concentrated in tropical and sub-tropical zones (although *C. gattii* infections have recently emerged on Vancouver Island and the adjacent mainland in British Columbia, Canada). The frequency and circumstances of human exposure to these organisms are not precisely understood, but exposure is assumed to occur following inhalation from the environment.



Infection is usually controlled effectively by the immune system, but remains latent, so that, if immune function later wanes, due to HIV/AIDS, immunosuppressing medication, or another condition, disease develops, in particular a life-threatening meningitis or meningoencephalitis. *C. neoformans* causes most infections in HIV-infected patients; *C. gattii,* in particular, also causes disease in apparently immunocompetent persons, treatment response is usually slow and these patients are at risk for developing mass lesions (cryptococcomas). Person to person or animal to person transmission does not occur.

Incidence and disease burden

Cryptococcal meningitis remains a very common opportunistic infection in patients with late stage HIV-infection. In fact, 80% - 90% of patients with cryptococcosis in the US have HIV/AIDS. In sub-Saharan Africa, 15%-30% of all patients with AIDS develop cryptococcal disease, Zimbabwe exceptionally has very high incidence with as many as 88% of patients with AIDS presenting with cryptococcal meningitis as HIV/AIDS defining opportunistic infection. Despite expansion of antiretroviral programs, cases have not decreased in most African countries. Furthermore, treatment is unsatisfactory: in Africa, mortality has ranged from 24% at 10 weeks to 95% at 12 weeks depending on the initial therapeutic regimen (see table, below). A recent CDC analysis estimated that in Africa, cryptococcosis-associated mortality at 3 months is ~70% [1] . The combination of high incidence and difficulties with treatment result in cryptococcosis being a very common cause of death in AIDS patients, accounting for 13 and 17% of all deaths in two cohorts of HIV patients from Uganda. In comparison, TB caused 6% and 5% of deaths respectively, in these



studies. While tuberculosis is much more common, treatment is available and effective; the high case fatality rate for cryptococcosis leads to a high death toll. A recent analysis estimated cryptococcal-related deaths at 181,100 (range 119,400 to 234,300) globally, with 75% (135,900) occurring in sub-Saharan Africa [2].

Clinical Features

HIV-associated cryptococcal meningitis and meningoencephalitis usually presents in late stage patients (CD4 cell count <100 cells/μl) as a subacute or chronic events with symptoms progressing over 1-2 weeks. These symptoms include headache, fever, and malaise and later, vomiting, double vision, normal or mildly elevated temperature, nausea and vomiting (with increased intracranial pressure), reduced vision, seizures and altered mental status. Signs, if present, may include meningism, papilloedema, cranial nerve palsies (particularly 6th nerve palsies reflective of raised cerebrospinal fluid (CSF) pressure, which is a very common complication) and reduced conscious level or coma.

In non HIV-infected patients, and especially in apparently immune-competent patients with *C. gattii* infection, the duration of presenting symptoms may be longer, and focal neurological lesions, hydrocephalus, and focal lung lesions are more common, reflective of an increased host inflammatory response, compared with HIV-infected patients.

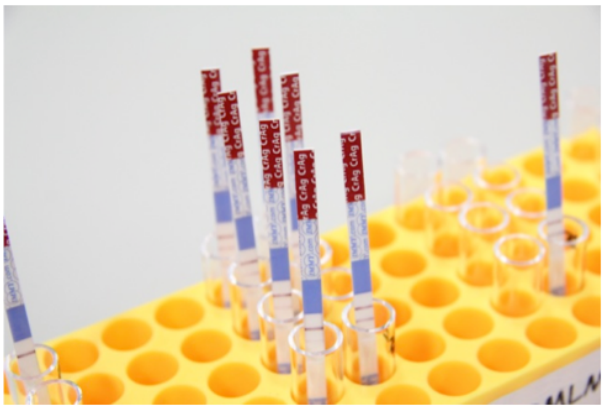
Lung involvement is probably under-recognized, especially in HIV-infected patients[1]. Immunocompetent patients may present with localized, self-limiting lung nodules, while in the immunocompromised, lung involvement is more diffuse and progressive and often leads to dissemination. In addition to the central nervous system and lung, just about any other organ system can be involved, including skin, lymph nodes and bone.

There is a period of asymptomatic disseminated infection prior to meningitis developing. Some patients have a positive blood culture for *C. neoformans*, but usually such patients are identified with a cryptococcal antigen test. The interval between asymptomatic antigenaemia and meningitis, which represents a window of opportunity to intervene and prevent meningitis, is about 3 weeks [3] – this is the basis of screening programs (see below).

