Assessing the burden of tuberculosis among pregnant and postpartum women:

Implications for integrating tuberculosis screening into routine antenatal care in Guinea-Bissau

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Summary

This study aims to address critical diagnostic and data gaps in tuberculosis (TB) care among pregnant and postpartum women in Guinea-Bissau, a high-burden, resource-limited setting. Recognising that current TB screening during antenatal care (ANC) relies largely on unstructured symptom questions, the study will integrate more systematic and innovative approaches into routine maternal health services.

The project will implement the Bandim TBscore II as a structured triage tool to classify symptom severity and guide referrals. To strengthen diagnostic capacity, two novel tools will be evaluated: stool-based GeneXpert testing (a method recommended in children and explored here as a feasible alternative for pregnant women) and artificial intelligence-powered chest X-ray interpretation software designed to enhance TB detection where radiological expertise is lacking.

The study will also generate comprehensive, population-based data on TB and TB infection (TBI) among pregnant, postpartum women and women of reproductive age in Guinea-Bissau. The results are intended to inform health policy, both locally and in high-income countries, by providing evidence to improve TB screening protocols and care for this vulnerable group. Ultimately, the study seeks to develop scalable strategies that can be replicated across low- and middle-income countries to advance maternal and child health and support global TB eradication efforts.

1. Background

Tuberculosis (TB) remains a major global health challenge, with an estimated 10.8 million new cases and 1.25 million deaths in 2023, making it the leading cause of death from a single infectious agent worldwide (1). The burden is highest in low- and middle-income countries (LMICs), where access to diagnostics and treatment is limited (2). Sub-Saharan Africa accounts for about 23% of global TB cases and 31% of TB-related deaths (3). Among the affected populations, women of reproductive age (WRA), especially those who are pregnant or postpartum, represent a critically neglected group in TB eradication efforts.

Pregnant and postpartum women are at increased risk of developing active TB and experiencing adverse outcomes (4-6), yet TB in this population remains under-diagnosed and under-researched. While an estimated 200,000 pregnant or postpartum women develop active TB annually (7), prevalence estimates vary widely and are rarely based on systematic data. In

Mozambique, TB prevalence was reported at 505 cases per 100,000 pregnant women, increasing to 1,626 cases per 100,000 among those with HIV co-infection, while in South Africa, 2.5% of HIV-positive pregnant women were diagnosed with active TB (8, 9). Immunological and physiological changes during pregnancy and postpartum, combined with increased nutritional demands, heighten vulnerability to TB (4, 5) and increase the risk of severe maternal and neonatal outcomes including maternal death, stillbirth, preterm birth, and neonatal mortality (10-15). These risks are further compounded in TB-HIV high double-burden settings (16), where diagnostic delays can be fatal.

Despite this, conventional TB screening approaches are largely inadequate for detecting tuberculosis during pregnancy, leaving a critical diagnostic gap. Standard diagnostics, such as GeneXpert sputum testing, require quality sputum samples that are often difficult to obtain in pregnant women (17). Symptom-based screening alone is unreliable due to symptom masking by pregnancy-related physiological changes (18-20). Overburdened healthcare systems and limited access to radiological expertise further hinder timely diagnosis and treatment, contributing to a major gap in the TB care cascade. Innovative, feasible diagnostic approaches are urgently needed to overcome these constraints.

The lack of robust data on TB and TBI prevalence among pregnant and postpartum women severely hampers global progress towards the WHO End TB Strategy (21) and maternal and child health goals. Data on TB in pregnancy is highly uncertain, fragmented, and rarely integrated into policy and antenatal care guidelines. These evidence gaps hinder understanding of the full scope of the problem and obstruct the development of effective interventions, especially in LIMCs.

Guinea-Bissau, one of the 30 TB-HIV double-burden countries, reports a TB incidence of 361 per 100,000 population (22) and a case detection rate of only 35% (23), indicating critical diagnostic gaps. TB is the leading infectious cause of death among WRA in the country, accounting for 14% of all deaths in this group (24). Yet, there are no reliable estimates of the burden of TB or TBI among pregnant or postpartum women. Current TB screening during ANC consists solely of informal symptom questions (e.g., “do you cough”), an approach insufficient due to symptom masking during pregnancy (25) and potentially limited health-seeking among symptomatic women. More systematic, active TB screening during pregnancy is urgently needed.

