College of Medicine and Health Sciences

School of Medicine and Pharmacy

THERAPEUTIC ASPECTS OF CRYPTOCOCCAL MENINGITIS IN HIV PATIENTS AND FACTORS CONTRIBUTING TO MORTALITY. CASE OF CHUK.

Dissertation submitted for fulfillment of the requirements for the award of the degree of Masters of medicine in Internal Medicine, School of Medicine and Pharmacy, College of Medicine and Health Sciences at University of Rwanda

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DECLARATION

I, Dr. UWIMANA Marie Grace, to the best of my knowledge hereby declare and certify that the work presented in this dissertation entitled "**Therapeutic aspect of cryptococcal meningitis in HIV patients and factors contributing to mortality. Case of CHUK"** is entirely my own and original work and it has never been presented or submitted in whole or in part to any other university and was checked for plagiarism using Turnitin

UWIMANA Marie Grace Date……. Signature

Supervisors:

We, hereby declare that this dissertation has been submitted with our approval as the supervisors.

Dr RUTAGANDA Eric Date …… Signature

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To you my husband Dr MUTABAZI EMMANUEL, your love and support transformed my dream into reality.

My thanks are finally presented to all my colleagues, friends and relatives for their endurance and charity throughout my life, I can say "May our Almighty God bless you ".

Dr. UWIMANA Marie Grace

DEDICATION

This work is dedicated;

To you mother, for your love, hardworking and patience.

To my husband MUTABAZI Emmanuel for your great love and support.

To My daughters IRADUKUNDA Ange Henriette and ISHIMWE Louange Ghislaine and to my son SHEMA Jules Fidel.

TO you colleagues, brothers and sisters for your encouragements.

ABSTRACT

Introduction: Cryptococcal meningitis remains a major cause of HIV-related morbidity and mortality Worldwide with a greatest burden in LMIC. The higher mortality is related to delayed diagnosis of both HIV and CM, substandard treatment of CM with failure to manage high ICP and amphotericin B toxicity.

Objective: This study aimed to assess therapeutic aspects of CM in HIV patients and identify factors contributing to mortality.

Methods: We conducted a prospective observational study in patients with HIV related CM at CHUK from 1st February 2019 to 31st January2020. Data included demographic, clinical features, treatment and outcome. Patient's outcome was recorded at 2 weeks and 10 weeks of treatment.

Results: We identified 41 patients, 27 male and 14 females sex ratio (M: F) of 1.93:1, 18 to 69 years with a mean age of 40 years. Therapeutic LP was performed in 44.9% and CSF opening pressure was measured in 4.9% of cases. In consolidation, 57.1% showed a good adherence to fluconazole.CM treatment protocol was fully respected in 36.6%, partially respected in 48.8% and not respected in 14.6% of cases with a mortality of 29.1% and 58.5% at 2 weeks and 10 weeks respectively. Failure to perform therapeutic LP, non-respect of CM treatment protocol and poor adherence to fluconazole in consolidation phase were factors associated with mortality. ($p<0.05$)

Conclusion: CM remains a life-threatening condition in HIV patients in CHUK with a significant mortality. Efforts should be made regarding management of CM.

Key word: Therapeutic Aspect,CM,HIV, CHUK.

LIST OF ABBREVIATIONS

AIDS: Acquired immunodeficiency syndrome

ART: Antiretroviral therapy

CHUK: Centre Hospitalier Universitaire de Kigali (Kigali University Teaching Hospital)

CmH_2O: Centimeter of water

CNS: Central nervous system.

CD4: Cluster of differentiation 4

CFU: Colony formulating unit

CT: Computer tomography

CM: Cryptococcal meningitis

C-IRIS: Cryptoccocal-immune reconstitution inflammatory syndrome

CSF: Cerebral spinal fluid

CHBI: Community Health based insurance

CMHS: College of medicine and health sciences

HIV: Human immunodeficiency virus

ICP: Intracranial pressure

IRB: Institutional review board

IV: Intravenous

K^+: Potassium

KCL: Potassium chloride

LP: Lumbar puncture

MRI: Magnetic resonance imaging

MLIC: Middle- and Low-Income Countries

NS : Normal saline

Meq : Mille-équivalent

RNA: Ribonucleic acid

UR: University of Rwanda

WHO: World Health Organization

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I. INTRODUCTION

I.1. Background

Cryptococcal meningitis is a major cause of mortality in HIV positive patients worldwide mainly in MLIC where the incidence of HIV infection is relatively elevated.[1]

The global annual incidence of CM accounts for 223100 cases with 73% coming from Sub-Saharan Africa. In addition, its fatality rate is estimated at 35to 65% among HIV infected African and 14 to 26% in industrialized countries. CM results into serious morbidities such as irreversible loss of vision, deafness and other irreversible neurocognitive impairments.[5, 10, 17]

Many researches and international guidelines regarding diagnostic and management have been elaborated during the past three decades of HIV pandemic in order to improve the long-term survival of HIV patients. Even with the increase availability of potent ART in Sub Saharan Africa, the rate of CM remains stabilized at relatively high incidence estimated at 3% per year among HIV infected adults.[1, 10]

I .2.Problem statement

The mortality attributed to HIV related CM remains remarkably elevated in LMIC. This higher mortality is related to delayed diagnosis of both HIV and CM, below standard treatment of CM mainly failure to manage high ICP, amphotericin B toxicity and lack of fist line antifungal therapy[1, 5, 10]

In Rwanda, there is a lack of data on management profile of CM and its contribution on overall mortality in HIV patients. We conducted a study intended to assess therapeutic aspects of CM in HIV positive patients, identify different factors contributing to mortality in this population in order to contribute on an improved management and outcome of HIV positive patients with CM.

I.3. Research questions.

What is the association between patients' profile, management and outcome of cryptococcal meningitis in HIV patients at CHUK?

I.4. Objectives

I.4.1 General Objective

To contribute in management and improve outcome of HIV patients with CM at CHUK.