Diagnosis of cryptococcosis

The cryptococcal antigen test is perhaps the best test in microbiology – it is fast, inexpensive and has a sensitivity and specificity of >99% [4]. The test can be done on serum, plasma, finger prick blood, CSF and urine, a probably bronchoalveolar lavage although there are few data [5–8]. It is less sensitive on saliva [9]. All species of *Cryptococcus* causing meningitis are detected by the antigen test [4]. The cryptococcal lateral flow assays have a very high sensitivity (>95%) and sensitivity (~99%) in serum and CSF and sensitivity of ~85% in urine. It is stable at all room temperatures, quick to perform (10-15mins) and can be performed at the bedside.

Diagnosis of cryptococcal meningitis is usually not a problem in HIV-infected patients, if a lumbar puncture is done, since the organism load is high. The antigen test is highly sensitive for CSF ~100%. The simple, quick, and widely available, Indian ink examination of the cerebrospinal fluid (CSF) is positive in around 70–80% of AIDS patients, while those with infection who are India ink negative are invariably positive on antigen testing and culture of the CSF. However, lumbar puncture is moderately invasive and may require travel to a referral centre, meaning that in many patients presenting with a headache to primary care clinics, diagnosis is initially delayed, with potentially fatal consequences.



However, cryptococcal antigen is also positive in blood in nearly all patients with HIV-associated cryptococcal meningitis and is detectable in urine in most patients. While previously, antigen testing was done using a latex agglutination format requiring some processing of the blood sample in a central laboratory, two point-of-care, "dipstick" antigen tests, are both more sensitive and potentially much more widely accessible [4,10]. These antigen tests enable rapid diagnosis in clinic settings and rapid referral for assessment and earlier treatment.

In non-HIV-associated disease, especially in apparently immune-competent patients, the sensitivity of all diagnostic tests is reduced due to lower organism loads and the diagnosis is occasionally difficult to exclude. Large-volume CSF cultures and repeated lumbar punctures may occasionally be needed in this setting.

Screening patients with low CD4 counts for cryptococcal disease

The development of latex agglutination (sensitivity: 93-100%, specificity: 93-98%) and enzyme immunoassay (sensitivity and specificity ~99%) techniques for the rapid detection of the cryptococcal polysaccharide capsular antigen (CrAg) has revolutionized the diagnosis and screening of cryptococcosis [4,11].

HIV/AIDS patients with CD4 counts of less than 100cells/mm^3 are at the highest risk for developing cryptococcal disease. *Cryptococcus* spp. shed their polysaccharide capsular antigen very early into the bloodstream during dissemination i.e. between 5-234 (median 22) days before the development of clinical cryptococcal disease – asymptomatic antigenaemia [3]. The knowledge of this phenomenon allows a "window" of opportunity for a subclinical cryptococcal disease to be diagnosed through routine screening of high risk patients.

Patients who screen negative are enrolled into routine HIV care as per national/WHO guidelines and those who screen positive are referred for lumbar puncture for CSF analysis to rule out cryptococcal meningitis. "Pre-emptive" antifungal (fluconazole) therapy is initiated for patients with asymptomatic cryptococcaemia while patients with positive CSF analysis for cryptococcal meningitis are started on optimal therapy.

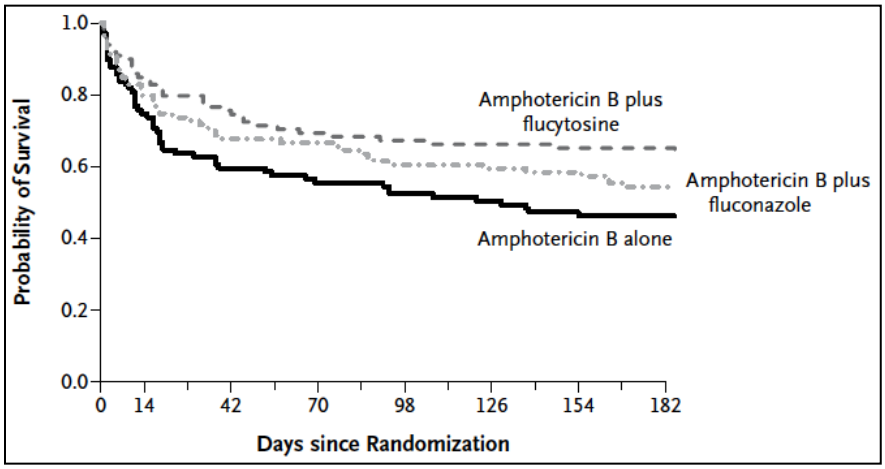
CrAg screening is a very cost-effective public health intervention associated with reduced mortality by decreasing the incidence of cryptococcal meningitis among HIV patients especially in a population with high prevalence (>3%) of CrAg antigenaemia [7,12,13].

