

East African Medical Journal Vol. 90 No. 12 (Supplement) December 2013

CRYPTOCOCCUS MENINGITIS IN A COHORT OF HIV POSITIVE KENYAN PATIENTS : OUTCOME AFTER TWO WEEKS OF THERAPY

A. E. O. Otedo, MBChB, MMed (Int.Med.); Cert. Gastroenterology, Consultant Physician/ Gastroenterologist, DDMS/ Senior Medical Specialist, Kisumu District Hospital, P.O. Box, 4685-40103, Kisumu, C. F. Otiemo, MBChB, MMed (Int. Med), Associate Professor, Consultant Physician, Department of Clinical Medicine and Therapeutics, College of Health Sciences, University of Nairobi, P.O. Box, 19676-00202, Nairobi, J. Jowi, MBChB, MMed (Int.Med), Associate Professor, Consultant Physician, Neurologist, Aga Khan Hospital, P. O. Box 30700-00100, Nairobi, G. O. Oyoo, MD, MMed (Int. Med.), FACR, Senior Lecturer, Consultant Physician and Rheumatologist and E. O. Omonge, MBChB, MMed, (Int. Med), Lecturer, Department of Clinical Medicine and Therapeutics, College of Health Sciences, University of Nairobi, P.O. Box, 19676-00202, Nairobi, Kenya

Request for reprints to: Dr. A.E.O. Otedo, Senior Medical Specialist, Kisumu District Hospital, P.O. Box, 4685-40103, Kisumu, Kenya, email: otedoamos@yahoo.com

**CRYPTOCOCCUS MENINGITIS IN A COHORT OF HIV POSITIVE KENYAN PATIENTS: OUTCOME AFTER TWO WEEKS OF THERAPY**

A. E. O. OTEDO, C. F. OTIENO, J. JOWI, G.O. OYOO and E. O. OMONGE

**ABSTRACT**

**Background:** Cryptococcus meningitis is the most lethal meningitis in patients with HIV/AIDS. It is invariably fatal if not treated appropriately and promptly. In sub-Saharan Africa with the highest prevalence of HIV/AIDS, response to treatment of cryptococcal meningitis has seldom been assessed.

**Objective:** To describe the clinical features, laboratory findings, CD4+ cell counts and clinical outcome after a two-week treatment course of patients having cryptococcal meningitis.

**Design:** Longitudinal, prospective, consecutive entry study.

**Setting:** Kisumu District Hospital, Nairobi Rheumatology Clinic and Mater hospital between July 2001 and May 2007.

**Subjects:** One hundred and forty one patients with cryptococcus meningitis.

**Main outcome measures:** CD4+ cell count, cerebrospinal fluid (CSF) biochemistry/ microbiology, morbidity and mortality.

**Results:** One hundred and forty one patients (80 males and 61 females) with cryptococcus meningitis were included. Mean age and CD4+ cell counts was  $36.12 \pm 9.1$  years (15-75) and  $66.9 \pm 102.8$  cells/ $\mu$ l (1-1058) respectively. One hundred and forty one (83%) patients had CD4 + cell counts  $<100$  cells/ $\mu$ l implying severe immunosuppression. Two (1.4%) patients had CD4+ cells  $> 350$  cells/ $\mu$ l and 22 (15.6%) patients had CD4+ cell counts between 100-350 cells/ $\mu$ l. Ten (six males and four females) died within one week (four amphotericin B, three fluconazole, three no treatment). Eighty one patients were simultaneously initiated on HAART.

**Conclusion:** Cryptococcal meningitis has a good clinical outcome when promptly and appropriately managed despite the low CD4+ cell count. Measures to avail amphotericin B and fluconazole at the mid level healthcare facilities must be enhanced.

**INTRODUCTION**

Cryptococcal meningitis is caused by the environmental encapsulated fungus, cryptococcus neoformans. It is a systemic AIDS defining illness and the most common, lethal fungal meningitis in patients with human immunodeficiency virus/ acquired immunodeficiency syndrome (HIV/ AIDS) worldwide. It is common in patients with severe immunosuppression and is almost invariably fatal if not treated appropriately and promptly (1-7).