I.4.2. Specific objectives

1. To know patients' characteristics (demographic, CD4 count, duration of HIV) from February 2019 to January2020.

2. To assess the management of HIV patients with CM at CHUK (regimen used, raised ICP and amphotericin B toxicity management).

3. To demonstrate the association between patients' profile, management and outcome of CM in HIV patients at CHUK from February 2019 to January 2020.

II. LITERATURE REVIEW

II.1.Introduction

CM is an infectious disease affecting meninges (inner layers of skull and spinal cord) caused by the encapsulated yeast "Cryptococcus".[7, 10, 16] More than 30 species of Cryptococcus are found in environment and two species are responsible for human disease:

Cryptococcus var Neoformans: Commonly found in immunosuppressed individuals worldwide and spread from the soil contaminated with bird particularly pigeon excreta.

Cryptococcus Gatti: Associated with numerous species of eucalyptus trees and responsible of infection mainly in immunocompetent individuals in tropical and subtropical regions. [7, 10, 16]

CM is an AIDS defining illness seen in individuals with CD4 less than100cells/mm^3.Cryptococcus infection is acquired through inhalation of the yeasts and remains latent in the respiratory system for months to years[10].

II.2. Epidemiology

Among 31.8 million who were living with HIV Worldwide in 2013, 21.1 million were living in Sub-Saharan Africa. 4.3 million of them were having CD4 counts less than 100 cells per μL globally and was at high risk of opportunist infection including CM. The Global annual incidence of CM was estimate at 223.100 worldwide with 73% arising from Sub-Saharan Africa [5].

The Global annual mortality of CM was estimated118.100 cases with 135900 cases occurring in Sub-Saharan Africa[5].

II.3. Clinical manifestation

The signs and symptoms of cryptococcal meningitis are not specific. The majority of patients present with acute to sub- acute onset of one or more in the following symptoms: Headache 93-96%, Seizure 33%, Fever 66%, Vomiting 77%, Blurred Vision 38%, Neck Stiffness 69%, Altered Mental Status 38%.[10]

Less frequently, patients may present with gait and cognitive impairment due to obstructive hydrocephalus .[10]

II.4. Diagnosis

The diagnosis of CM is achieved by CSF analysis and CSF opening pressure measurement remarkably elevated above200mmH_20 at 70% of patients .[1, 3, 10]

Different modalities are used for diagnosis of CM in CSF including:

India ink staining: Its sensitivity is estimated at 50–70% especially in case of yeast burdens higher than 103 CFU/mL.[1, 2, 3, 6]

Fungal culture of CSF samples: This is the gold standard diagnostic test. It becomes positive within 48 to72 hours for patient who never been exposed to antifungal therapy and up to 4weeks for those who are on treatment. Its sensitivity estimated at 75to 90% .[16]

Detection of cryptococcal antigen (CrAg) in CSF, its sensitivity and specificity are 93–100% and 93–98% respectively.[16]

II.5.Management

The key elements in management of CM include three phases of antifungal therapy, detection and management of raised ICP, management of amphotericin B based regimen toxicities .[2]

II.5.1. Antifungal therapy.

Antifungal therapy is given in three phases: induction, consolidation, and maintenance.[1, 2,3,6,16,16]

II.5.1.1.Induction phase

Induction therapy aimed to achieve rapid sterilization of CSF. Rwanda National protocol follows 2016 WHO guidelines, which recommends two weeks of induction with IV Amphotericin B (0.7 to 1.0 mg/kg /day) plus flucytosine (100mg/kg/day divided in four doses) as the first line therapy. Unfortunately, the availability of flucytosine is limited in sub-Saharan Africa, it is replaced by oral fluconazole 800mg/day as alternative[1 3,6,8,10,18]

The combination of Amphotericin B with a high dose of fluconazole (1,200 mg/day) given in five to seven day is an option when 14 days of amphotericin is not feasible (i.e., in case of amphotericin B toxicities) [6,7.] In absence of Amphotericin B and flucytosine, a high dose(1200mg/day) of fluconazole monotherapy is used for 10-12 weeks [1, 2, 3]

Liposomal amphotericin B (3to 4mg/kg/day) is preferable over amphotericin B deoxycholate because of its safety. Nevertheless, its access remains extremely limited in LMIC because of its high cost.[6, 8, 1 0]

The combination of Antidepressant drug sertraline with antifungal therapy have been proven to improve the rate of fungal clearance in CSF within two weeks .[3, 8, 9]

Corticosteroids are not used routinely in induction phase for the only reason that it decreases fungal clearance in CSF and associated with adverse events (infection, renal, cardiac and gastrointestinal disorders).[3, 8, 11]

II.5.1.2.Consolidation phase

Consolidation phase starts after 2 weeks of induction therapy and consists of fluconazole 400mg to 800mg per day for eight weeks. It can also be individualized based on response to antifungal therapy during induction phase. A high dose of fluconazole (1200mgper day) is used in consolidation phase when CSF sterility has not been achieved [1, 2, 3, 6, 8, 18]

II.5.1.3 Maintenance phase

After completion of two phases of therapy, patients with fungal culture negative start on fluconazole 200 mg/day as maintenance. [1, 2, 3, 8.18] .Discontinuation of maintenance therapy is based on patient's immunity and can be considered when patient has an undetectable HIV RNA levels for more than three months with CD4 counts ≥ 100 cells/μL.[3, 7,8,18]

In absence of HIV viral load testing, maintenance phase should be continued for one year and stopped if CD4 counts are >200 cells/μL. Fluconazole should be reinstituted in case of immunologic failure, ART stoppage, or a decrease in CD4 counts less than 100cell/μL[3, 8, 18]

II.5.2.Prevention, monitoring and management of Amphotericin B toxicities

Amphotericin B deoxycholate toxicities are barriers to optimal induction treatment in LMIC. The Major side effects of Amphotericin B include kidneys dysfunction, hypokalemia, hypomagnesaemia, anaemia, febrile reactions and phlebitis. [18]

