

Identifying common opportunistic infections among people with advanced HIV disease

Policy Brief



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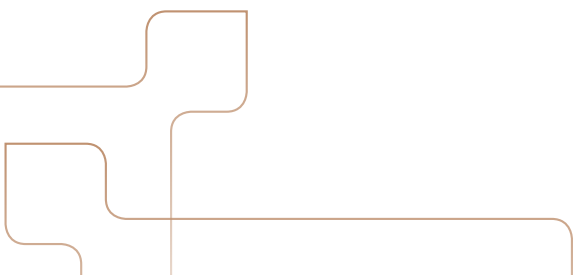
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Introduction

Definition of advanced HIV disease

For adults, adolescents, and children older than five years, advanced HIV disease is defined as a CD4 cell count <200 cells/mm³ or a WHO clinical stage 3 or 4 event at presentation for care (1,5).

- All children living with HIV younger than five years should be considered as having advanced HIV disease at presentation.
- Although children younger than five years are defined as having advanced HIV disease at presentation, those who have been receiving ART longer than one year and are clinically stable (established on ART) should not be considered to have advanced HIV disease and should be eligible for multimonth dispensing.

An estimated 630 000 people died from AIDS-related causes in 2022 (2), mainly in countries in sub-Saharan Africa. Many of these people died from tuberculosis (TB), cryptococcal meningitis and severe bacterial infections, although the

leading causes of death vary by region: in Latin America, histoplasmosis may cause more deaths than TB among people living with HIV (3).

WHO recommends a package of interventions that should be offered to everyone presenting with advanced HIV disease; this package including screening, treatment and/or prophylaxis for major opportunistic infections, rapid ART initiation and intensified adherence support interventions (4).

Diagnostics play a critical role in ensuring the success of the package of interventions being offered for advanced HIV disease. Therefore, understanding their importance and how they are best implemented are vitally important to successfully scaling up the advanced HIV disease package of care in HIV programmes in settings with a high burden of HIV infection.

This policy brief summarizes key WHO guidance on identifying the primary opportunistic infections causing many deaths among people living with HIV in HIV programmes in settings with a high burden of HIV infection.

Table 1. Key screening and diagnostic interventions in the WHO-recommended package of care for people with advanced HIV disease

	Intervention	CD4 cell count	Adults and adolescents	Children <10 years
Screening and diagnosis	Screening tools for TB disease for adults and adolescents: WHO-recommended four-symptom screen, chest X-ray, C-reactive protein Screening tools for TB disease among children: symptom screening for children living with HIV	Any	Yes	Yes (symptom screen only)
	WHO-recommended molecular rapid diagnostics as the first test for pulmonary TB diagnosis among those who screen positive for TB	Any	Yes	Yes
	Lateral flow urine lipoarabinomannan assay (LF-LAM) to assist in diagnosing TB among people with symptoms and signs of TB	≤200 cells/mm ³ (inpatient) ≤100 cells/mm ³ (outpatient) Or any CD4 count with symptoms or if seriously ill	Yes	Yes, if younger than five years, assumed to have advanced HIV disease
	Cryptococcal antigen for screening (in blood) and for diagnosis (in blood and cerebrospinal fluid (CSF))	Recommended for <100 cells/mm ³ and considered for 200 cells/mm ³	Yes	No
	Histoplasma antigen to assist in diagnosing histoplasmosis among people with symptoms and signs of histoplasmosis	Any	Yes	Yes

Source: Consolidated guidelines on HIV prevention, testing, treatment, service delivery and monitoring: recommendations for a public health approach, 2021 update (5).

Methods

Rationale, scope and development

This document responds to requests from Member States as well as from representatives of civil society for the World Health Organization to provide further technical guidance tools on existing recommendations for advanced HIV disease to promote further uptake of guidance in countries. The document was developed following internal WHO consultations with relevant technical officers, followed by external peer review. The document summarizes existing WHO recommendations on advanced HIV disease and key opportunistic infections, as well outputs of previous WHO expert consultations. The intended audience for this document is healthcare workers, national HIV programme managers, representatives of civil society.

Confidentiality undertaking and Declarations of interest (DOIs)

Confidentiality undertakings as well as declaration of interests were collected prior to engaging with external experts. The responsible technical officer in consultation with the Team Lead assessed the responses and found no conflicts of interest.