Approximately 7-10% of those infected with human immunodeficiency virus/acquired immunodeficiency syndrome (HIV/ AIDS) will develop cryptococcal meningitis during the course of their illness (8-11). It is more prevalent in patients with CD4+ cell counts lower than 100 cells/ $\mu$ l and suffice it to note that cryptococcal meningitis is almost always fatal if not treated promptly and appropriately (12). Cryptococcus is initially occult in 50% of these patients or patients with higher CD4+ cell counts  $>300$  cells/ $\mu$ l (9-11). Poor prognostic indicators in patients

with cryptococcal meningitis include positive Indian ink test, high cerebrospinal fluid (C.S.F) cryptococcal antigen titers, very low C.S.F white cell count (< 20 white cells/ml), low CSF glucose concentration (< 2 mmol/L) and high cerebrospinal fluid (CSF) opening pressures (> 60 cm H<sub>2</sub>O) (1, 6, 13). A study done in Zimbabwe showed that impaired mental status (GCS < 13) and hyponatraemia (Na<sup>+</sup> < 130 mmol/L) were associated with rapid clinical deterioration (1).

It frequently takes more than two weeks for drug treatment of cryptococcus meningitis to sterilise cerebrospinal fluid (C.S.F), achieve clinical improvement and reduce morbidity and mortality. Current options for treating cryptococcal meningitis include amphotericin B alone, amphotericin B plus flucytosine, or fluconazole alone or fluconazole after an initial one to two weeks administration of amphotericin B. None of these treatments is entirely satisfactory due to low rates of C.S.F sterilisation with low dose of amphotericin B, 0.4 mg/kg/day (9, 10, 14). This dose is associated with a success rate of 40% and mortality rate of 8% at 22 weeks of treatment (9,10,14). Intravenous amphotericin B with or without flucytosine is usually the standard therapy for cryptococcal meningitis in patients with acquired immunodeficiency syndrome (AIDS) (14). Ministry of health does not always have amphotericin B in its kit but occasionally supplies fluconazole.

A standard two week induction course of amphotericin B deoxycholate (0.7 mg/kg/day) and flucytosine 100 mg/kg/day is recommended (12,16).

In sub-Saharan Africa few health facilities use amphotericin B and ministry of health erratically supplies fluconazole. Prompt and appropriate treatment is usually associated with good clinical outcome and reduced mortality. Longitudinal studies evaluating the treatment outcomes of patients with cryptococcal meningitis from sub-Saharan Africa are scanty yet, it is here where there is the predominant global burden of HIV infection.

This study was undertaken to define the clinical presentation, CD4<sup>+</sup> cell count, mode of treatment and clinical outcome after two weeks of treatment.

## MATERIALS AND METHODS

Two hundred and fifty one patients (145 males and 106 females) who had meningitis were examined and screened. One hundred and ten (65 males and 45 females) patients were excluded (Indian ink negative)

and 141 (80 males and 61 females) patients were included in the study.

Informed signed consent was obtained from each patient, (<18 years old or in coma, signed by parent/guardian). A standard questionnaire was run for each study patient and included; biodata, symptoms and signs at presentation. Each study patient underwent a thorough clinical examination by one of the authors. Diagnostic counselling and testing (DCT) (pre- and post- test counselling) was done and sustained for each patient. Under aseptic technique, blood was taken for complete blood count (CBC), CD4<sup>+</sup> cell count and ELISA test for HIV. A lumbar puncture for cerebrospinal fluid (C.S.F) was also performed in every subject after doing a funduscopy. CSF was subjected to microscopy, gram stain, biochemistry, acid alcohol fast bacilli (Aafb) smear (Ziehl-Nielsen staining) and Indian ink staining. C.S.F cryptococcal antigen titer was done for five patients who were already on HAART and had severe headaches and negative Indian ink test.