II.5.2.1. Prevention of amphotericin B toxicity

The patient should receive one liter of NS mixed with 20 mmol of KCL given over two hours prior to Amphotericin B infusion and then maintenance with oral potassium supplement twice per day and oral magnesium once per day. [3, 16, 18]

It is important to flash the lines with NS and remove the bag from IV administrative set to minimize the risk of phlebitis. The symptoms of an infusion reaction can be minimized or prevented by premedication with acetaminophen (usual adult dose of 650 to 1000 mg orally) and/or diphenhydramine (usual adult dose, 25 to 50 mg orally or intravenously) if symptoms are severe IV hydrocortisone 25mg should be given. [3, 16]

II.5.2.2 .Monitoring of amphotericin B toxicity

Potassium and creatinine are checked at baseline and two to three times per week (mainly in the 2nd week of amphotericin B administration) and hemoglobin should be checked at baseline and weekly. [3,7,8,16,18]

II.5.2.3.Management of amphotericin B toxicity

When hypokalemia is significantly low (K <3.3 mol/l), The supplementation of potassium is increased to 40 Meq IV and/or one (8-mEq KCl) to two tablets oral potassium three times daily.[1, 8, 16, 18]

When creatinine increases by twofold from the baseline, increase pre-hydration to 1L every 8hours and consider temporal omission of amphotericin B doses. Once creatinine improves, resume amphotericin B at 0.7 mg/kg/day in alternate-day and monitor creatinine. If creatinine continues to rise, amphotericin B is stopped and replaced by fluconazole at 1200 mg/day, especially if seven doses of amphotericin B have been received.[1, 3, 16, 18]

Transfusion should be considered for severe amphotericin B–related anaemia, may also be a reason to stop amphotericin B prematurely and replaced by fluconazole 1200mg daily.[16, 18]

II.5.3. Management of raised intracranial pressure

Raised ICP is defined as CSF opening pressure ≥ 25 cm H_2O and is a common complication of CM. Elevated ICP is mainly due to a failure of CSF resorption via the arachnoid villa caused by physical obstruction of cryptococcal polysaccharide capsule. [3, 18]

Elevated ICP is characterized by headaches, vomiting, papilledema, reduced visual acuity, blindness, cranial nerve palsy's (most commonly cranial nerve VI), confusion, altered mental status, and coma. It can be also asymptomatic.[3, 18]

HIV patient with suspected CM should have an initial LP and measure baseline CSF opening pressure. LP should be repeated early and more than once with measurement of CSF opening pressure in order to assess for high ICP regardless of the presence of symptoms or signs. Persistence or recurrence of symptoms or signs of raised ICP determine the frequency of repeat therapeutic LP. It can be repeated daily until the symptoms resolve or CSF opening pressure is normalized (<20cmH_2o) for at least two days. [3, 8, 13, 18]

The data on the maximum volume of CSF that can be safely drained at one lumbar puncture are still lacking. Usually, 20–25 ml is enough to reduce the opening pressure sufficiently .[3]

II.6 Outcome and determinants of mortality of cryptococcal meningitis

Despite antifungal treatment, HIV related CM mortality in the developing world remains between 15% and 43% at two weeks and 22 to 96% at 10 to 12 weeks. The median time to death following hospital admission with CM is 10–13 days. Altered mental status, CSF fungal burden, older age (>50 year), high peripheral white blood cell count, fluconazole-based induction treatment, and slow clearance of CSF infection are associated with two weeks mortality. However, Low body weight, anemia (Hb <7.5g/dl) and high CSF opening pressure are independently associated with mortality at 10 weeks [4]

III. METHODOLOGY

III.1. Study design and setting

We conducted a prospective observational study in HIV positive patients with confirmed CM at CHUK, Internal medicine Department. Patients were recruited from admitting wards between 1st February 2019 and 31st January 2020.

University Teaching Hospital of Kigali (CHUK) is the largest hospital located in District of Nyarugenge at KN4 Ave, Kigali city, it is also the biggest referral hospital of the country with a capacity of 519 beds. CHUK provides quality healthcare to the population, training, clinical research and technical support to district hospitals. CHUK has 18 clinical services and specialties among which Internal Medicine Department consists of four inpatients Units with 68-bed capacity.

III.2. study population

Patients with HIV- related CM confirmed by CSF analysis meeting selection criteria.

III.3.Selection criteria

III.3.1 Inclusion criteria

Male and female patients aged 16years and above

HIV infected patients with confirmed CM with no other known underlying immunosuppressive condition.

III.3.2.Exclusion criteria.

HIV negative patients diagnosed with CM.

Patients who refused to comply for enrolment to the study.

Patients with altered mental status who are not accompanied by next of kin

III.4 .Sampling method

This was a non-random purposeful study.

III.5.Data collection and analysis

Data collection was done using a coded data sheet/questionnaire of variables under investigation (see appendix). Investigator recorded data herself. The investigator did not influence in any case the treating team in the management or discharge plan of the patients.

Patients were primarily admitted to CHUK through the main accident and emergency department of the hospital. From there, emergency team then internal medicine team always did initial assessment on day or night call was contacted for review. After a full assessment, all necessary investigations including CSF analysis if meningitis suspected were requested then the patients were admitted in internal medicine wards. After CSF analysis, the patients who were confirmed with CM and meeting inclusion criteria were recruited in the study within 24hours.

Data were initially extracted from the file of the patients and interview to the patients or next of kin if the patient was not able to sign consent and provide information. Patients admitted in absence of a researcher were retrieved using medical register in different wards of internal medicine within 24 hours post admission.

Data collection was focused on demographic and management of CM, including four elements: Successful induction phase with amphotericin B based regimen, management of raised intracranial pressure with serial LP according to initial opening pressure, prevention and management of amphotericin B toxicities. During the hospital stay, patients were followed once a week in order to ascertain whether any complication occurred and its management and whether survived to discharge. After two weeks of treatment (induction phase), outcome of the patient was recorded. Those who survived to discharge were followed using phone call once in four weeks until 10 weeks (end of consolidation phase) and outcome at 10 weeks was recorded.