CD4 cell count testing

In 2016, WHO recommended initiating antiretroviral therapy (ART) for all people living with HIV regardless of WHO clinical stage and at any CD4 cell count (5). This expansion of ART access has led to an increase in the average CD4 cell count when ART is initiated in most settings (6); however, this average can mask the fact that the proportion of people living with advanced HIV disease has remained relatively stable (7). The proportion of individuals with advanced HIV disease when initiating ART or already receiving ART is estimated to exceed 42% in some settings (8).

CD4 cell count testing is the gateway to identifying people with advanced HIV disease when HIV is diagnosed, when they return to care after treatment interruption or when they present with acute and/or severe illness. Clinical evaluation is not a reliable proxy for CD4 cell count (9). CD4 cell count also is not consistently correlated with signs and symptoms of illness, and up to 50% of the people with advanced HIV disease may be asymptomatic or do not have clinically identifiable symptoms (10).

CD4 testing can be performed using a variety of technologies, including laboratory-based CD4 analysers, point-of-care technologies and device-free semiquantitative rapid tests (11–13).

In the WHO African Region, about 35 countries have access to CD4 testing according to 2021 estimates, of which only 21 countries have CD4 testing available in at least 50% of programme sites (14). Recent analyses suggest that, although some low- and middle-income countries have

reasonable aggregate CD4 testing capacity to conduct advanced HIV disease screening, HIV facilities in most settings globally have limited CD4 testing available (15–17).

Given the high risk of morbidity and mortality among people living with advanced HIV disease (18), priority should be given to more rapidly identifying such people.

Both laboratory-based and point-of-care CD4 testing options are available. Although laboratory-based testing can process more samples, the results are often delayed, leading to longer times on average to linkage to care and appropriate clinical decision making. Point-of-care technologies are well suited to supporting rapid and, ideally, same-day identification of advanced HIV disease. Several studies have demonstrated reliable performance of point-of-care CD4 technologies, including the device-free technology (13,19,20). WHO prequalification of in vitro devices remains critical for technical evaluations of these devices, and several CD4 technologies are WHO prequalified (21).

Most programmes have redirected resources towards purchasing viral load devices, and this has been accompanied by a reduction in CD4 testing when ART is initiated (17). Several key organizations along with representatives of civil society are reiterating the need for more CD4 testing options to diagnose advanced HIV disease, issuing an open letter addressed to pharmaceutical companies to express concern about the expected shortfall in CD4 diagnostics (22).

The use of CD4 testing declined with the introduction and scaling up of the recommendation to treat all people living with HIV, regardless of CD4 cell count, and the shift to using viral load alone for treatment monitoring (17). Several countries are scaling up their CD4 testing capacity to support the identification of people with advanced HIV disease and to link individuals to life-saving interventions (23).

Box 1. Who needs a CD4 test?

WHO supports the use of CD4 testing in the following situations.

1. To identify advanced HIV disease among individuals:
 - a. initiating ART after being diagnosed with HIV;
 - b. re engaging or re-entering care with HIV programmes following a period of disengagement;
 - c. to support identifying treatment failure or when suspected to have treatment failure;
 - d. to assess hospitalized or seriously unwell people living with HIV when clinically unstable and when clinically indicated.
2. To assess eligibility to stop co-trimoxazole prophylaxis.

HIV-associated TB

People living with HIV are up to 18 (15-21) times more likely to develop TB disease than those without HIV, and TB is a predominant cause of mortality and morbidity among people living with HIV(76). An estimated 187 000 [158 000–218 000] people living with HIV died from TB in 2021 (77), and identifying TB disease is a core component of the WHO-recommended advanced HIV disease package of care. Once TB disease is ruled out, offering TB preventive treatment as part of a comprehensive package of HIV care is necessary to prevent TB among people living with HIV (4).

Systematic screening to identify TB disease among people living with HIV is an important priority for national programmes. The urgency is greater for individuals who have advanced HIV disease since they have greater risk of mortality and morbidity. The presence of TB bloodstream infection among seriously ill people living with HIV predicts 30-day mortality (24). Autopsy studies have found that seriously ill people living with HIV often have disseminated TB (25–27). Rapid turnaround of testing services is vital to inform appropriate clinical management, and near point-of-care tests and point-of-care rapid tests may be used for rapid identification. WHO recommends using LF-LAM alongside a WHO recommended molecular test as part of the advanced HIV disease package of care (4), following systematic screening for TB disease.