Only 141 patients (80 males and 61 females) with positive Indian ink test, 136 and positive cryptococcal antigen test 5, in C.S.F were enrolled for this study. This strict inclusion/exclusion criterion could have excluded patients with less florid cryptococcal meningitis. Chest X-ray was done at admission (if indicated). HIV serology was confirmed using an ELISA test (welcozyme) with a sensitivity and specificity of 99.7 and 99.9% respectively. Sera testing positive were subjected to ELISA test for confirmation of the HIV status. CD4<sup>+</sup> cell count was analysed using FACS (fluorescent activated cell sorter) flow cytometry machine with a sensitivity of 1-2000 cells/ $\mu$ l (FACS count machine from Baxton, Dickson).

*Ethical Approval:* The ethics and standards committee Kisumu District Hospital approved the study.

*Intervention:* Patients diagnosed to have cryptococcal meningitis were admitted in the medical wards and started on amphotericin B 50mg daily for two weeks. Five patients were initiated on intravenous fluconazole due to deranged renal functions and could therefore not be given amphotericin B. Those who could not afford were put on fluconazole (oral or intravenous) 400mg daily for two weeks and thereafter, fluconazole 200mg daily (patients buy, its erratically supplied by the hospital). Patients were reviewed daily in the medical wards till discharge, which was between 10–31 days depending on their clinical response and

co-morbid state. No post-mortem was performed for those who died while on treatment. Convulsions were managed using phenytoin sodium or carbamazepine. Some patients did not receive anti-fungal treatment because fluconazole was out of stock (when they were admitted) and they could not afford to buy fluconazole or amphotericin B. They only received palliative care.

Eighty one patients could afford to buy HAART and were initiated on them. The study was conducted when the National Ministry of Health HAART access programme was not in place.

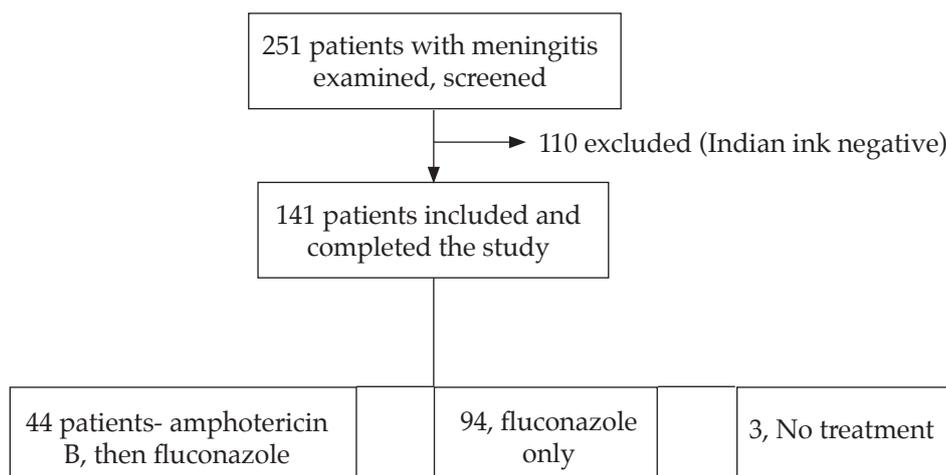
*Statistical analysis:* The data were cleaned and analysed using SPSS (version 11). They were expressed as mean  $\pm$  SD, tables and bar charts.

## RESULTS

One hundred and forty one patients (80 males and 61 females) were included in the study conducted between July 2001 and May 2007. All the patients tested positive for HIV and cryptococcus meningitis (136 Indian ink positive, 5, C.S.F cryptococcal antigen test positive).