We defined:

C*omplete fulfillment* of the protocol by complete respect of all three elements including in management of CM, which included effective three-phased therapy, effective screening, prevention and management of amphotericin B toxicities, effective raised ICP management.

Incomplete (partial) fulfillment of the protocol, if one or two components of management of CM are respected

No fulfillment of the protocol, none of three components of CM management is respected.

Data were recorded using Epi-data version 3.1 and analyzed using Statistical Package of social sciences software (SPSS), version16.0. Pearson's chi square was calculated to compare continuous variables and p value < 0.05 was considered statistically significant. A multivariate analysis was done to determine variables associated with increased risk of mortality.

III.6.Ethical considerations

The research protocol obtained approval, respectively from Internal medicine department, Ethical committee at CHUK and UR ethical and research committee at School of Medicine (CMHS /IRB). An informed consent was obtained from patients or next of kin in case the patients were not able to give his/her consent. Participation in the study was voluntary and did not affect in any case the patient's management. The information obtained was treated confidentially and only used for research purposes by a researcher.

IV.RESULTS

Demographic characteristics

Forty-one HIV positive patients diagnosed with CM were enrolled and followed for 10 weeks. Table one below demonstrates baseline demographic characteristics of population; 80.5% of patients were from city of Kigali and 19.5% were coming from other four Provinces of the country. 65.9% of populations were males and 34.1% were females with male to female ratio of 1.93:1. The patients' age was ranging between 18 and 69 years with a mean age of 40years. 48.8% of patients were married, 36.6 % were single, divorced and widows represented 14.6%. CBHI was the most used insurance at 75.6%.

Table 1: Patients' Demographic characteristics

Variable (n=41)		**Frequency**	**%**
Province of origin	North	4	9.8
	South	1	2.4
	East	1	2.4
	West	2	4.9
	Kigali city	33	**80.5**
Gender	Male	27	**65.9**
	Female	14	34.1
Age range	16 to 30 years	8	19.5
	31 to 44 years	15	36.6
	45 to 59 years	17	**41.5**
	60 years and above	1	2.4
Marital status	Married	20	**48.8**
	Single	15	36.6
	Divorced	3	7.3
	Widow	3	7.3
Type of medical insurance	CBHI	31	**75.6**
	No insurance	6	14.6
	Other insurances	4	9.8

Clinical characteristics

Clinical characteristics of population**;** 73.1% were living with HIV for more than 5years; the majority (75.6%) were on ART with poor adherence estimated at 93.5%. The most presenting symptoms were headache for 95.1% followed by vomiting for 87.8% and Meningeal signs for 63.4 %. (Figure 1 and 2).

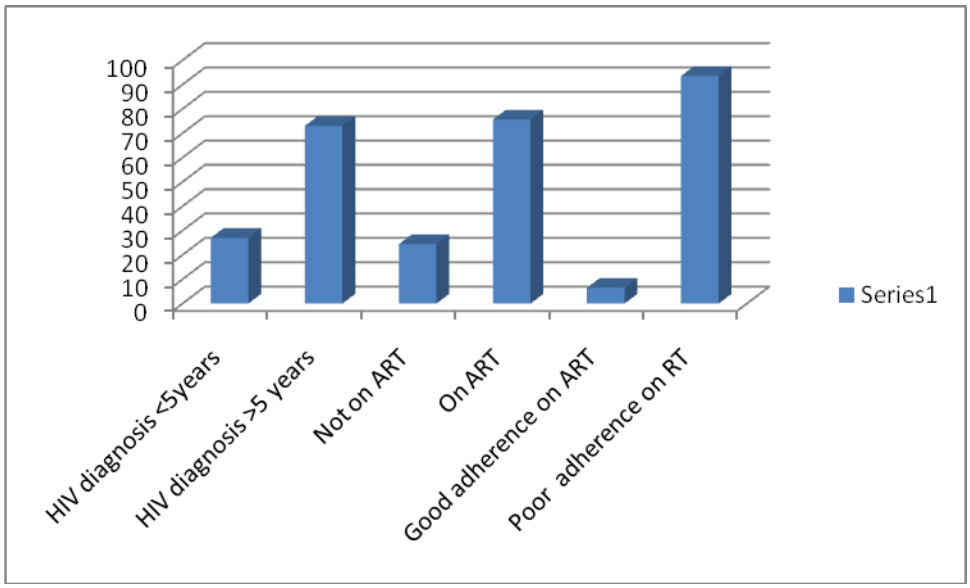

Figure 1: Past medical history

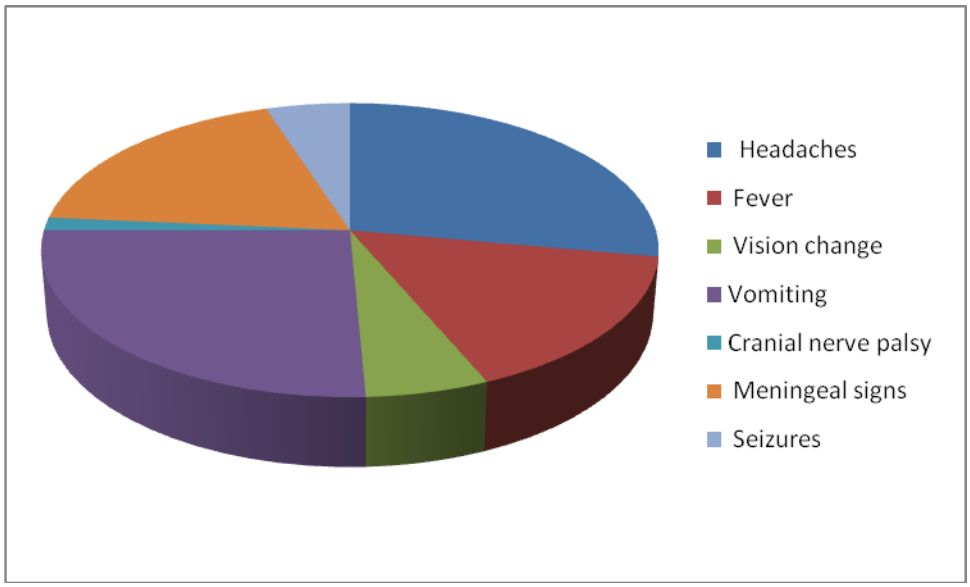

Figure 2: Presenting symptoms

Management Profile.