Screening for TB

Systematic screening for TB disease is a core activity within HIV programmes as part of comprehensive care (4). WHO guidelines primarily encourage using the WHO-recommended four-symptom screen (current cough, fever, weight loss or night sweats), which remains the minimum necessary for screening (Box 2). When feasible and available, countries may also consider additional tools to further improve the sensitivity and specificity of screening, including using chest X-ray and C-reactive protein (28). WHO has published detailed guidance on approaches to screening for TB disease, and this is also part of the advanced HIV disease package of care (4,28).

Box 2. Summary of key recommendations on systematic screening for TB among people living with HIV

People living with HIV should be systematically screened for TB disease at each visit to a health facility.
(strong recommendation, very-low-certainty evidence)

Among adults and adolescents living with HIV, systematic screening for TB disease should be conducted using the WHO-recommended four-symptom screen, and those who report any one of the symptoms of current cough, fever, weight loss or night sweats may have TB and should be evaluated for TB and other diseases.

(strong recommendation, moderate-certainty evidence)

Adult and adolescent inpatients with HIV in medical wards where the TB prevalence is >10% should be tested systematically for TB disease with a molecular WHO-recommended rapid diagnostic test.

(strong recommendation, moderate certainty of evidence for test accuracy)

Source: Consolidated guidelines on tuberculosis. Module 2: screening – systematic screening for tuberculosis disease (28).

Diagnosing TB disease

WHO currently recommends several categories of technologies that may be used to diagnose TB disease. The most commonly available tests in many settings are near point-of-care molecular assays, also known as molecular WHO-recommended rapid diagnostic tests.

Molecular assays

Both laboratory-based and near point-of-care molecular assays are available or being developed that can support TB diagnosis. mWRDs are considered the most sensitive diagnostic tests for detecting TB disease (29). Assays may require sample preparation, and most are PCR amplification and detection tests in a single self-enclosed unit, in the form of a cartridge. Generally, the assays are automated once the cartridge containing the sample is inserted into the device. This enables specimens to be collected away from a laboratory; however, the device itself often requires a stable power supply, temperature control and annual recalibration of its modules. It is important to note that test sensitivity has been found to reduce along with CD4 cell count, and people with advanced HIV disease may have difficulties in producing sputum, thus requiring other specimens to be used where indicated(29). These operational considerations often require that the device be present in a laboratory environment, which can lead to delays in results (not always processed and provided on the same day of sample collection or clinic visit).

Table 2 summarizes the target population, sample types eligible for testing and whether information on rifampicin drug resistance is also available for numerous available near point-of-care molecular TB assays. Complete information on the diagnostic accuracy of these tests is available (29). In addition, some near point-of-care and laboratory-based molecular assays can test for numerous diseases (such as TB, HIV, hepatitis and/or human papillomavirus infection) and therefore present an opportunity for diagnostic integration across disease areas.

Table 2. Summary of molecular assays for diagnosing TB disease

Diagnostic test	Pulmonary TB sample	Extrapulmonary TB sample	Rifampicin resistance	Isoniazid resistance
Xpert® MTB/RIF (Cepheid, Sunnyvale, CA, USA); Xpert® MTB/RIF Ultra (Cepheid, Sunnyvale, CA, USA)	Sputum, gastric aspirate, nasopharyngeal aspirates	Meningitis: cerebrospinal fluid Lymphadenopathy: lymph node aspirate, lymph node biopsy, blood, pleural fluid or peritoneal fluid or pericardial fluid or synovial fluid or urine	Yes	No
Truenat® MTB, MTB Plus and MTB-RIF Dx tests (Molbio Diagnostics, Goa, India)	Sputum	–	Yes	No
TB-LAMP (Eiken Chemical, Tokyo, Japan)	Sputum	–	No	Yes
Moderate complexity automated NAATs RealTime MTB Assay (Abbott Laboratories, Abbott Park, USA), the BD MAX™ multidrug-resistant TB assay (Becton, Dickinson and Company, Franklin Lakes, NJ, USA), the Hain FluoroType® MTBDR assay (Bruker/Hain Lifescience, Nehren, Germany) and the Roche COBAS® MTB and MTB-RIF/INH assays (F. Hoffmann–La Roche, Basle, Switzerland)	Sputum	–	Yes	No
Urine LF-LAM Abbott Determine™ TB LAM Ag (Abbott Laboratories, Abbott Park, USA) For people living with HIV only (adults, adolescents and children) with signs and symptoms or advanced HIV disease or low CD4 count	Not for use on pulmonary samples	Urine	No	No

The choice of diagnostic test depends on the prevailing national policy for TB diagnostics. The key diagnostic tools summarized here are those recommended by WHO. Source: Consolidated guidelines on HIV prevention, testing, treatment, service delivery and monitoring: recommendations for a public health approach, 2021 update (5).