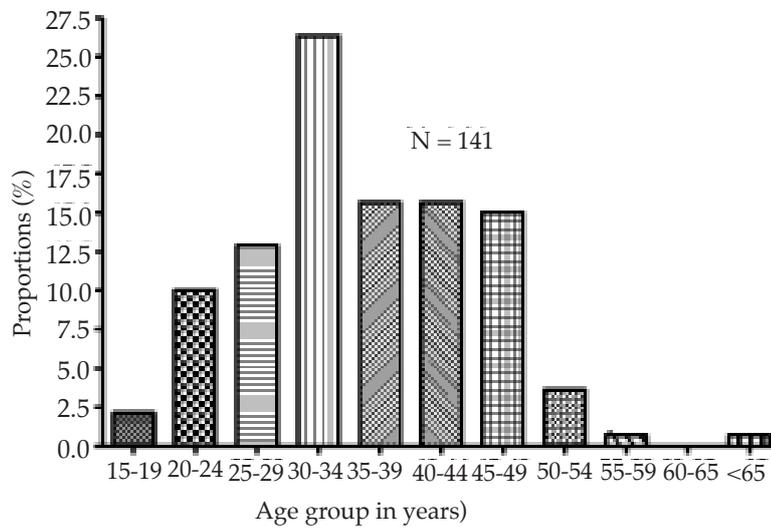
The mean age range was  $36.12 \pm 9.1$  (15-75) years. The mean CD4+ cell count was  $66.9 \pm 102.8$  (1-1058) cells/ $\mu$ l. 2 (1.4%) patients had CD4+ cell counts  $> 350$  cells/ $\mu$ l, 22 (15.6%) patients had CD4+ cell counts of 100-350 and 117 (83%) patients had CD4+ cell counts  $< 100$  cells/ $\mu$ l. Ten patients died within one week (three no treatment, three fluconazole, and two amphotericin B).

**Figure 1**  
*Enrolment profile*



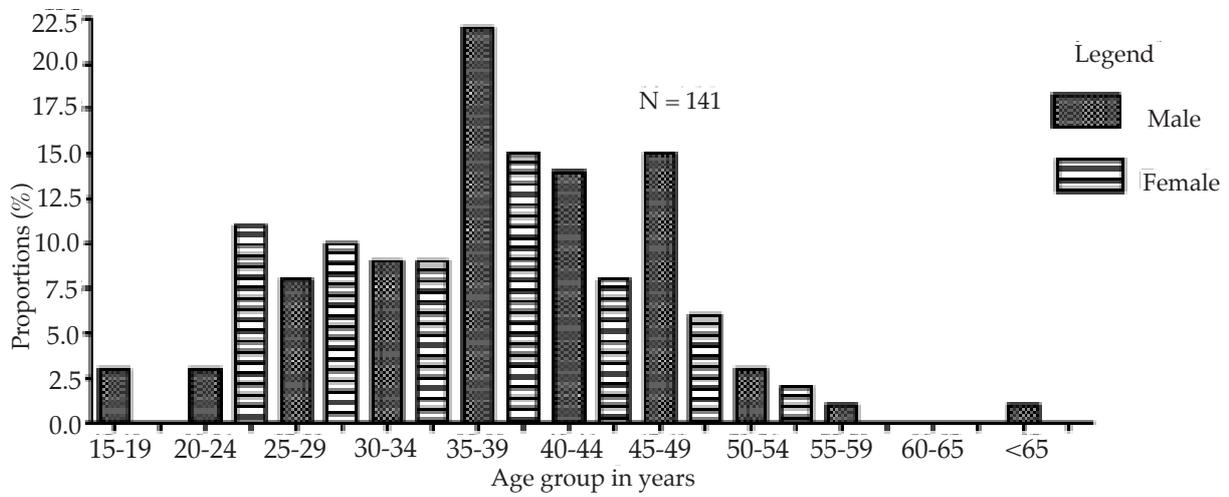
**Figure 2**

*Age distribution of HIV positive patients who had cryptococcal meningitis*



**Figure 3**

*Age and gender distribution of HIV patients who had cryptococcal meningitis*



**Table 1***Presenting symptoms of 141 patients with HIV/AIDS and Cryptococcal meningitis.*

Symptom	No. (Percentage)
Headache	141 (100)
Neck stiffness	97 (68.8)
Confusion /coma	47(33.3)
Fits (seizure)	92 (65.2)
Fever	70(49.6)
Neurological deficit	10 (7.0)
Blurred vision	27 (19.1)

**Table 2***Presenting clinical signs of 141 patients with HIV/AIDS and cryptococcal meningitis*

Sign	No (Percentage)
Fever	70 (49.6)
Meningeal signs (stiff neck, Kernig's sign, Brudzinski's sign)	97 (68.8)
Motor weakness	10 (7.0)
Papilloedema	17 (12.0)
Confusion/coma	47 (33.3)
Cranial nerve palsy	9(6.4)