All patients received antifungal treatment and the respect of antifungal dosage was estimated at 65.9%. During Amphotericin B toxicity prevention, 31.7% received IV fluids plus potassium supplements while 68.3% received IV fluid only. In addition, 75.6 % received IV Paracetamol plus IV Hydrocortisone as premedication. Therapeutic lumbar puncture was performed at least once in 56.1%. %. And CSF opening pressure was measured for only two of 41 patients representing 4.9%; both had high CSF opening pressure. In consolidation phase, 22.6% abandoned fluconazole and 72.4% continued fluconazole with 57.2% of good adherence. (**Table 2)**

Considering the respect of CM treatment protocol, it was fully respected in 36.6%, partially respected in 48.8% and not respected in 14.6% of cases. The mortality was 58.5% within 10 weeks of follow up with 29.3% occurring in the first two weeks**. (Table2**)

Table 2: Management profile

Variables		**%**
Antifungal treatment	Received	100
	Not received	0
Antifungal dosage	Respected	65.9
	Not respected	**34.1**
AmphoB toxicity prevention	IV Paracetamol	24.4
	IV Paracetamol+ Hydrocortisone	**75.6**
	IVF + K CL	31.7
	IVF alone	**68.3**
CSF opening pressure	Measured	**4.9**
	Not measured	96.1
Level of CSF opening pressure	Normal (<200mmhg	0
	High(>200mmhg)	100
Therapeutic LP performed	Performed	56.1
	Not performed	**44.9**
Adherence on fluconazole	Good adherence	57.1
	Poor adherence	**42.9**
Respect of protocol	Fully respected	**36.6**
	Partially respected	48.8
	Not respected	14.6
Outcome at 2 weeks	Not Survived	**29.3**
	Survived	70.7
Outcome at 10 weeks	Survived	41.5
	Not survived	**58.5**

Factors associated with mortality after two weeks of follow up

Mental change was statistically associated with outcome with 17/20(85%) of patients presented without mental change who survived after two weeks of follow up and 9/12(75%) of deaths in patients presenting mental change (p=0.006). Therapeutic lumbar puncture was associated with outcome with good survival (78.8%) in patients who underwent therapeutic lumbar puncture at least once and relatively increased mortality 7/12 (58.3%) in patients for whom therapeutic lumbar puncture were not perfomed (p=0.013). The respect of CM protocol within two weeks of follow up was found to be statistically associated with the outcome with a very large majority of survival after two weeks (93.3%) in patients for whom the protocol was correctly followed and many deaths (66.7%) within patients for whom the protocol was not respected at all. (p=0.018) **(Table 3)**

Table 3: Bivariate analysis of factors associated with mortality after two weeks of follow up

Variables		**2 weeks outcome**			
		Not survived	**Survived**	**Total**	**P value**
Gender	Male	8(29.6%)	19(70.4%)	27(100%)	0.944
	Female	4(28.5%)	10(71.5%)	14(100%)	
Age	16 to 30 years	3(37.5%)	5(62.5%)	8(100%)	0.660
	31 to 44 years	3(20%)	12(80%)	15(100%)	
	45 to 59 years	6(35.3%)	11(64.7%)	17(100%)	
	>60 years	0(0%)	1(100%)	1(100%)	
Headache	No	1(50%)	1(50%)	2 (100%)	0.505
	Yes	11(28.2%)	28(71.8%)	39(100%)	
Vomiting	No	1(20%)	4(80%)	5(100%)	0.539
	Yes	11(30.6%)	25(69.4%)	36(100%)	
Mental change	No	3(15%)	**17(85%)**	20(100%)	**0.006**
	Yes	9(42.9%)	12(51.7%)	21(100%)	
Seizures	No	10(29.4%)	24(70.6%)	34(100%)	0.965
	Yes	2(28.6%)	5(71.4%)	7(100%)	
Treatment of Ampho B toxicity	IVF alone	5(55.6%)	4(44.4%)	9(100%)	0.386
	K^+ supplement	2(28.6%)	5(71.4%)	7(100%)	
	IVF+k+ supplement	1(10%)	9(90%)	10(100%)	
	IVF+Transfusion	0(0%)	1(100%)	1(100%)	
	K^+supplement+ Transfusion	0(0%)	2(100%)	2(100%)	
	None	1(25%)	3(75%)	4(100%)	
Therapeutic LP performed	Not performed	7(38.9%)	11(61.1%)	18(100%)	**0.013**
	Performed	5(21.7%)	**18(78.3%)**	23(100%)	
respect of CM treatment protocol	Fully respected	1(6.7%)	**14(93.3%)**	15(100%)	**0.018**
	Partially respected	7(35%)	13(65%)	20(100%)	
	Not respected	4(**66.7%)**	2(33.3%)	6(100%)	

Factors associated with mortality after 10 weeks of follow up

Adherence to fluconazole was associated with outcome after 10 weeks of follow up, with a good survival (91.7%) within patients with good adherence and a significant mortality (55.6%) within patients with poor adherence. (p=0.018). The overall respect of CM treatment protocol was associated with outcome after 10 weeks of follow up, with more survival (78.6%) among patients for whom the protocol was well followed and increasing mortality 53.8% and 100% for patients to whom the protocol was followed partially and not followed respectively. (p=0.017) **(Table 4)**

Table 4: Bivariate analysis of factors associated with mortality after 10 weeks of follow up