LF-LAM

LF-LAM is an immunocapture assay that detects lipoarabinomannan in urine, which is a polysaccharide present on the cell wall of the TB bacteria that is released from metabolically active or degenerating TB bacterial cells (29).

LF-LAM remains the only currently available true point-of-care rapid diagnostic test for TB disease and can produce results rapidly. It is used as a rule-in test for TB for people with advanced HIV disease to facilitate rapid linkage to treatment (29,30). In most settings, test results may be available within 25 minutes, and the test does not require any special equipment or reagents; the basic components needed are urine collection cups, blotting paper, micropipettes with tips and a timer (31).

Table 3. Diagnostic accuracy of LF-LAM for diagnosing adults with advanced HIV disease for TB, regardless of signs and symptoms of TB

CD4 count		Inpatient	Outpatient	All settings*
<200 cells/mm ³	Sensitivity	64% (35–87%)	21% (8–48%)	26% (9–56%)
	Specificity	82% (67–93%)	96% (89–99%)	96% (87–98%)
<100 cells/mm ³	Sensitivity	57% (33–79%)	40% (20–64%)	47% (30–64%)
	Specificity	90% (69–97%)	87% (68–94%)	90% (77–96%)

*Additionally, the presence of signs and symptoms, CD4 cell count have an effect on sensitivity and specificity of the test. Source: Consolidated guidelines on tuberculosis. Module 3: Diagnosis – rapid diagnostics for tuberculosis detection (29).

In addition, LF-LAM can be used for individuals who might be incapable of producing sputum. It is suggested that everyone living with advanced HIV disease receive an LF-LAM test to support rapid TB diagnosis. The current WHO recommendations on using LF-LAM are included in this policy brief (Table 3).

LF-LAM may thus have additional advantages over near point-of-care molecular assays that could include testing in the absence of laboratory facilities, good turnaround times for reporting results and opportunities for task-sharing (29).

WHO's operational handbook offers several algorithmic approaches for testing for TB disease (30). The core clinical algorithm for individuals with advanced HIV disease is systematic screening (WHO four-symptom screening), urine LF-LAM followed by a near point-of-care molecular assay (if available) to determine possible resistance or diagnose TB if LF-LAM is negative.

Table 4. WHO recommendations for using LF-LAM for people living with HIV

Population	Clinical action
Inpatient settings	WHO strongly recommends using LF-LAM to assist in the diagnosis of active TB in HIV-positive adults, adolescents and children: <ol style="list-style-type: none"> 1.1. with signs and symptoms of TB (pulmonary and/or extrapulmonary) (strong recommendation, moderate certainty in the evidence about the intervention effects); or 1.2. with advanced HIV disease or who are seriously ill (strong recommendation, moderate certainty in the evidence about the intervention effects); or 1.3. irrespective of signs and symptoms of TB and with a CD4 cell count of less than 200 cells/mm³ (strong recommendation, moderate certainty in the evidence about the intervention effects).
Outpatient settings	2. WHO suggests using LF-LAM to assist in the diagnosis of active TB in HIV-positive adults, adolescents and children: <ol style="list-style-type: none"> 2.1. with signs and symptoms of TB (pulmonary and/or extrapulmonary) or seriously ill (conditional recommendation, low certainty in the evidence about test accuracy); and 2.2. irrespective of signs and symptoms of TB and with a CD4 cell count of less than 100 cells/mm³ (conditional recommendation, very low certainty in the evidence about test accuracy).
	3. WHO recommends against using LF-LAM to assist in the diagnosis of active TB in HIV-positive adults, adolescents and children: <ol style="list-style-type: none"> 3.1. without assessing TB symptoms (strong recommendation, very low certainty in the evidence about test accuracy); 3.2. without TB symptoms and unknown CD4 cell count or without TB symptoms and CD4 cell count greater than or equal to 200 cells/mm³ (strong recommendation, very low certainty in the evidence about test accuracy); and 3.3. without TB symptoms and with a CD4 cell count of 100–200 cells/mm³ (conditional recommendation, very-low-certainty evidence for test accuracy).