**Table 3***Baseline characteristics**Demographic and laboratory profile of the 141 patients with HIV/AIDS and Cryptococcus meningitis.*

Parameter	Value
M:F ratio	80:61
Age (years)	36.12 ± 9.1 (15-75)
Mean CD4+ cell count (350-1600 cells/ $\mu$ l)	66.9 ± 102.8 (1-1058)
CD4+ cell counts (cells/ $\mu$ l)	
> 500	2 (1.4% )
350-499	4 (2.8% )
200-349	14 (10.0% )
< 200	121 (85.8%)
*CDC clinical staging	
A	3 (2%)
B	2 (1.4%)
C	136 (96.4%)
AST (5-40 IU/L)	49.7 ± 17.3 (23-97)
ALT (5-37 IU/L)	37.3 ± 14.2 (21-94)
Creatinine(60-120 $\mu$ mol/L)	43.9 ± 19.9 (61.3-117.4)
Mean CSF proteins(15-45 mg/L)	166.5 ± 77.3 (311-2570)
Mean CSF glucose (0.83-2.03 mmol/L)	1.9 ± 0.7 (0.1-3.9)
Mean CSF lymphocytes (cells/ $\mu$ l)	6 (0-19)
Drugs used	Amphotericin B-44 patients, Fluconazole 94 patients, No treatment-3 patients.

## DISCUSSION

This study included 141 patients (80 males and 61 females) with cryptococcal meningitis, 126 of whom were highly active anti-retroviral therapy (HAART) naïve and 15 were already initiated on HAART (5-12 months). The study was conducted when HAART and fluconazole were not yet freely available and accessible at the public hospitals for all patients (supply was erratic, amphotericin B was out of supply). The mean age was  $36.12 \pm 9.1$  years (15-70). The clinical symptoms and signs all indicated meningitis. Cerebrospinal fluid from five patients was Indian ink negative and cryptococcal antigen titer test positive. Cryptococcal meningitis is the most common neurological opportunistic infection in HIV/AIDS and almost invariably fatal if poorly managed (21, 22).

Research in Thailand showed that success rates in patients treated with 0.4 mg/kg/day of amphotericin B was 40%, with a mortality rate of 8% at two weeks (8-10,14). Indeed other studies show that, despite the acute mortality rates during initial therapy, the 12 month survival rates among all patients on maintenance therapy with fluconazole 200 mg daily and addition of HAART are significantly improved (15,19,20).

At week two, 40 patients on amphotericin B and 81 patients on fluconazole were all discharged home. Four patients on amphotericin B and six patients on fluconazole stayed longer in the hospital for about three weeks (six had tuberculosis, four had global motor weakness and were undergoing physical therapy). Ten (8.8%) died within the first week of treatment. Three out of ten were not on anti-fungal treatment (Fluconazole was out of stock in the hospital and they could not afford to purchase), seven out of ten received anti-fungal treatment (3 amphotericin B and 4 –intravenous fluconazole). A study done in Zambia showed that patients with cryptococcal meningitis who did not receive anti-fungal treatment were all dead at week seven of follow up (15).

Only 30 of the discharged patients were followed up subsequently. The other 111 patients were not followed up for reasons:

- (i) Being transferred to other health facilities, which had free fluconazole and HAART like Kenyatta National Hospital and New Nyanza General Hospital Kisumu according to their request.
- (ii) Logistics and could not afford transport back to the hospital. The follow up of the cohort was important since it was noted in Zambia that at

week 24, most of the patients had died but the cause of mortality was not reported (15).

It has been demonstrated that combination therapy of amphotericin B and flucytosine was associated with a faster clearance of cryptococcus from C.S.F. and a better clinical outcome (16). In public hospitals, amphotericin B is not regularly supplied and patients had to self-purchase. The administration of the drug requires hospitalization.

Poor prognostic indicators associated with increased mortality in cryptococcus meningitis have been identified as low CD4+ cell count with a high visible cryptococcal count at admission of  $> 900 \times 10^3$ /ml, Glasgow coma scale (GCS) < 13, low body weight on admission < 40 kg, higher systolic blood pressure >150 mm Hg on admission, a higher BUN, higher CSF protein > 45g/L and higher CSF opening pressure >60 cmH<sub>2</sub>O (8,17, 18).