In this project, we will address key diagnostic, and evidence gaps by integrating optimised TB screening protocols and novel diagnostics into routine ANC in Guinea-Bissau, an example for a high-burden, resource-constrained setting. The Bandim TBscore II (26), previously validated in Guinea-Bissau (27), offers a more structured triage tool and will be used in this project to guide referrals based on symptom severity classification. To improve diagnostic capacity, this project will evaluate two innovative tools adapted for low-resource settings. First, we will assess the use of stool-based GeneXpert testing, currently recommended for children (28, 29), as a feasible alternative for pregnant women. Second, we will implement artificial intelligence-powered (AI) chest X-ray interpretation software (Qure.ai, India) (30), which has shown promise in improving TB detection where radiology expertise is limited (31, 32).

Moreover, in this project, we will also generate contextualised, population-based data on TB and TBI burden among pregnant, postpartum women and WRA in Guinea-Bissau. Drawing from the findings of this project, we seek to inform health policy and aid the design of replicable strategies for TB screening and care in LMICs.

Furthermore, findings from this project will inform health policy in high income countries such as Denmark, regarding screening of pregnant women from LMICs, as it currently is done in Sweden (15).

2. Objectives

2.1. Primary objective

To evaluate the effectiveness of integrating a TB screening protocol and novel diagnostic procedures into routine ANC for improvement of timely detection of TB in pregnant women, to assess active TB disease prevalence among women of reproductive age (WRA) and pregnant women, and to assess TBI prevalence among pregnant women in Guinea-Bissau.

2.2. Secondary objectives

a. To assess the burden of active TB among WRA and pregnant women;
b. To assess the prevalence of TBI among pregnant women;
c. To assess the incidence of developing active TB among pregnant and postpartum women with and without HIV infection;

d. To assess the impact of integrating active TB screening and innovative diagnostic methods into routine ANC on timely detection and treatment of TB in pregnant and postpartum women.

2.3. Research question

Does integrating a standardized TB screening protocol and novel diagnostic procedures into routine antenatal care improve timely detection and increase detection rates of active tuberculosis among pregnant and postpartum women in Guinea-Bissau?

2.4. Hypothesis

Employing an integrated TB screening protocol and novel diagnostic methods will improve timely diagnosis and double detection rates of active TB among pregnant and postpartum women.

3. Methods

3.1. Setting and epidemiology

The project will be conducted in Bissau, the capital of Guinea-Bissau. Here, the Bandim Health Project (BHP) (33) has been running a Health and Demographic Surveillance System (HDSS) since 1978, covering over 100,000 persons in six suburban neighbourhoods. Since 1996, the HDSS has recorded all facility-detected TB cases in the study area, with ongoing long-term follow-up in BHP's TB cohort. For this purpose, the BHP has built up a close collaboration with the three primary healthcare facilities located in the study area. Based on this framework, BHP has conducted large population-based studies on TB epidemiology, interventions and diagnostics (34, 35).

A complete population census is conducted approximately every three years. Each month, all households with women of reproductive age are visited to identify new pregnancies. Pregnant women are referred to local health centres for antenatal care, where they are followed monthly from the second trimester onwards. All newborns are registered, and mothers are interviewed about pregnancy, delivery, and household conditions. Children are then followed up every three months until the age of three. At routine visits, information on nutritional status, vaccinations, morbidity, and hospitalisations is collected. In addition, BHP records routine vaccinations and monitors TB and HIV consultations and treatments at three health centres within the study area. BHP also registers all births at the maternity ward and all paediatric hospitalisations at the national Hospital Simão Mendes, located two kilometres outside the study area.

3.2. Design

The project comprises three interconnected studies designed to evaluate innovative screening approaches and to generate comprehensive evidence on TB among WRA, pregnant, and postpartum women:

a. Study 1: Assess the burden of active TB among WRA and pregnant women to establish baseline prevalence and trends.
b. Study 2: Estimate the prevalence of TBI and the incidence of active TB among pregnant and postpartum women.
c. Study 3: Evaluate the integration of systematic TB screening and innovative diagnostic methods into routine ANC to improve timely case detection.

Study 1: Assessing the burden of active TB among WRA and pregnant women: Based on already collected data from the BHP's HDSS and its TB cohort, we will assess the burden of active TB among WRA and pregnant women from 2003 to 2025. The HDSS provides a continuously updated population registry, which will be linked to clinical TB diagnosis data through individual identifiers, allowing us to determine both TB cases and population at risk for rate calculations. We estimate that approximately 90,000 women aged 15 to 49 years will be included in the analysis. Using descriptive statistics and logistic regression models, we will examine trends in TB incidence over time and assess associations with background factors such as age, socioeconomic status, HIV status, and parity. Analyses will account for time trends and clustering at the community and household levels to improve precision and address potential confounding. The results will be contrasted to the findings of WP3.

