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Utility, acceptability and applicability of a nucleic acid amplification test in comparison with a syndromic approach in the management of sexually transmitted diseases at Mulago National Referral Hospital in Uganda (ASTRHA): protocol for an open-label, randomised controlled trial

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ABSTRACT

Introduction Sexually transmitted diseases (STDs) are a major cause of long-term disability. Urethral discharge syndrome (UDS), abnormal vaginal discharge (AVD) and genital ulcer disease (GUD) are very common in low-income and middle-income countries (LMICs), where, due to lack of resources, these infections are managed according to a syndromic approach. Although microbiological diagnosis using nuclear acid amplification tests (NAAT) is already a standard to prescribe targeted treatments in industrialised countries, no randomised clinical trials have been conducted to evaluate clinical usefulness and acceptability of NAAT in comparison with syndromic approach in LMICs. The results of this study could inform diagnostic guidelines since they may suggest an update of the current recommendation if microbiological diagnosis using NAAT in the management of STD is demonstrated to be both useful and acceptable in an LMIC context.

Methods and analysis The primary objective of this randomised, open-label trial is to evaluate the clinical usefulness of a NAAT and its acceptability in comparison with a clinical syndromic approach and to explore whether this test could replace the syndromic approach in the management of STDs at a national referral hospital in Uganda. 220 patients presenting to the STD clinic at Mulago Hospital in Kampala, Uganda with AVD, UDS or GUD will be randomised to either standard of care (syndromic management) or NAAT-based treatment with a 1:1 ratio. All the patients will be asked to return after 2 or 3 weeks for a control visit. Primary outcome will be therapeutic appropriateness.

Ethics and dissemination This trial was approved by the Mulago Hospital Research and Ethical Committee (MHREC2023-97) and the Uganda National Council for Science and Technology (HS31000ES). Patients will give

STRENGTHS AND LIMITATION OF THIS STUDY

⇒ This randomised clinical trial will assess whether nuclear acid amplification test-based sexually transmitted disease management will improve not only therapeutic appropriateness, but also clinical outcome and microbiological cure as patients will be re-evaluated after 2–3 weeks.
⇒ The study will assess the acceptability of the molecular test in a low-income and middle-income country setting by calculation of the percentage of patients retained into the study with the prescribed treatment and through a questionnaire.
⇒ The study will be conducted in a national referral hospital, so the results cannot be immediately transferred to the whole country.
⇒ The primary endpoint is not clinical cure, in order to avoid the study being underpowered; the chosen primary endpoint (appropriateness of therapy) is however relevant both for individual patients and in terms of public health (eg, with respect to reduction of inappropriate antibiotic use, which may promote the emergence and the dissemination of drug-resistant strains).
⇒ The study will not assess acceptability through psychometrically assessed new measures such as the acceptability of intervention measure, but this can be evaluated in future studies if our study generates positive results.

informed consent to participate before taking part in the study. Results will be published in peer-reviewed journals in open-access formats and data made available in anonymised form.

Trial registration number NCT05994495.

INTRODUCTION

Sexually transmitted diseases (STDs) are a major cause of long-term disability, including infertility, ectopic pregnancy and premature birth. In Uganda, prevalence of STDs increased from 22% in 2006 to 27% in 2011 and up to 1.5 million cases were reported between 2015 and 2017.[1] The high prevalence of STDs and of associated adverse health outcomes make STD control a public health priority, especially in most-at-risk populations. STDs also facilitate the sexual transmission of HIV. Recently, the WHO guidelines recommended the introduction of microbiological diagnosis based on the results of quality-assured molecular assays to manage STDs.[2] However, in settings with limited or no molecular tests or laboratory capacity, WHO recommends syndromic treatment without or with limited laboratory testing (eg, direct examination) to ensure treatment on the same day of the visit. For lack of adequate resources, and for the timing needed to develop and implement the new rapid unexpensive desk top diagnostic tools, syndromic management is still widely used to manage STDs in many low-income and middle-income countries (LMICs), including Uganda. On one hand, this type of approach has many advantages in LMICs, but on the other hand it does not allow an exact diagnosis to be determined and a targeted therapy to be prescribed.

There is currently paucity of data regarding the use of microbiological diagnosis using nucleic acid amplification techniques (NAAT) as the standard of care for the management of STDs in resource-limited setting, particularly in terms of clinical usefulness, appropriateness of therapy and acceptability. No randomised trials have been conducted so far to evaluate clinical usefulness and acceptability of microbiological diagnosis using molecular test in comparison with syndromic approach without or with limited laboratory tests.

The global STDs strategy, endorsed by the World Health Assembly in 2016 aims to end STDs as a public health threat by 2030.[3] Thus, appropriate STDs diagnosis and treatment is crucial to prevent the transmission and sequelae of untreated or overtreated infections.