The mean CD4+ cell counts of the patients were very low  $66.9 \pm 102.8$  cells/ $\mu$ l (range of 1-1058). This signifies severe immunosuppression and has been associated with a large yeast biomass in the sub-arachnoid space. This requires prompt, appropriate, anti-fungal treatment (12, 19). Twenty one (14.9%) patients had co-morbidities (hepatitis B virus 4, cholecystitis 3 and pulmonary tuberculosis 14). The other 120 (85.1%) patients had cryptococcal meningitis alone. Indeed the ten patients who died had very low CD4+ cell counts of < 50 cells/ $\mu$ l (1-49). Forty seven (33.3%) patients presented with confusion/coma with a G.C.S (glass coma scale) between 5-8. These are patients who require intravenous medication, which are not freely and readily available in public health facilities and the Centre for Diseases Control (CDC) supplies oral fluconazole to few government hospitals with comprehensive care clinics. This may increase mortality due to cryptococcal meningitis due to the low bioavailability of oral fluconazole.

Forty seven (33.3%) patients in this cohort had confusion/coma. A study done in Tanzania showed that low CD4+ cell counts plus coma predicts cryptococcal meningitis. Thus, cryptococcal meningitis is common among Tanzanian HIV positive in-patients presenting with headache and/or altered mental status (23).

The clinical diagnosis of cryptococcal meningitis requires high index of suspicion and the laboratory back up must be well equipped to isolate the fungus and or the antigen. The hospitals should therefore have adequate and appropriate anti-fungal drugs for immediate initiation of treatment to reduce mortality due to Cryptococcal meningitis. The review by Jowi

and a study in Zambia also underscore the necessity of early diagnosis, prompt appropriate treatment and HAART initiation in managing cryptococcus meningitis (15, 22).

This requires scaling up and adequate supply of amphotericin B and fluconazole in the comprehensive care clinics where the majority of patients are managed. With the inadequate state of investigation capability of the HIV care clinics, it would be necessary to carry out a study where high dose intravenous or oral fluconazole can be used to treat cryptococcal meningitis instead of the nephrotoxic amphotericin B in resource constrained settings or in HIV infected patients who already have compromised renal functions.

