

# Burden of *Cryptococcal antigenemia* Among Symptomatic HIV Patients in Agbor, Nigeria

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*Cryptococcal antigenemia* is a significant opportunistic infection among HIV-positive individuals, contributing to high morbidity and mortality if not detected early. This study aimed to determine the burden of cryptococcal antigenemia (CrAg) among antiretroviral therapy-naïve patients with CD4 counts below 200 cells/mm<sup>3</sup> at Central Hospital, Agbor, Nigeria. A cross-sectional hospital-based study was conducted, utilizing the lateral flow assay for CrAg detection. Of the 188 participants, 127 (67.6%) were female, and 61 (32.4%) were male, with a mean age of  $37.37 \pm 12.01$  years. The prevalence of CrAg positivity was 4.3% (95% CI: 1.50%–7.39%), with 2 males (3.3%) and 6 females (4.7%) testing positive. Although CrAg positivity was higher in females, the difference was not statistically significant. As a precursor to cryptococcal meningitis, a leading cause of mortality in HIV-infected individuals, early CrAg detection can enable timely intervention to reduce disease progression and improve outcomes. This study underscores the importance of routine CrAg screening in high-risk HIV populations to mitigate the burden of cryptococcal disease.

**Keywords:** Cryptococcal, Antigenemia, Antigen, Infection, Prevalence

## Introduction

*Cryptococcus neoformans* infection is a life-threatening condition, particularly among individuals with immunosuppressive diseases such as HIV/AIDS. Cryptococcosis is a major opportunistic infection in people with advanced HIV disease (AHD), defined as a CD4 cell count  $\leq 200/\mu\text{L}$ , and contributes significantly to AIDS-related mortality worldwide (World Health Organization [WHO], 2023; Katoch et al., 2023; Oladele et al., 2023). It primarily manifests as meningitis, accounting for approximately 15–20% of HIV-related deaths globally, with an estimated 75% of these occurring in sub-Saharan Africa (Derbie et al., 2020; Rajasingham et al., 2022). In 2017, the global prevalence of cryptococcosis among individuals with AHD was estimated at 6.0%, with variations between high- and low-income countries (Eric et al., 2023). The infection, caused by *Cryptococcus neoformans* and *Cryptococcus gattii*, affects the membranes surrounding the brain and spinal cord, leading to symptoms such as severe headache, fever, and confusion (Dao et al., 2024; Taha et al., 2024). The fungus possesses virulence factors, including a polysaccharide capsule, melanin deposition in the cell wall, growth at 37°C (host temperature), and production of extracellular enzymes like laccases and urease (De Leon-Rodriguez et al., 2018).

Cryptococcal antigen (CrAg) is a detectable biomarker in the blood of infected individuals, enabling early diagnosis through methods such as enzyme immunoassay and lateral flow assay

(LFA) (Wake et al., 2023). The LFA, a point-of-care test, provides rapid results within 15 minutes, making it invaluable in resource-limited settings (Negri et al., 2024). CrAg testing has revolutionized the diagnosis of cryptococcal disease, particularly in areas with limited laboratory infrastructure, offering high sensitivity and specificity for detecting cryptococcal meningitis (Jarvis et al., 2009; Kabanda et al., 2014; Vijayan et al., 2013). Studies have consistently validated its diagnostic performance (Lindsley et al., 2011; Escandón et al., 2013; Huang et al., 2015; Liang et al., 2023). Since 2011, the WHO has recommended routine CrAg screening for early detection, now integrated into HIV national treatment guidelines (Dalhatu et al., 2016).

In Nigeria, cryptococcal antigenemia is a significant concern, though its prevalence remains unclear. Since 2016, Nigeria's National HIV Guidelines have mandated CrAg screening for antiretroviral therapy (ART)-naïve adults with CD4 counts below 200 cells/mm<sup>3</sup>, followed by pre-emptive antifungal therapy to prevent cryptococcal meningitis if meningitis is ruled out (Oladele et al., 2020). A study in North-Central Nigeria reported a 36% hospital-based frequency of cryptococcal meningitis, with high mortality rates (Gomerep et al., 2010). This study aims to determine the prevalence of cryptococcal antigenemia among HIV-seropositive and seronegative individuals attending Central Hospital Agbor, Nigeria.

## Materials and Methods

### Study Design

A prospective study was conducted between January and March 2024, enrolling a total of 188 inpatient and outpatient HIV-infected individuals aged above 16 years with CD4 counts below 200 cells/mm<sup>3</sup> at the Antiretroviral Therapy (ART) Centre, Central Hospital Agbor, a publicly owned secondary health-care facility. The study included all eligible patients admitted to inpatient wards or attending the outpatient ART centre with a documented CD4 count of less than 200 cells/mm<sup>3</sup>. A trained clinician evaluated each participant for overt clinical symptoms or indicators of cryptococcal meningitis, which served as an exclusion criterion. Following informed consent, participants underwent cryptococcal antigen (CrAg) testing using a lateral flow assay (LFA).

### Ethical Considerations

Participants were informed about the study's objectives and procedures, and written informed consent was obtained from those who agreed to participate. The study was approved by the Institutional Review Board of Central Hospital, Agbor. To ensure confidentiality, samples and questionnaires were delinked from personal identifiers and assigned unique codes.