Source: Consolidated guidelines on tuberculosis. Module 3: Diagnosis – rapid diagnostics for tuberculosis detection (29).

Numerous molecular WHO-recommended rapid diagnostic tests are currently available on the market and may be used to diagnose TB disease following a WHO-recommended four-symptom systematic screening. Incorporating systematic screening, molecular tests and LF-LAM into the advanced HIV disease package of care is a key step in reducing the number of people dying from HIV-related causes.

Rapidly identifying TB is a critical programmatic activity among people living with HIV to help link to appropriate care or deliver preventive treatment for those without TB disease (5). Ensuring equitable and widespread global access

to LF-LAM and mWRDs is a key step in improving care for individuals with advanced HIV disease, since these people suffer disproportionately from TB-related mortality.

Cryptococcal disease

Cryptococcus species are single-celled fungi that have a polysaccharide (complex sugar) capsule containing glucuronoxylomannan, which is the target for cryptococcal antigen testing (32).

WHO provides recommendations for screening and diagnosis for cryptococcal disease, introduced in 2018 and updated in 2022. These recommendations primarily target adults and adolescents; routine screening of children is not recommended since cryptococcal disease is very rare among children (4).

For children, appropriate differential diagnosis is required for illness consistent with central nervous system disease to rule out more common causes of meningitis.

WHO also recommends that, where possible, cryptococcal disease should be confirmed following a positive antigen screening test by obtaining a CSF sample with a lumbar puncture and tested with either a cryptococcal antigen assay, CSF culture or India ink if the former is unavailable (4,33). Lumbar puncture and CSF examination are not required to initiate pre-emptive treatment in settings where lumbar puncture is not feasible and should not result in delays in initiating treatment, especially when clinical suspicion is high.

Box 3. Overarching principles and recommendations for screening for cryptococcal disease

Overarching principle

Screening for plasma, serum or whole blood cryptococcal antigen is the optimal approach for guiding resources in a public health approach and is the preferred approach for identifying infection when managing people 10 years and older presenting with advanced HIV disease.

Recommendations

Screening^a for cryptococcal antigen followed by pre-emptive antifungal therapy among cryptococcal antigen-positive people to prevent the development of invasive cryptococcal disease is recommended before initiating or reinitiating antiretroviral therapy (ART) for adults and adolescents living with HIV who have a CD4 cell count <100 cells/mm³.

(strong recommendation, moderate-certainty evidence)

This may be considered at a higher CD4 cell count threshold of <200 cells/mm³.

(conditional recommendation, moderate-certainty evidence)

All people living with HIV with a positive cryptococcal antigen screening should be carefully evaluated for signs and symptoms of meningitis and undergo lumbar puncture, if feasible, with CSF examination and India ink or CSF cryptococcal antigen assay to exclude meningitis. India ink has low sensitivity, and a negative result on India ink should be confirmed by CSF cryptococcal antigen testing or CSF culture.

When cryptococcal antigen screening is not available, fluconazole primary prophylaxis should be given to adults and adolescents living with HIV who have a CD4 cell count <100 cells/mm³.

(strong recommendation, moderate-certainty evidence)

This may be considered at a higher CD4 cell count threshold of <200 cells/mm³.

(conditional recommendation, moderate-certainty evidence)

^aAll people living with HIV with a positive cryptococcal antigen result on screening should be carefully evaluated for signs and symptoms of meningitis and undergo a lumbar puncture if feasible with CSF examination and cryptococcal antigen assay (or India ink if cryptococcal antigen assay is not available) to exclude meningitis.

Source: Guidelines for diagnosing, preventing, and managing cryptococcal disease among adults, adolescents and children living with HIV (52).

Cryptococcal antigen testing

Cryptococcal antigen testing may use various specimens – whole blood, serum plasma and CSF. Tests are mainly qualitative; semiquantitative tests are also available, and their clinical utility is a subject of ongoing research. A significant benefit of the cryptococcal antigen lateral flow assay tests are their high sensitivity and specificity and device-free operability.

Box 3 summarizes the WHO principles and recommendations for screening for cryptococcal disease.

A systematic review from 2021 estimated that the prevalence of serum cryptococcal antigen among 3600 adults living with HIV and clinically suspected of having cryptococcal disease presenting with central nervous system symptoms (such as neck pain and rigidity or altered mental status) was 63% (95% confidence interval 45–81%) (34).