Variables		**10 weeks outcome**			
		Not survived	**Survived**	**Total**	**P value**
Gender	Male	9(50%)	9(50%)	18(100%)	0.228
	Female	3(27.3%)	8(72.7%)	11(100%)	
Age	16 to 30 years	1(25%)	3(75%)	4(100%)	0.355
	31 to 44 years	4(30.8%)	9(69.2%)	13(100%)	
	45 to 59 years	6(54.5%)	5(45.5%)	11(100%)	
	>60 years	1(100%)	0(0%)	1(100%)	
Mental change	No	7(38.9%)	11(61.1%)	18(100%)	0.728
	Yes	5(45.5%)	6(54.5%)	11(100%)	
Seizures	No	10(41.6%)	14(58.4%)	24(100%)	0.945
	Yes	2(40%)	3(60%)	5(100%)	
Adherence to fluconazole	Good	1(8.3)	11(**91.7**)	12(100)	**0.018**
	Poor	5(55.6)	4(44.4)	9(100)	
Respect of CM treatment protocol	Fully respected	3(21.4)	11(**78.6**)	14(100)	**0.017**
	Partially respected	7(53.8)	6(46.2)	13(100)	
	Not respected	2(100)	0(0.00	2(100)	

Factors strongly associated with mortality

On multivariate analysis, factors associated with mortality were therapeutic lumbar puncture (OR=2.29;95%CI:0.58to4.42; P=0.023), adherence on fluconazole (OR=2.062;95%CI:0.06 to3.82; P=0.018) and overall respect of CM treatment protocol (OR= 1.07;95%CI 0.63to 5.82; P=0.007). **(Table5)**

Table 5: Multivariate analysis of variables associated with mortality.

Multivariate analysis of variable associated with 2 weeks outcome			
Variable	**P value**	**Odd ratio**	**95% CI**
Mental change	0.050	0.23	[0.052 - 9.05]
Therapeutic lumbar puncture	0.023	2.29	[0.58-4.42]
Multivariate analysis of variable associated with 10 weeks outcome			
Variable	**P value**	**Odd ratio**	**95% CI**
Adherence on fluconazole	0.018	2.062	[0.06-3.82]
Overall respect of CM protocol	0.007	1.07	[0.63 - 5.82]

V. DISCUSSION

Our patients were predominantly young male with mean age of 40 years and male to female ratio of 1.93 to1. Our results are comparable with the ones found by Mdodo R.et al in Kenya where the most participants were young male with age range of 19 to 60 years and mean age of 35years.[1]The majority of our patients were living in city of Kigali. This predominance was related to the high prevalence of HIV (1.9 times higher) in urban areas (4.3% in city of Kigali) compare to the rural areas.[26]

There is a remarkable gap in performance of therapeutic lumbar puncture at the extent that 18 patients in 41 did not receive any therapeutic lumbar puncture. In addition to that, CSF opening pressure was measured for only two patients in 41. Shmuel et al in Washington D.C and Melissa A. et al in-COAT trial done in Uganda and South Africa found the similar results[21, 14]. The paucity of performance of therapeutic lumbar puncture at CHUK might be explained by lack of clear hospital-based protocol of management of CM and health providers do not have enough information about the role of lumbar puncture in management of CM. The rarity of measurement of CSF opening pressure in LMIC is due to lack of manometers and practical ICP management protocol[16]; the same situation at CHUK. Fortunately, the study done in Tanzania by John Meda et al demonstrated a good evidence that intravenous tubing sets assembled to spinal needles coupled with a meter measuring stick are an accurate alternative to manometers to measure CSF opening pressure.[1]

Management of Amphotericin B toxicities was not accurate; some patients received IV fluids alone instead of IV fluids combined with potassium supplements as per protocol. On the other hand, the study done in Uganda by Nathan C. Bahr et al shows an improved survival with IV fluids coupled with universal electrolyte supplementation of K+ andMg2+, electrolyte monitoring and standardized electrolyte replacement[23]. Furthermore, a high number of patients received routinely additional hydrocortisone, which is normally used in case of severe infusion reaction[3]. The routine use of corticosteroids has been discouraged in the study done by J. Beardsley for the reason that it slows fungal clearance in CSF[12]. The reason of inaccurate management of amphotericin B toxicities at CHUK are not well studies, but it is thought to be either due to unavailability of mediations (mainly potassium supplement) in the hospital or lack of protocol insisting on electrolytes management as a package of management of CM.

The mortality rate was remarkably high and estimated at 29.3 % within two weeks and 58.5% after 10 weeks of follow up.

This high mortality is superior to one found in the prospective trials done in Thailand, Uganda, Malawi and South Africa by Joseph N et al where mortality was estimated at 17% at 2 weeks and 34 % at 10weeks.[4] However this mortality stays in the range of one reported in LMIC estimated between 35% and 65%.[6]

Regarding the factors associated with mortality, therapeutic lumbar puncture was found to be associated with mortality. Our findings correlate with the ones of the study done in Uganda by Melissa A. et al and Tihana Bicanic et al, which shows relative improvement in survival with therapeutic lumbar puncture performed at least once regardless of initial intracranial pressure[14, 2].

Accurate management of cryptococcal meningitis should include appropriate antifungal therapy without leaving aside the control of intracranial pressure.

In accordance with the study done by Jarvis et al where non adherence to fluconazole as secondary prophylaxis was associated with CM relapse and mortality estimated at 33%, we found that poor adherence on fluconazole was likewise significantly associated with mortality.[24]

The overall respect of CM treatment protocol contributed to mortality. Comparable results were found in the study by Shmuel Shoham et al where they found a major deviation on CM treatment guidelines, with failure to manage high ICP among others, estimated at 54% .[21]

Potential limitations of our study are System related problems, which might have led to inadequate management thus contributing negatively to the outcome while these were not part of the study. Indeed, the study period constrained us to be limited on two phases of therapy (induction and consolidation). Long-term outcome counting three phases may be a subject for another study in the future. As strength of the study, data were collected prospectively, which minimized confounding errors.