In screen positive patients, about 40% have meningitis, usually asymptomatic. Cryptococcal antigen titres in blood are higher in those with meningitis. A cutoff titre of 1:160 separates most of those with and without meningitis. One of the commercially available lateral flow cryptococcal antigen tests has a double concentration line, approximating to a titre of 1:160.

As some patients are reluctant to have a lumbar puncture, this additional data can be used to treat the patient as if they have meningitis [14].

Treatment of Cryptococcal Meningitis

World Health Organization (WHO, 2013) recommends daily intravenous amphotericin B (0.7-1mg/Kg/d of amphotericin B deoxycholate) plus 6-hourly oral flucytosine (100mg/Kg/d) for 2 weeks as the gold standard for induction therapy, as this combination results in the most rapid control of infection [15–17]. Flucytosine with amphotericin B synergistically results in quicker clearance of viable yeast from the cerebrospinal fluid than is seen with amphotericin B alone or amphotericin B plus fluconazole. Induction therapy is considered successful only after CSF culture is negative for *Cryptococcus* spp in a patient with significant clinical improvement. In HIV-infected patients, after 2 weeks, given the duration-dependent side effects of amphotericin B and flucytosine, patients should be switched to fluconazole at a dose of 400mg/d for a minimum of 8-10 weeks. Although less rapidly fungicidal, fluconazole is well-tolerated and widely available even in resource-limited settings. If flucytosine is not available, fluconazole at 800mg/d can be given with amphotericin B as alternative induction therapy.



Unfortunately in many centers in Africa, 2 weeks induction therapy with amphotericin B cannot be safely sustained. In addition to the costs of the drug, which may be substantial in local terms, there are the requirements for hospitalization and intravenous drug administration. Nursing time and expertise in siting and maintaining IV access for a drug which causes considerable phlebitis, additional IV fluid and electrolyte replacement, and regular, rapid and reliable laboratory monitoring for renal function, electrolytes, and hemoglobin are all significant hurdles for many places. Without proper monitoring, amphotericin B can lead to fatal hypokalaemia. In these circumstances, amphotericin B courses of 5-7 days may have very significant benefit while being much less toxic and requiring less intense monitoring.

In the absence of amphotericin B, high dose oral fluconazole (1200 mg/d for the first 2 weeks) plus flucytosine is more effective than fluconazole alone. However, since flucytosine is not currently widely available, fluconazole monotherapy is still the most widely used initial treatment across Sub-Saharan Africa. 10-week mortality with this treatment, even using the higher doses of fluconazole now recommended, is in the order of 50-60%.

Antiretroviral therapy and maintenance therapy

In HIV-associated meningitis, fluconazole is given at 400-800 mg/d for 2 to 10 weeks, then at 200 mg/d until immune reconstitution has occurred with antiretroviral therapy. Antiretroviral therapy is currently started after 2 to 6 weeks of antifungal treatment, in

order to prevent other HIV-related complications without exacerbating immune reconstitution reactions. Initiation of antiretroviral therapy at the time of diagnosis of cryptococcal meningitis increases mortality [18].

Raised intracranial pressure

Raised CSF pressure is extremely common and, if untreated, is associated with worse symptoms (blindness or permanent neurological deficits) and increased acute mortality. CSF pressure should be monitored carefully during the induction phase. The uses of mannitol, acetazolamide or corticosteroid to control CSF pressure unassociated with immune reconstitution inflammatory syndrome (IRIS) are of no proven value. There is good evidence that careful mechanical drainage with repeated lumbar punctures (or, in the most severe cases, a lumbar drain or intraventricular drain) relieves symptoms and reduces mortality [18,19]. However, across Africa, manometers to measure CSF pressure are not generally available, and, although attempts to measure and manage CSF pressure using intravenous giving sets have been made, currently CSF pressure is not managed in most resource limited settings.