Screening for cryptococcal disease requires a blood-based cryptococcal antigen test along with identifying the signs and symptoms of cryptococcal disease.

Blood-based screening enables individuals with cryptococcal antigenaemia to be identified and offered timely pre-emptive treatment. In addition, it helps rule individuals out for cryptococcal disease and reduce polypharmacy for individuals with advanced HIV disease. Evidence of the diagnostic accuracy and clinical utility of these tests is growing (11, 35–37).

Cryptococcosis may be diagnosed in several ways, including microscopy, histopathological approaches from tissue samples, fungal cultures and detecting circulating cryptococcal antigens in various body fluids, primarily in CSF. WHO recommends using CSF-based cryptococcal antigen testing for diagnosing cryptococcal disease.

Since early diagnosis is key to reducing morbidity and mortality from cryptococcal disease (37), countries should give priority to reliable access to rapid diagnostic assays, preferably lateral flow assays for use in whole blood, serum, plasma or CSF, especially since several devices are WHO prequalified (38). Several cryptococcal antigen tests are available at relatively low cost (39), and these should be feasible to implement in most settings. Cryptococcal antigen lateral flow assay screening has been demonstrated to be highly cost-effective through mathematical modelling (40,41).

The threshold of CD4 cell count for cryptococcal antigen testing as part of WHO guidelines on cryptococcal disease supported the need for screening individuals for cryptococcal disease at thresholds of <100 cells/mm³ but also encourages screening for disease at a CD4 threshold of <200 cells/mm³ (42). This enables streamlining with the advanced HIV disease package of care with a consistent CD4 threshold of 200 cells/mm³ for identifying cryptococcal disease and triggering screening, treatment and/or prophylaxis.

The diagnostic accuracy of several cryptococcal antigen tests was systematically assessed for the 2018 WHO guidelines for managing cryptococcal disease. These included CSF latex fluid agglutination, enzyme immunoassays and lateral flow assays. The results of the systematic review are published in the guidelines (33). All cryptococcal antigen assays exhibit good and comparable accuracy and performance and can support countries in implementing robust, accurate, and inexpensive options for rapid cryptococcal diagnosis with sensitivity and specificity typically being >90% across all antigen testing technologies (Box 4).

A review from 2021 found no evidence of any difference between these tests, with both types of specimens approaching very high sensitivity and specificity of ideal test characteristics (34). Using plasma specimens for cryptococcal antigen testing is a viable option in settings that have ready access and trained health-care workers to operate a centrifuge device. Serum and whole-blood specimens, however, may be more feasible in a broader range of settings, especially decentralized settings that require rapid results to support more rapid clinical decision-making and may not have access to laboratory settings for plasma separation.

Microscopy and microbiological culture

Microscopy of culture samples from CSF on Sabouraud dextrose agar is considered the gold standard for diagnosing cryptococcal disease (43). Fungal colony growth is characterized by the formation of opaque white- or cream-coloured colonies; the opacity of the colonies results from the encapsulated nature of cryptococcus. Quantitative cultures are both a measure of disease severity and independently associated with clinical outcomes. These quantitative cultures may be used to measure the clearance of cryptococcus, but there is currently a lack of standardization of approaches to measure clearance (44). Culture of CSF samples has been used in a variety of settings where antigen-based tests and India ink are not available. However, cultures have limited applicability since they can be resource intensive and more expensive than antigen-based tests for diagnosing cryptococcal infection.

India ink preparation

For the past several decades, the standard method for identifying cryptococcal disease was using CSF specimens with an India ink preparation. This reveals characteristic white refractile fungal cells with a halo or clear zone of India ink pigment around the cells against a dark background on microscopy (45). However, this technique requires experienced laboratory technicians and appropriate laboratory support for consistent reporting. The India ink preparation remains a useful diagnostic tool in settings that do not have access to newer, easier antigen-based technologies; however, the India ink test in CSF specimens is less sensitive than CSF culture (46).

Finally, histopathology is a resource intensive and challenging approach requiring tissue biopsy and a variety of reagents to identify fungal elements in each sample and thus not always feasible for wide-scale use in programmatic settings.