VI.CONCLUSION AND RECOMMANDATION

VI.1.Conclusion

Despite the availability of diagnostic modalities and the use of appropriate antifungal treatment available, we are still experiencing an increased mortality rate in HIV patients with CM in CHUK. This mortality increases gradually from induction to consolidation phase.

The factors mostly correlated with an increased risk of mortality are failure to perform serial lumbar puncture and measurement of CSF opening pressure, which are the cornerstone in management of raised intracranial pressure commonly found in CM and poor adherence on fluconazole during consolidation phase.

This highlights the need of new measures emphasizing on prompt management of raised intracranial pressure by serial therapeutic lumbar puncture as required and develop strategies for long-term support to patients aiming to an improved compliance to antifungal treatment in consolidation phase.

VI.2.Recommendations

In the light of these results, we recommend the following:

To CHUK internal medicine department:

To promote a CM treatment protocol including antifungal therapy, appropriate management of amphotericin B toxicities, raised ICP management by serial LP, CSF opening pressure measurement and documentation.

To primary health facilities and HIV clinics:

To enhance follow up of patients mainly those who are living with HIV for many years by insisting on adherence to ART and educate them on CM symptoms for early recognition and consultation.

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25. Rwanda Population-Based HIV Impact Assessment Rphia 2018–2019

VIII.APPENDICES.

1.Data collection form

Identification

Study number	
Date of enrollment	
Hospital number of the patient	
Date of admission	
Date of discharge	
Mobile phone/ contact	1. 2. 3.
HIV clinic or Health center following the patient	

2. Social Demographic

Age	
Gender	Male Female
Marital status	Single Married Separated /divorced

Residence	Province District
Medical insurance	Mutuelle de santé Other insurance. :… No insurance
Weight	
Height	
BMI	
Ubudehe category	1 2 3 4 Unclassified

3. Medical history

Diagnosis of HIV	At admission < 1 year 1-5 years 5-10years >10years
Patient on anti retroviral therapy?	Yes No

Adherence on medication	Good Poor
Recent CD4 Count	>200 cells/mm^3 100-199cells/mm^3 50-99cells/mm^3 <50cells/mm3 unknown
Viral load	

4.Clinical presentation

Duration of symptoms	< 1 week 1-2 weeks 2-4 weeks >4 weeks
Signs and symptoms at presentation	Mental changes Headache Fever Vision changes Vomiting Weight loss Cranial nerve palsy Meningeal signs

	Others …
CSF opening pressure measured at presentation.	Yes No
If yes, opening pressure	< 25cm H_2O > 25cm H_20

5. Management

Antifungal treatment given	Yes No
If yes, which one?	IV.Amphotericin (0.7-1mg/kg) plus Fluconazole 800mg-1200mg daily IV.Amphotericin B alone Fluconazole alone Other option…
Dose respected	Yes No
Prevention of Amphotericin B toxicity	IV.Fluid given per protocol Yes No K+ supplement given per protocol Yes No

	Other medication received…
Amphotericin B toxicity screened per protocol	Yes No
If yes,	K+… Creatinine… Hb…
Therapeutic serial LP performed	Yes No
If yes, how many times per week	1× 2 × 3-4× >4 times
Amphotericin B toxicity managed.	Yes No

If yes, which medication received?	IV.Fluid KCL, Transfusion Amphotericin B stopped Other option …
Fluconazole maintained in consolidation phase	Ye no
If yes, how is the adherence	Good Poor

6. Evolution and outcome

Outcome at 2 weeks	Patient died Patient improved Patient not improved
If died; cause of death	
CM therapeutic protocol respected.	Fully respected Partially respected Not respected at all
Outcome at 10 weeks	Patient survived Patient not survived
Cause of death	

UNIVERSITY OF RWANDA

COLLEGE OF MEDICINE AND HEALTH SCIENCES

CMHS INSTITUTIONAL REVIEW BOARD (IRB)

Kigali, 26th /10/2018

Dr UWIMANA Marie Grace
School of Medicine and Pharmacy, CMHS, UR

Approval Notice: No 366/CMHS IRB/2018

Your Project Title ***"Therapeutic Aspects of Cryptococcal Meningitis in HIV Patients and Factors Contributing to Mortality. Case of CHUK"*** has been evaluated by CMHS Institutional Review Board.

		Involved in the decision		
		Yes	No (Reason)	
Name of Members	**Institute**		**Absent**	**Withdrawn from the proceeding**
Prof Kato J. Njunwa	UR-CMHS	X		
Prof Jean Bosco Gahutu	UR-CMHS	X		
Dr Brenda Asiimwe-Kateera	UR-CMHS	X		
Prof Ntaganira Joseph	UR-CMHS	X		
Dr Tumusiime K. David	UR-CMHS	X		
Dr Kayonga N. Egide	UR-CMHS	X		
Mr Kanyoni Maurice	UR-CMHS	X		
Prof Munyanshongore Cyprien	UR-CMHS	X		
Mrs Ruzindana Landrine	Kicukiro district		X	
Dr Gishoma Darius	UR-CMHS	X		
Dr Donatilla Mukamana	UR-CMHS	X		
Prof Kyamanywa Patrick	UR-CMHS		X	
Prof Condo Umutesi Jeannine	UR-CMHS		X	
Dr Nyirazinyoye Laetitia	UR-CMHS	X		
Dr Nkeramihigo Emmanuel	UR-CMHS		X	
Sr Maliboli Marie Josee	CHUK	X		
Dr Mudenge Charles	Centre Psycho-Social	X		

After reviewing your protocol during the IRB meeting of where quorum was met and revisions made on the advice of the CMHS IRB submitted on 16th October 2018, **Approval has been granted to your study.**

Please note that approval of the protocol and consent form is valid for **12 months**.
You are responsible for fulfilling the following requirements:

1. Changes, amendments, and addenda to the protocol or consent form must be submitted to the committee for review and approval, prior to activation of the changes.
2. Only approved consent forms are to be used in the enrolment of participants.
3. All consent forms signed by subjects should be retained on file. The IRB may conduct audits of all study records, and consent documentation may be part of such audits.
4. A continuing review application must be submitted to the IRB in a timely fashion and before expiry of this approval
5. Failure to submit a continuing review application will result in termination of the study
6. Notify the IRB committee once the study is finished