Study 2: Assessing TBI prevalence and incidence of active TB development among pregnant women: In a cross-sectional investigation nested within the larger framework of WP3, we will estimate the prevalence of TBI among pregnant women through QuantiFERON-TB testing. Blood samples (4 mL of blood – 1 mL per QFT-Plus Blood Collection Tube) (36) will be collected from all pregnant women, attending their first ANC at the three collaborating health centres, for 8 months. These samples will then be analysed at LNSP by trained laboratory technicians, ensuring accurate results that contribute valuable epidemiological data on TBI rates. Data analysis will include descriptive statistics to summarise prevalence and logistic regression models to explore associations between TBI and background factors such as age, HIV status, and nutritional status. Further, all women will be followed through the pregnancy

and 6 months into the postpartum period to observe for possible development of active TB disease.

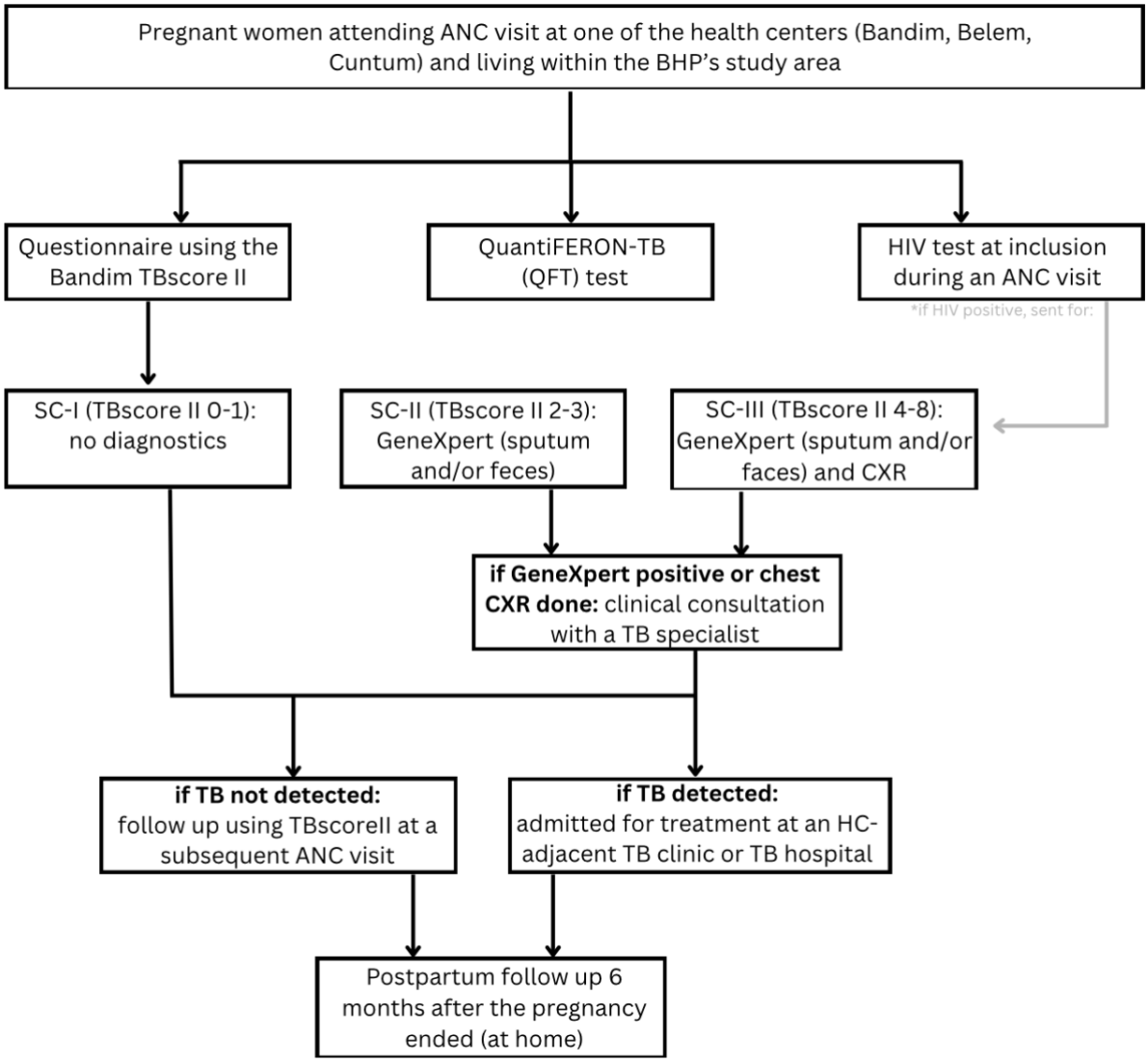
Study 3: Integrating systematic TB screening and innovative diagnostic methods into routine ANC: In a stepped-wedge cluster randomised trial, we will evaluate the impact of integrating systematic TB screening and innovative diagnostic methods into routine ANC. Standard care (baseline) will be compared with an enhanced protocol (intervention).