WHO has identified *Neisseria gonorrhoeae* (NG) as a high-priority pathogen because of widespread antimicrobial resistance to penicillin, tetracyclines, macrolides (including azithromycin), sulphonamides, trimethoprim and quinolones, including emergent resistance to the 'last line' extended-spectrum cephalosporins, cefixime and ceftriaxone. The emergence of decreased susceptibility of NG to extended-spectrum cephalosporins together with already-existing resistance to other antibiotics makes NG a multidrug-resistant organism.[4]

The emerging NG-resistant strains to the current recommended first-line and second-line antibiotics in Uganda[5 6] may be partially attributable to the syndromic approach being used in the management of STDs at all health facility levels including the National Referral and Teaching Hospital. Syndromic evaluation for the primary approach to guide treatment of STDs can lead to undertreatment or inadequate treatment or even overtreatment of patients with consequences for both the individual patient and the entire community with the possible emergence of drug resistance strains (not only in NG but also in other pathogens responsible for STDs such as *Mycoplasma genitalium*[7]), increasing costs and potentially avoidable toxicity. If nothing is done to control this serious problem, it may render health systems helpless with a biosecurity risk to the entire world. Furthermore, the syndromic approach does not permit determination of the prevalence of individual aetiologies in a population. The primary objective of this trial is to evaluate the clinical usefulness of a NAAT in terms of appropriateness of therapy, clinical and microbiological outcomes, diagnostic accuracy and acceptability in comparison with clinical syndromic approach and to explore whether this test could replace the syndromic approach in the management of STDs at a national referral hospital in Uganda. Primary and secondary objectives of the trial are summarised in table 1.

Table 1 Primary and secondary objectives

Primary objective	► To evaluate whether microbiological diagnosis using NAAT significantly improves the appropriateness of therapy of STDs in comparison with the syndromic approach without or with limited laboratory tests currently in use.
Secondary objectives	► To evaluate whether the use of NAAT for the management of STDs improves clinical outcome and microbiological cure compared with the syndromic approach. ► To assess the degree of concordance between actual pathogens diagnosed through the molecular test and the presumed pathogens indicated by the syndromic approach. ► To evaluate the acceptability of the NAAT. ► To estimate the actual relative prevalence of causative agents of STDs in our study setting.

NAAT, nucleic acid amplification test; STD, sexually transmitted disease.

As reported by WHO, new diagnostics are needed to help guide diagnosis and treatment decisions to foster antibiotic stewardship of existing and new antibiotics. WHO reported that if syndromic evaluation remains the primary approach to guide treatment of STDs, there is a significant risk of misdiagnosis and antibiotic overuse, which has been shown to lead to antimicrobial resistance.[4]

New molecular tests could enable a rapid and accurate diagnosis that would overcome these problems.

The turnaround time for the molecular test in study is much shorter (210 min) compared with standard laboratory investigations that can be used in the syndromic approach (eg, culture). Since the test in question can provide results for mixed infections, it is an advantage to the other laboratory test which provides for only single causative organisms yet patients mostly present with mixed infections; besides this, the test will contribute to delay the emergence of drug-resistant strains and reduce on waste of drugs due to unnecessary treatment. It is imperative, therefore, for a national referral and teaching hospital to put in place a relatively improved syndromic approach in the management of STDs to avoid delay of patient treatment during microbiological results processing.

We speculate that the change in approach would allow a significant improvement in terms of appropriateness of therapy, reduction of the collateral damage (emergence and transmission of drug resistances), toxicity and pharmacoeconomic costs.

Results of our study could inform diagnostic guidelines since they may suggest an update of the current recommendation if the test-and-treat approach using the multiplex PCR technique is demonstrated to be both useful and acceptable in an LMIC.

Finally, this study could open further areas of research; if this study reveals poor acceptability of NAAT by patients because they are unable to wait for the time needed to get the result and receive the appropriate treatment, a study could be conducted in the future to assess the feasibility and acceptability of prescribing the treatment in local settings with the help of pharmacists.

METHODS AND ANALYSIS

Design

This is an operational, randomised, open-label trial to assess the clinical usefulness, acceptability and clinical outcomes of an aetiological approach using NAAT versus a clinical syndromic approach for the management of STDs. Participants will be recruited from the STD clinic of Mulago National Referral Hospital in Kampala, Uganda which also houses the National STD Reference laboratory/control unit of Ministry of Health. The STD clinic is located at upper Mulago, in Kawempe Division, Kampala City Authority in Uganda. At Mulago STD clinic, about 20–30 adult patients are seen every day—Monday to Friday. The syndromes considered in the study will be urethral discharge syndrome (UDS) in males, abnormal vaginal discharge (AVD) and genital ulcer disease (GUD) in both genders. The pathogens responsible for each of these syndromes are shown in table 2. According to Ugandan clinical guidelines, the standard of care for the management of STDs is based on a syndromic clinical approach that may include some laboratory investigations, when needed, but not NAAT that is the focus of our study.[8] The treatment regimens were formulated following the current National STD Management Guidelines 2016 and Centre for Disease Control and Prevention STI guideline, 2021.