Sincerely,

Date of Approval: The 26th October 2018

Expiration date: The 26th October 2019

Professor Kato J. NJUNWA
Chairperson Institutional Review Board,
College of Medicine and Health Sciences, UR

Cc:

- Principal College of Medicine and Health Sciences, UR
- University Director of Research and Postgraduate Studies, UR

C.H.U.
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Ethics Committee / Comité d'éthique

January 21st, 2019 **Ref.: EC/CHUK/006/2019**

Review Approval Notice

Dear **Uwimana Marie Grace**,

Your research project: ***"Therapeutic aspects of cryptococcal meningitis in HIV patients and factors contributing to mortality at CHUK"***

During the meeting of the Ethics Committee of University Teaching Hospital of Kigali (CHUK) that was held on 21st January, 2019 to evaluate your request for ethical approval of the above mentioned research project, we are pleased to inform you that the Ethics Committee/CHUK has approved your research project.

You are required to present the results of your study to CHUK Ethics Committee before publication.

PS: Please note that the present approval is valid for 12 months.

Yours sincerely,

ETHICS COMMITTEE
CHUK

Mr. Emmanuel MUNYANEZA
The Secretary, Ethics Committee,
University Teaching Hospital of Kigali

<<University teaching hospital of Kigali Ethics committee operates according to standard operating procedures (Sops) which are updated on an annual basis and in compliance with GCP and Ethics guidelines and regulations>>

B.P. :655 Kigali- RWANDA www.chk.rw Tél. Fax : 00 (250) 576638 E-mail :chuk.hospital@chukigali.rw

2.Consent form

URUPAPURO RWO KWEMERA KUGIRA URUHARE MU BUSHAKASHATSI

Nitwa UWIMANA Marie Grace umuganga w'umunyeshuri wiga ibijyanye n'indwara zo mu mubiri muri Kaminuzay'u Rwanda, nkaba ndigukora ubushakashatsi kuri Mugiga iterwa na kiributokoke mu bantu babana n, ubwandu bw'agakoko gatera SIDA, n,uburyo bavurwa mu bitaro bya CHUK.

Urasabwa kubanza gusobanukirwa intego y'ubu bushakashatsi, inyungu n'ingaruka zishobora kubaho igihe wemeye kubugiramo uruhare.

Intego: kureba uko abarwayi babana n, ubwandu bw'agakoko gatera SIDA barwaye mugiga iterwa Na kiributokoke bavurwa mu bitaro bya CHUK.

Ngaruka:

Nta ngaruka ugize uruhare muri ubu bushakashatsi azajya.

Inyungu:

Ibizava muri ubu bushakashatsi bizafasha kandi kuvura neza abarwayi babana n, ubwandu bwa SIDA bagaragaweho n'indwara yamugiga itewe na kiributokoke. Nta nyungu y'amafaranga uwagize uruhare muri ubu bushakashatsi azabukuramo.

Ibanga:

Amakuru yose kuri buri muntu azajya abikwa n'umushakashatsi kugirango akoreshwe mu bushakashatsi gusa.

Ibibazo:

Umuntu wese wemeye kugira uruhare muri ubu bushakashatsi yemerewe kubaza ibibazo byose igihe cyose yifuza ubundi busobanuro. Nimero ya telefoni yanjye ni: 0788610741.

Uburenganzira bwo kwivana mu bushakashatsi

Ufite uburenganzira bwo kwivana mu mubare w'abakorerwaho ubushakashatsi igihe mbishakiye kandi nta ngaruka ugize.

Amasezerano yo kwemera gukorerwaho ubushakashatsi

Maze gusoma ibyanditse hejuru kandi nabisobanukiwe. Nasobanuriwe birambuye mu rurimi numva intego, inyungu n'ingaruka muri ubu bushakashatsi. Nasobanuriwe n'uko nemerewe kwivana mu mubare w'abakorerwaho ubushakashatsi igihe mbishakiye nta ngaruka ngize.Nshyize umukono kuri aya masezerano nsobanukiwe kandi nemera ko nkorerwaho/umurwayi wanjye akorerwaho ubushakashatsi.

Umukono wanjye……itariki……

Umukono w’umurwaza ……..itariki……

Nasobanuriwe umurwayi/umurwaza muburyo birambuye intego, inyungu n’ingaruka by’ububushakashatsi.

Ubushakashatsi…….itariki:…….

2. CONSENT FORM

Patient's number:……..

I am Dr UWIMANA Marie Grace a postgraduate student at University of Rwanda in the department internal medicine who is carrying out a study on **"Therapeutic aspects of cryptococcal meningitis in HIV patients and factors contributing to mortality. Case of CHUK"**

You will be required to understand its purpose, risks and benefits before you agree to participate in it.

Aim: To contribute in management and improve outcome of HIV patients with CM at CHUK.

Risks to the participants

There are no major risks in this study.

Benefits

The information from the study will provide useful input for management of cryptococcal meningitis.

There are no financial benefits to be provided to the participants in the study.

Confidentiality:

All information will be kept confidential by the principal investigator for purposes of the study strictly.

Questions

Participants are free to ask questions or seek any clarifications about the study when they wish.

My phone number: 0788610741.

Rights to withdraw from the study

You are free to withdraw from the study at any time without any consequence.

Statement of consent

I have read the information above and understood the content. I have had a full explanation of the nature and purpose of the study, risks and benefits in a language I understand. I have understood that I have right to withdraw from the study at anytime.

By signing this consent form, I understand that I am accepting to be enrolled in this study.

I hereby sign for myself…/next of kin……as a proof to participate in the study.

Names :……..Date :……
……

I have explained the purpose of the study to the participant to the best of my knowledge and he has fully understood the purpose, benefits and risks to him or her.

Signature:……Date :……