At baseline, the women will be assessed for TB symptoms using the Bandim TBscore II and referred for GeneXpert sputum testing at the decision of midwifes, alongside routine HIV testing. At intervention, pregnant women will be interviewed regarding TB exposure and previous TB, and systematically screened with the Bandim TBscore II (26), which categorises patients based on the severity of their symptoms. Women exhibiting moderate symptoms (TBscoreII=2-3) suggestive of TB will be referred for GeneXpert testing sputum and/or faecal samples, while individuals with more severe symptoms (TBscoreII=4-8) will additionally receive chest radiography, which will be analysed with AI software (Qure.ai, India) (30). All HIV-positive women will be tested immediately with both GeneXpert and Xray. Women with positive GeneXpert results will be referred for treatment, while women with Xray will attend a consultation with a TB specialist. Participants will be reassessed for TB symptoms at each subsequent ANC visit and at 6 months postpartum.

The primary endpoint is diagnostic yield. Based on an assumed TB prevalence of 0.22% among pregnant women, an anticipated doubling of yield through enhanced screening protocols, and an estimated 8 inclusions per cluster per week, 720 women will be recruited over 32 weeks (>90% power), with 60 women per cluster per time interval (8 weeks).

Detection rates will be compared between baseline and intervention using a generalised linear mixed effects model accounting for time and intra-cluster correlation. Repeated measures model will be used to analyse patient characteristics. The sample size was estimated using the “steppedwegde” function in STATA (37, 38).

Figure 1. Flowchart for Study 2 and Study 3 (intervention).



3.2. Enrolment

3.2.1. Inclusion criteria

Study 1: Pregnant women in the study area between 2003-2025. Data retrieved from the individual-level longitudinal data from BHP's HDSS and TB cohort. in the study area between 2003-2025.

Study 2 and 3: Pregnant women ≥ 15 years of age from the study area attending ANC consultations at three health centres (HCs): Bandim, Belem, and Cuntum.

3.2.2. Exclusion criteria

Study 1: Pregnant women ≤ 15 years of age.

Study 2 and 3: Pregnant women who are unable to provide informed consent, reside outside of the study area, are ≤ 15 years of age, and/or receive TB treatment at the time of, or a year prior to, enrollment.

4. Sample size and statistical analysis

Study 1: The analysis will include approximately 90,000 women of reproductive age recorded in the HDSS between 2003 and 2025. The large sample size ensures sufficient power to estimate TB incidence rates and assess time trends. Descriptive statistics will be used to summarise TB cases, and logistic regression models will evaluate associations with age, socioeconomic status, HIV status, and parity. Analyses will account for clustering at the household and community level to improve precision and reduce bias.

Study 2: We expect to enrol all pregnant women attending ANC at the three collaborating health centres over an 8-month period, estimated at approximately 720 women. QuantiFERON results will provide prevalence estimates of latent TB infection with narrow confidence intervals. Logistic regression will be used to assess associations with background characteristics, while follow-up data will allow calculation of incidence rates of active TB. Analyses will adjust for potential confounders such as HIV status and nutritional status.

Study 3: This study is designed as a stepped-wedge trial. We aim to recruit 720 pregnant women over 8 months, corresponding to an average of 8 inclusions per week per cluster. With an assumed TB prevalence of 0.22% and an expected doubling of diagnostic yield in the intervention arm, the study is powered at >90% to detect meaningful differences between groups. Detection rates will be compared between baseline and intervention using a generalised linear mixed effects model accounting for time and intra-cluster correlation. Repeated measures model will be used to analyse patient characteristics. The sample size was estimated using the “steppedwegde” function in STATA (37, 38).

5. Significance and potential impact

5.1. Major advances

The project introduces a major advance by integrating active TB screening protocols and novel diagnostic tools directly into routine ANC. This approach will, for the first time, provide reliable prevalence and incidence estimates of TB and TBI among pregnant and postpartum women in Guinea-Bissau. Such data are currently lacking, undermining the ability to monitor

progress toward the WHO's End TB Strategy (21) and maternal and child health goals. By targeting a neglected high-risk population, the project has the potential to reduce preventable maternal deaths, stillbirths, and neonatal mortality through earlier detection and treatment of active TB diesese.