Alternative amphotericin B formulations

Lipid formulations of amphotericin B (i.e. liposomal amphotericin B 3-4mg/kg/d) can be used in place of conventional amphotericin B and are less nephrotoxic, although not more effective. They are preferred for cryptococcal meningitis in patients immunosuppressed through organ transplantation, who do not tolerate conventional amphotericin B well. Amphotericn B-based induction is often prolonged beyond 2 weeks in these cases, and in the non-HIV, non-transplant patient group including those who are immune-competent and those infected with *C. gattii*.

Recent data from a phase II randomized controlled non-inferiority trial from Tanzania showed that single dose 10mg/kg of liposomal amphotericin B is well tolerated with a non-inferior early fungicidal activity (EFA) compared to 14-day courses of 3mg/kg liposomal amphotericin B in the treatment of HIV-associated cryptococcal meningitis [20].

Patients with severe non-meningeal cryptococcosis should also be treated initially with amphotericin B, while for localized or less severe, non-meningeal infection, fluconazole is effective.

No role for steroid in the management of cryptoccocal meningitis

There is no place for corticosteroid in the management of HIV-associated cryptoccocal meningitis. In the most recent study of patients with AIDS-associated cryptococcal meningitis, adjunctive dexamethasone did not reduce mortality and was associated with more adverse events and disability and slower clearance of *Cryptococcus* from the spinal fluid [21].

Discontinuation of therapy

Therapeutic goal in the treatment of HIV-related cryptoccocal meningitis is to control acute infection, followed by life-long suppression of *C. neoformans* with low dose fluconazole. Most authorities recommend discontinuation of maintenance fluconazole monotherapy in HIV patients who remain symptom free of cryptococcal disease and have reconstituted their CD4 counts to more than 200 cells/μL for more than 6 months.

Treatment Outcomes

Untreated, cryptococcal meningitis is invariably fatal. If diagnosed rapidly, treated initially with amphotericin B plus flucytosine and with CSF pressure managed pro-actively, the 10-week mortality is probably around 20%, as reflected in results from studies from the USA and Europe (see table 1 below). In patients treated late, with fluconazole monotherapy, the outcome is much worse- >50% 10 week mortality, as reported below (table 2), in series from Africa. *C. gattii* infections also probably respond less well. In those with underlying disease that cannot be controlled (ie lymphoma or untreated HIV infection), the outcome is also poor.

A cohort study from Botswana recruiting 236 individuals with HIV-associated cryptococcal meningitis showed an overall mortality of 62%. The two-week, 10-week and 1-year mortality were 26%, 50%, and 65% respectively [22].

New treatment option

A recent study in Tanzania has shown that the antidepressant drug, sertraline in combination with fluconazole improve two-weeks CSF fungal clearance rate and clinical outcomes and is superior to fluconazole monotherapy or short course amphotericin B therapy [23].

Table 1: Outcomes of therapy for HIV-associated cryptococcal meningitis in developed country settings

Country	Reference	Year	Induction Treatment	2 week mortality	10 week mortality	Comments
USA	Saag, 1992 [24]	1988-1989	Amphotericin B 0.4-0.5mg/kg/d +/- Flucytosine 150mg/kg/d, or fluconazole 200-400 mg/d	12%	17%	*lethargy or obtundation Comatose patients and those unlikely to survive 2 weeks excluded.
USA	Van der Horst, 1997 [25]	1991-1994	Ampotericin B 0.7mg/kg/d +/- Flucytosine 100mg/kg/d	5.5%	10-23%*	Comatose patients excluded. *exact 10 week mortality unknown as only subset of patients re-randomized at 2 weeks
USA	Robinson, 1999 [26]	1986-1993	Amphotericin B 0.3-0.7mg/kg/d + Flucytosine 150mg/kg/day	12%	26%	
France	Lortholary 2006 [27]	1990-1996	Amphotericin based therapy 75%, fluconazole monotherapy 25%	-----	19%	No difference between pre-ART and ART era (21% vs 18%).
France	Dromer, 2008 [28]	1997-2001	Amphotericin B + 5FC 52%. Amphotericin B monotherapy or fluconazole montherapy in the remainder	6.5%*	15%**	*Just HIV positive patients. **12 week data.