### REFERENCES

- Hyderman R.S., Gangaidzo I.T., Hakim J.G, *et al.* Cryptococcosis meningitis in human immunodeficiency virus infected patients in Harare, Zimbabwe. *Clin. Infect. Dis.* 1998; **26**: 284 – 289.
- Mayanje – Kizza H., Oishi K., Mitarai S, *et al.* Combination therapy with fluconazole and flucytosine for cryptococcal meningitis in Ugandan patients with AIDS. *Clin. Infect. Dis.* 1998; **26**: 1362 – 1366.
- Moosa M.Y.S., Coovadia Y.M. Cryptococcal meningitis in Durban, South Africa: A comparison of clinical features, laboratory findings and outcome for human immunodeficiency virus (HIV) – Positive and HIV negative patients. *Clin. Infect. Dis.* 1997; **24**: 131 – 134.
- Powderly W.G. Recent advances in the management of cryptococcal meningitis in patients with AIDS. *Clin. Infect. Dis.* 1996; **22** (suppl.2): 119 – 123.
- Sharkey P.K., Graybill J.R, Johnson E.S., *et al.* Amphotericin B lipid complex compared with Amphotericin B in the treatment of cryptococcal meningitis in patients with AIDS. *Clin. Infect. Dis.* 1996; **22**: 315 – 21.
- Van der Horst C.M., Saag M.S., Cloud G.A. *et al.* Treatment of cryptococcal meningitis associated with the acquired immunodeficiency syndrome. *N. Engl. J. Med.* 1997; **337**:15-21.
- Khanna N., Chandramuki A., Desai A., *et al.* Cryptococcal infections of the central nervous system: An analysis of predisposing factors, laboratory findings and outcome in patients from South India with special reference to HIV infection. *J. Med. Microbiol.* 1996; **45**:376-379.
- A.P. Pitisuttithum, S. Tansuphasawadikul, Simpson A. J. H., Howe P. A. and white N. J. A prospective study of AIDS – associated cryptococcal meningitis in Thailand treated with high dose Amphotericin B. *J. Infect.* 2001; **43**: 226-233.
- Zuger A., Lovie E., Holzman R.S., Simberkoff M.S., and Rahal J.J. Cryptococcal disease in patients with the acquired Immunodeficiency Syndrome. Diagnostic features and outcome of treatment. *Ann. Intern. Med.* 1986; **104**: 234-240.
- Kovacs J.A., Kovacs A.A., Polis M. *et al.* Cryptococcosis in the acquired immunodeficiency Syndrome. *Ann. Intern. Med.* 1985; **103**: 533-538.
- Chuck S. L. and Sande M. A. Infections with cryptococcosis Neoformans in the acquired Immunodeficiency Syndrome. *N. Engl. J. Med.* 1989; **321**: 794 – 799.
- Gupta S. K. and Sarosi G. A. Cryptococcol meningitis. *Curr. Treat. Opt. Infect. Dis.* 2002; **4**: 503 – 511.
- Diamond R. D, and Bennett J. E. Prognostic factors in cryptococcal meningitis. A study in 111 cases. *Ann. Intern. Med.* 1974; **80**: 176 – 181.
- Saag M.S., Robinson P., Grieco M.H., Sharkey P.K., Thomson S.E., Sugar A.M., Tuazon C.U., Fisher J.F., Powderly W. G. and Cloud G. A. *et al.* Comparison of amphotericin B with fluconazole in the treatment of acute AIDS – associated cryptococcal meningitis. The NIAID mycoses study group and AIDS clinical trials group. *N. Engl. J. Med.* 1992; **326**:83 –89.
- Mwaba P., Mwansa J., Chintu C., Pobe J., Scarborough M., Portsmouth S., and Zumla A. Clinical presentation, natural history, and cumulative death rates of 230 adults with primary cryptococcal meningitis in Zambian AIDS patients treated under local conditions. *Post. Grad. Med. J. Engl.* 2001; (77); **914**: 769 – 773.
- Brouwer A.E., Rajanuwang A. Chierakul W., Griffin G.E., Larsen A.R., White N.J. and Harrison T.S. Combination antifungal therapies for HIV associated cryptococcal meningitis: a randomised trial. *Lancet.* 2004; **363**:1764 – 6177.
- Clark R.A., Greer D., Atkinson W., Valaines T.G. and Hyslop N. Spectrum of cryptococcus Neoformans infection in 68 patients infected with human immunodeficiency virus. *Rev. Infect. Dis.* 1990; (12): 768 – 777.
- Wuthiekanun V., Howe P.A., Simpson A.J. *et al.* Antifungal susceptibilities of cryptococcosis neoformans in North East Thailand. *Trans. Roy. Soc. Trop. Med. Hyg.* 1999; (93): 96-97.
- Leenders A.C., Reiss P, *et al.* Liposomal Amphotericin B (AmBisome) compared with Amphotericin B, both followed by oral fluconazole in the treatment of AIDS associated cryptococcal meningitis. *AIDS* 1997; **11**: 1463 – 1471.
- Aberg J.A., Powderly W.G. Cryptococcal disease. Implications of recent clinical trials on treatment and management. *AIDS. Clin. Rev.* 1997; **98**: 229 – 248.
- French N., Gray K., Watera C. *et al.* Cryptococcal infection in a cohort of HIV -1 infected Ugandan Adults. *AIDS* 2002; **16**: 1031 – 1038.
- Jowi J.O. Neurological manifestations of HIV Infection and AIDS. A review. Ed. Prof. William Lore; 2005. Chap 3 page 10.
- Kisenge P.R., Alexander T.H., Venance P.M., Mchele J.P.D., Ndealilia S.S., Andreas M. and Houpt E.R. Low CD4 counts plus coma predicts cryptococcal meningitis in Tanzania. *BMC infectious Diseases* 2007; **7**: 39.