Importance of lumbar puncture

Performing lumbar puncture is a critical skill for diagnosing cryptococcal disease in programmatic settings. Lumbar puncture is a relatively straightforward procedure but requires appropriate training on the indications, contraindications, risks and management of complications. A study assessing the safety of performing lumbar puncture in a rural hospital in central Africa found no procedure-attributable deaths or nervous system sequelae when local guidelines were followed; the most serious post-lumbar puncture adverse events were typically self-limiting (transient confusion, headache and backache) (47). Adequate counselling is a vital component of a successful lumbar puncture procedure; lumbar puncture refusal has been documented, with some study sites reporting as high as 25% refusals or deferrals (48,49).

Cryptococcal antigen screening is an integral part of the WHO advanced HIV disease package of care and is a key tool for reducing mortality. Individuals identified as having advanced HIV disease – either entering or re-entering care – should be screened for cryptococcal disease and offered prophylaxis or treatment in accordance with the WHO advanced HIV disease algorithm (4). However, access to cryptococcal antigen testing is relatively limited in low- and middle-income countries with a high burden of HIV infection (50,51).

Routine screening of children is not recommended, since cryptococcal disease is very rare among children (4); appropriate differential diagnosis is required for illness consistent with central nervous system disease to rule out more common causes of meningitis.

WHO recommendations for induction therapy for cryptococcal disease were updated in 2022, recommending a single, high dose of liposomal amphotericin in combination with flucytosine and fluconazole (52). The introduction of these recommendations relies on the ability to rapidly identify individuals with cryptococcal disease – thus reiterating the need for scaling up of cryptococcal screening and diagnosis in programmes globally.

Box 4. Country implementation examples

A feasibility study in rural Lesotho showed promising results when supervised lay counsellors delivered cryptococcal antigen tests (72). In Malawi, the national HIV programme includes a cadre of HIV diagnostic assistants who develop key diagnostic tests, including rapid CD4, LAM and cryptococcal antigen testing for people with advanced HIV disease. This is currently being evaluated.

Reflex screening for cryptococcal disease has been introduced successfully into national policy in South Africa, where reflex cryptococcal antigen screening is performed on remnant CD4 samples and may save programme costs compared with provider-initiated screening. The scaling up of reflex screening also included the development of a dashboard with two new indicators to track progress (73,74).

¹ Induction therapy refers to part of a three-stage management approach (induction, consolidation and maintenance) for treatment for cryptococcal meningitis.

Histoplasmosis

Histoplasmosis is a chronic disease caused by the fungus *Histoplasma capsulatum*. It is an important coinfection for individuals with advanced HIV disease, frequently manifesting as a disseminated infection. It is considered endemic in some regions of North, Central and South America and in some countries in Asia and Africa. Most reports of histoplasmosis are from the Region of the Americas, annually reporting up to 15 600 new cases and 4500 deaths among people living with HIV. The low reported rates in Africa probably result from underreporting from a lack of appropriate diagnostics (53).

Clinical misidentification, misclassification and underdiagnosis of histoplasmosis are common and have important consequences for national HIV programmes. Histoplasmosis is often clinically misdiagnosed as TB, and individuals with histoplasmosis therefore often do not receive appropriate treatment or might be untreated altogether, thus contributing to increased mortality (54). An autopsy study among people with HIV in Brazil found that histoplasmosis was a major contributor to 22% of deaths (55). Challenges in timely identification are further compounded by significant interregional variation in disease prevalence (54).

WHO recommends using an antigen test for diagnosing histoplasmosis. Histoplasmosis has traditionally been diagnosed with conventional laboratory approaches, such as culture and histopathology with microscopy, but these approaches have important limitations since histology requires invasive biopsy and fungal cultures take up to six weeks for identification.

Antibody-based tests are less sensitive for individuals who are immunocompromised and have little or no utility for diagnosis for people living with HIV. Antigen tests, available as both an enzyme immunoassay and a lateral flow assay, have been a key approach for diagnosis, especially from urine specimens. Antigen tests have been used systematically for people with advanced HIV disease in Paraguay (56), Guatemala (57), Colombia (58), Honduras, Panama and Nicaragua (59) and Brazil (60), with prevalence estimates of 7–10% when testing all individuals with CD4 <200 cells/mm³, 20% when testing symptomatic individuals (59,60) or 29% (58) among people living with HIV admitted to the hospital. Up to 10% of the histoplasmosis cases also have coinfection with active TB, which complicates management because of drug-drug interactions with itraconazole.