Table 2: Outcomes of therapy for HIV-associated cryptococcal meningitis in resource-limited settings

Country	Reference	Year	Induction Treatment	2 week mortality	10 week mortality	Comments
Thailand	Imwidthaya, 2000 [29]	1996-1997	Amphotericin B 0.5-0.8mg/kg/d	-----	43%	

Thailand	Pitisuttothum, 2001[30]	1997-1999	Amphotericin B 0.7mg/kg/d	16%	~40%*	*Exact 10 week figure not reported.
Thailand	Brouwer, 2004 [31]	2002	Amphotericin B +/- Flucytosine 100mg/kg/d and/or fluconazole 400mg/d	14%	22%	
Cambodia	Micol, 2007 [32]	2004	Amphotericin B 0.7mg/kg/d	10%*	37%**	*3 and **12 week figures.
Peru	Dammert, 2008 [33]	1998-2001	Amphotericin B 0.7mg/kg/d	13%	19%	
Brazil	Pappalardo, 2007 [34]	1995-1997	Amphotericin B based therapy 80% Fluconazole plus flucytosine 20%*	31%	63%**	*Exact doses of treatment not specified. **deaths during "second phase" of treatment. Timing not specified.
Zambia	Mwaba , 2001 [35]	1998-1999	Fluconazole 200mg/d*	39%	96%**	*Data only reported for those who received treatment. 400mg stat dose initially. **12 week figure
Uganda	Mayanja-Kizza, 1998 [36]	1994	Fluconazole 200mg/d	40%	64%*	*8 week figure
			Fluconazole 200mg/d + 5FC	16%	44%*	
South Africa	Schaars, 2006 [37]	1999-2002	Fluconazole 200mg or 400mg/d	25%**	~50%***	**In hospital. *** median follow up for discharged patients 36 days, lost=censored
South Africa	Bicanic, 2007 [38]	2005	Amphotericin B 1mg/kg/d*	17%	37%	*Amphotericin only given for 1 week. 5 of 54 patients received fluconazole as initial therapy.
Uganda	Kambugu, 2008 [39]	2001-2002	Amphotericin B 0.7mg/kg/d	46%	-----	Comatose patients excluded.
		2006-2007		20%	43%	
South Africa	Bicanic, 2008 [40]	2005-2006	Amphotericin B 0.7-1mg/kg/d plus Flucytosine 100mg/kg/d	6%	24%	
Botswana	Bisson, 2008 [41]	2005-2006	Amphotericin B 1mg/kg/d	17%*	-----	*In hospital figure
Uganda	Longley, 2008 [42]	2005-2007	Fluconazole 800 mg/d	37%	60%	
			Fluconazole 1200mg/d	22%	48%	
Malawi	Jackson, 2012 [43]	2008	Fluconazole 1200mg/d	37%	58%	
			Fluconazole 1200mg/d plus 5FC 100mg/kg/d	10%	43%	

Opportunities to reduce Global Disease Burden and improve patient outcomes:

A number of feasible initiatives, if widely implemented, could have a very substantial impact on reducing the global cryptococcal disease burden:

1. Prevention of clinical disease in those newly diagnosed with HIV with a low CD4 cell count (<100) through screening for cryptococcal antigen prior to initiation of antiretroviral therapy, the "screen and treat" approach; and pre-emptive fluconazole therapy in those patients who test antigen positive (around 5% in many settings). Recent research has shown:

a. Over 50% cases in sub-Saharan Africa currently present **AFTER** a diagnosis of HIV has been made.

b. Antigen screening after an HIV diagnosis, before antiretroviral therapy, identifies those at risk of developing meningitis – 0/660 Ag negative patients developed meningitis, compared with 7/25 antigen positive patients [44].

c. The strategy is highly cost-effective.

The strategy has been endorsed for high incidence areas in WHO guidance, and is being implemented in South Africa and Rwanda.

2. Earlier diagnosis of all symptomatic cases through widespread use of new point-of-care antigen test in primary care as well as secondary care settings.

3. Optimization of antifungal therapy in resource limited settings through:
    a. Training and operational research on safe delivery of amphotericin B-based therapy (detailed advise on pre-emptive management of known toxicities is given in WHO guidelines).
    b. Increased access to flucytosine. Abandonment of fluconazole monotherapy and replacement with amphotericin B-based therapy (for one or two weeks depending on resource), or oral combination therapy with fluconazole and flucytosine, would likely substantially reduce the 10 week mortality from around 60% seen with fluconazole alone towards the 30-35% seen with amphotericin B-based combination treatment.
    c. Modelling suggests that having amphotericin B and flucytosine available across Africa will reduce HIV-related cryptococcal deaths by over 60,000 a year, and 300,000 over 5 years [45].
4. Systematic measurement and management of cerebrospinal fluid pressure through increased access to manometers in resource limited settings of high burden.

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