5.2. New approach

Unlike previous TB studies that focused on the general population or HIV-positive individuals, this project pioneers a maternal health-centered approach. Active screening and innovative diagnostics, such as GeneXpert fecal testing and use of AI, will be embedded within ANC settings, ensuring feasibility and sustainability in low-resource contexts. The strategy leverages existing maternal health services, minimizing additional infrastructure needs, while simultaneously introducing improved diagnostic capacity that can be scaled across similar high-burden, low-income settings worldwide.

5.3. Generalisability of trial results

The results of this project will be directly applicable to other TB-endemic, resource-limited countries where maternal health services form one of the most consistent points of healthcare contact for women. If effective, the integration of TB screening into ANC could be adopted and endorsed by the WHO and national governments, forming a replicable model for maternal TB care globally. The project's emphasis on sustainable, context-specific approaches enhances its generalizability across diverse health system settings.

5.4. Contribution to improved disease management and public health

By focusing on maternal populations, the project addresses a critical blind spot in global TB control. Undiagnosed TB in pregnancy not only threatens maternal survival but also drives poor neonatal outcomes and intergenerational cycles of ill health. Earlier identification and treatment of TB in this group can substantially reduce mortality and morbidity, while also preventing onward transmission. This directly contributes to the dual agendas of ending TB and reducing maternal and child mortality in high-burden countries, thereby strengthening health equity and alleviating poverty-related disease burdens.

5.5. Improvements in patient care

For the first time, pregnant and postpartum women in Guinea-Bissau will benefit from systematic TB screening during routine care, ensuring that high-risk individuals are identified and linked to treatment without delay. Embedding TB screening into ANC promotes a patient-centered approach that enhances trust in maternal health services and improves continuity of

care. The use of innovative diagnostic tools tailored to maternal populations further increases the accuracy of case detection, ensuring that women and their newborns receive timely and appropriate management. In this way, the project improves both clinical outcomes and the overall quality of maternal healthcare delivery.

6. Timeframe

Study 1 will rely on retrospective data analysis and is expected to be completed within the first 12 months. Study 2 will run concurrently with Study 3, with recruitment and sample collection over an 8-month period. Study 3 will also include ANC follow up and follow-up at six months postpartum, concluding to a 20-month period. After concluding the data collection period, we will proceed with data cleaning, advanced analyses, dissemination of findings, and manuscript preparation. All studies are set to be finalised by July 2028.

Table 1 – Timeframe for preparations.

	2025												2026										
	J	F	M	A	M	J	J	A	S	O	N	D	J	F	M	A	J	J	A	S	O	N	D
Preparations																							
Protocol writing																							
Funding applications																							
Database and forms																							
Ethics approval																							
Procurement																							
Workshop and training																							

Table 2 – Timeframe for working on-site.

	2026												2027												2028											
	J	F	M	A	M	J	J	A	S	O	N	D	J	F	M	A	J	J	A	S	O	N	D	J	F	M	A	J	J	A	S	O	N	D
Study 1																																				
Merging of datasets																																				
Data cleaning																																				
Data analysis																																				
Manuscript writing																																				
Dissemination																																				
Study 2																																				
Data collection																																				
Data analysis																																				
Manuscript writing																																				
Dissemination																																				
Study 3																																				
Patient enrollment																																				
ANC follow-up																																				
6-month postpartum follow-up																																				
Data analysis																																				
Manuscript writing																																				
Dissemination																																				
Data dissemination (local and regional)																																				
Policy meetings																																				

7. Ethical considerations

The Ethical Committee of Guinea-Bissau has approved the study on 9 December 2025, with a decision number 044/CNES/INASA/2025. Individual informed consent has been previously

obtained for HDSS and TB cohort data. Anonymity of participants will be preserved throughout all data analyses and the dissemination of findings.

8. Reporting and dissemination

This project is distinguished by its innovative approach to integrating active TB screening protocols and novel diagnostic tools into routine ANC and generating critical epidemiological data on active TB burden, prevalence, and incidence among pregnant and postpartum women. Thereby, this research aligns with the WHO's End TB Strategy (21) by fostering sustainable healthcare models tailored to the needs of high-risk maternal populations and contributes to the global efforts to end TB and advance maternal health equity. We plan to publish at least three peer-reviewed studies in international journals. The findings will also be shared at both local and international conferences and directly communicated to local health authorities through presentations and policy briefs, thereby ensuring that findings contribute directly to public health strategies both in low-, middle- and high-income settings.

9. Study group

Primary investigator: Anita Magdalena Zalisz, MSc, PhD Student, Department of Clinical Medicine, Aarhus University, and Bandim Health Project.

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