WHO and PAHO published recommendations for diagnosis and management of histoplasmosis for people living with HIV in 2020 (61), and these recommendations were subsequently integrated into the 2021 WHO consolidated guidelines on HIV (5). Algorithms that include appropriate histoplasmosis tests to avoid misclassifying individuals with histoplasmosis with other diseases in poorly resourced regions are an important need.

Other important coinfections

Individuals with advanced HIV disease risk developing a wide range of opportunistic infections, and timely identification is critical. WHO recently launched the fungal priority pathogens list, which also includes fungal species that primarily affect people living with HIV (62). Of the 19 fungal pathogens within the WHO fungal priority pathogens list, four opportunistic pathogens in particular cause invasive diseases in people living with HIV: *Cryptococcus neoformans*, *Histoplasma* spp., *Pneumocystis jirovecii* and *Talaromyces marneffeii*.

People with advanced HIV disease are at risk of illness and death from a wide range of infectious diseases. *Candida* spp., *Paracoccidioides* spp., *Coccidioides* spp. and *Aspergillus fumigatus* most commonly cause severe disease for other populations, but they are also important opportunistic pathogens for people living with HIV (63). Severe disease is common among people living with HIV caused by most of the pathogens on the fungal priority pathogens list.

Severe bacterial infections are another important cause of mortality for people living with HIV; WHO provides up-to-date clinical and programmatic considerations (64).

The advanced HIV disease package of care recommended by WHO is suggested as the minimum required considerations for all countries and settings with a high burden of HIV infection. Each country should further assess any additional coinfections that are endemic and adapt the package of care to ensure that critical diagnostics, treatment and prevention tools are made available for the diseases afflicting people with advanced HIV disease.

Talaromycosis

Talaromycosis is an invasive fungal infection that has been increasingly reported in South-East Asia among individuals with advanced HIV disease. A seasonal association has been reported, especially with the tropical monsoons. The disease is associated with an elevated mortality rate, with up to one third of individuals diagnosed with talaromycosis dying (65–67).

Despite the association with higher mortality, relatively little is known about its prevalence in the general population. Countries such as China, Thailand and Viet Nam report talaromycosis as a leading cause of HIV-related deaths. It also disproportionately affects poorer or rural areas of affected countries. It is also often difficult to identify, regularly mimicking other infections (66,67).

Diagnosis may be made with fungal culture, but this approach is associated with substantial delays, leading to delays in initiating treatment. PCR-based assays have a high reported specificity but are not well suited for screening approaches because of lower sensitivity (68). Several promising antigen tests for talaromycosis are undergoing clinical validation (69–71). Point-of-care antigen tests have the potential to enable screening for early asymptomatic disease and pre-emptive therapy to prevent fulminant talaromycosis and reduce the number of people dying from HIV-related causes in South-East Asia

There remain clear gaps in the diagnosis of talaromycosis as well as access to diagnostics and treatments. Talaromycosis has been included in the WHO fungal priority pathogens list and is considered a medium-level priority, primarily because of its regional predilection (62). Effective treatments are available, but access is very limited and they are expensive. There also remains a need for improved surveillance of this disease among people living with HIV, especially those with advanced HIV disease.

Conclusions

There remains a strong rationale to scale up the screening and diagnosis of advanced HIV disease and the key opportunistic infections. Linkage to effective treatments relies heavily on timely recognition of disease. WHO recommends both task-sharing considerations and an integrated approach to diagnostics (4). This will promote optimal use of resources while covering key infections causing mortality not just in advanced HIV disease but among all people living with HIV. Training health care workers with point-of-care technologies may offer potential task-sharing opportunities, which in turn may contribute to lower costs. An important conclusion is that for populations with a high burden of advanced HIV disease, the WHO-recommended package needs to be fully implemented to maximize health benefits while also being highly cost-effective across a wide range, as shown by recent modelling work (75).

WHO will continue to closely follow emerging interventions related to advanced HIV disease and will develop guidelines or guidance where appropriate to support and enable countries to further scale up services for advanced HIV disease.

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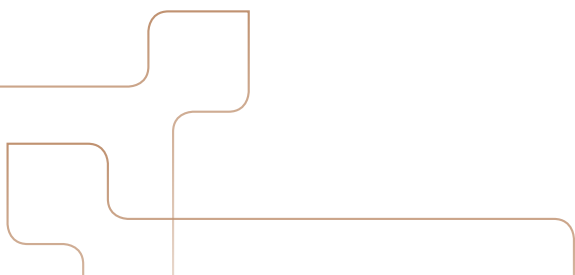
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