### Data Collection

Socio-demographic and clinical information were collected using structured questionnaires administered by trained clinicians to ensure data completeness. Clinical data, including presenting symptoms, coexisting diagnoses, and current treatments, were obtained through questionnaires and medical record reviews. Five millilitres (5.0 mL) of blood were aseptically collected from each participant into labelled EDTA vacutainer tubes. Plasma was separated by centrifugation at 3000 rpm for 5 minutes, and serum was transferred into another labelled tube for analysis.

### Serological Assay

Cryptococcal antigen testing was performed using the Cryptococcal Antigen Lateral Flow Assay (CrAg LFA) (Immy, USA), a non-automated immunochromatographic test for the qualitative detection of capsular polysaccharide antigens of the *Cryptococcus* species complex (*Cryptococcus neoformans* and *Cryptococcus gattii*) in serum, plasma, and whole blood (Huang et al., 2015). Paired assays of serum samples from known CrAg-positive patients and control subjects were conducted simultaneously to ensure quality control.

### Statistical Analysis

Data were entered and analyzed using the Statistical Package for the Social Sciences (SPSS) Version 25. Descriptive statistics, including mean and standard error of the mean (SEM), were calculated. Frequency tables were used to present variables.

## Results

A total of 188 individuals participated in this study, comprising 127 (67.6%) females and 61 (32.4%) males, with a female-to-male ratio of 2:1 (Table 1). The seroprevalence of cryptococcal antigenemia among the study population was 4.3% (Table 2).

The distribution of cryptococcal antigen (CrAg) positivity across age groups is presented in Table 3. The age group 31–45 years exhibited the highest prevalence at 5.0%, followed closely by the 46–60 years age group with a prevalence of 4.7%. The age groups 61–75 years and 76–90 years showed no CrAg-positive cases (0%). Regarding gender distribution, a higher proportion of females (6, 4.7%) tested CrAg-positive compared to males (2, 3.3%) (Table 4).

**Table 1** Demographic Characteristics of Study Population

Gender	Frequency (%)
Male	61 (32.4%)
Female	127 (67.6%)
<b>Total</b>	<b>188 (100%)</b>

**Table 2** Prevalence of Cryptococcal Antigenemia Amongst Study Participants

CrAg Test	Frequency	Percentage (%)
CrAg Positive	8	4.25
CrAg Negative	180	95.75

**Table 3** Seroprevalence of Cryptococcal antigenia among age groups

Age Group	Subjects Tested	CrAg Positive	Total
16–30	61	2 (3.2%)	1.06
31–45	80	4 (5.0%)	2.13
46–60	43	2 (4.7%)	1.06
61–75	3	0 (0.0%)	0
76–90	1	0 (0.0%)	0
<b>Total</b>	<b>188</b>	<b>8</b>	<b>4.25</b>

**Table 4** Distribution of Cryptococcal Antigenemia Amongst Gender

Gender	Overall (n=188)	CrAg Positive	CrAg Negative
Male	61 (32%)	2 (3.3%)	59 (96.7%)
Female	127 (68%)	6 (4.7%)	121 (95.3%)
<b>Total</b>	<b>188</b>	<b>8</b>	<b>180</b>

## Discussion

This study found an overall cryptococcal antigen (CrAg) prevalence of 4.3% among HIV-infected individuals attending Central Hospital, Agbor, Nigeria, indicating a significant burden of cryptococcal disease in this population. This prevalence may be linked to environmental exposure to *Cryptococcus neoformans*, potentially from avian sources or other environmental reservoirs. The reactivation of latent infections during immunosuppression likely contributes to the presence of *Cryptococcus* species in HIV-infected individuals (Chukwuanukwu et al., 2013). Previous studies in Nigeria have reported CrAg prevalence ranging from 2.2% to 13.1% among HIV-infected patients (Joseph et al., 2015; Ogba & Abia-Bassey, 2015), which aligns with the 4.3% observed in this study. Variations in prevalence across studies may be attributed to differences in diagnostic methods employed.

Females exhibited a slightly higher CrAg prevalence (4.7%) compared to males (3.3%), though this difference was not statistically significant. This finding is consistent with Osazuwa et al. (2012), who also reported higher cryptococcal antigen levels among females. The reasons for this gender disparity warrant further investigation, potentially involving demographic, immunological, or environmental factors. Progress is being made in Nigeria to implement routine CrAg screening and pre-emptive treatment as part of the World Health Organization's (WHO) recommended package of care for advanced HIV disease (AHD), including CrAg screening and pre-emptive fluconazole therapy, through collaborative efforts between the Nigerian government and its developmental partners (Shroufi et al., 2021; World Health Organization, 2023).

Our data indicate that younger and middle-aged adults, particularly those in the 31–45 (5.0%) and 46–60 (4.7%) age groups, are more likely to test CrAg-positive. This age-related trend may be influenced by factors such as health-seeking behavior, occupational exposure, or the higher proportion of these age groups within the HIV-infected population, leading to more frequent interactions with healthcare facilities and increased screening and detection rates (Atchessi et al., 2018; Adewoye et al., 2023). The study underscores the critical need for enhanced awareness and integration of CrAg screening into HIV care strategies to reduce associated morbidity and mortality. Routine CrAg screening should be prioritized, particularly in high-prevalence settings like Central Hospital Agbor, to facilitate early detection and pre-emptive treatment.

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## Competing Interests

The authors declare that they have no competing interests.

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