Table 2 Syndromes considered in the study and their respective possible aetiological agents[8]

Syndromes	Pathogens	
UDS	► *Neisseria gonorrhoeae* ► *Chlamydia trachomatis* ► *Ureaplasma urealyticum* ► *Trichomonas vaginalis*	
AVD	**Vaginitis** ► *Candida albicans* ► *Trichomonas vaginalis* ► Bacterial vaginosis	**Cervicitis** ► *Neisseria gonorrhoeae* ► *Chlamydia trachomatis*
GUD	► *Treponema pallidum*: syphilis ► *Herpes simplex virus*: genital herpes ► *Haemophilus ducreyi*: Chancroid ► *Klebsiella granulomatis*: Granuloma inguinale ► *Chlamydia* strains: lymphogranuloma venereum	

AVD, abnormal vaginal discharge; GUD, genital ulcer disease; UDS, urethral discharge syndrome.

The protocol manuscript is reported following the guidance of the Standard Protocol Items Recommendations for Interventional Trials 2013 explanation and elaboration: guidance for protocols of clinical trial.[9]

The study was approved by the Mulago Hospital Research and Ethical Committee (MHREC2023-97) and Ugandan National Council of Science and Technology (HS31000ES) and is registered with ClinicalTrials.gov (NCT05994495).

Participants and eligibility criteria

The trial will enrol adults aged 18 years and above presenting with signs and symptoms of UDS, AVD and GUD at the Mulago Hospital STDs clinic who fulfil the inclusion criteria and provide written consent to participate.

We will exclude patients who reside farther than a 20-km radius from Mulago National Referral Hospital, to ensure patients do not miss follow-up visits, patients with other syndromes than those under study (eg, anorectal discharge), pregnant women and those having menstrual periods at presentation to the clinic. The inclusion and exclusion criteria are listed as follows.

Inclusion criteria

Adult males and females with UDS, AVD and GUD diagnosed as per the current National STD Management Guidelines 2016,[8] who have given informed, written and signed consent.

Exclusion criteria

- All patients living farther than a 20-km radius from Mulago National Referral Hospital.
- All patients presenting with any syndromes not listed above.
- Female patients in their menstrual period.
- Pregnant patients.

Sample size

A sample size of 90 patients in each arm will be enrolled to demonstrate by means of an exact Fisher's test carried out at a significance level of 0.05 (two sided) with a power of 0.80, a difference of 0.20 between the 2 treatment groups (experimental group and control group), with an expected proportion of appropriateness of therapy equal to 0.75. The sample size will be increased to 110 patients in each treatment arm to cater for a drop-out rate of about 20%.

After a comprehensive literature review, we reasonably assume that appropriateness of therapy prescribed through a syndromic approach will be around 75%. However, this estimate may be heterogeneous for intrinsic limitations of the available studies.

A study published in 2019 evaluated a novel multiplex real-time PCR method and it concluded that this method has high sensitivity (91%–100%) and specificity (99%–100%), relatively low cost, and simplicity of use for the simultaneous detection of 9 STD pathogens in genitourinary secretions.[10]

There are currently no randomised controlled trials comparing the syndromic approach and the use of molecular testing for the management of STDs. Indeed, most studies are observational, heterogeneous in nature and data available in Uganda are outdated.[11]

Based on the available data, it appears very difficult to define the diagnostic accuracy of the syndromic approach and thus the resulting therapeutic appropriateness. In fact, diagnostic accuracy varies according to the pathogen responsible and its prevalence, the syndrome analysed, the presence or absence of symptoms and the type of algorithm used (eg, with or without laboratory assistance) for the management of STDs.[12]

The syndromic approach seems to work well for UDS, with sensitivity ranging from 84% to 95% and specificity ranging from 7% to 90%, implying that patients could be overtreated.[12–14] Syndromic management for vaginal discharge has severe limitations. Systematic reviews and meta-analyses of the syndromic approach to diagnose and treat cervical infections revealed low accuracy, resulting in a high proportion of overtreatment, incorrect treatment and missed treatment as concluded by Wi *et al*.[12] Sensitivity and specificity of syndromic management to diagnose vaginal infection (*Trichomonas vaginalis* and bacterial vaginosis) vary according to the different flow charts. Sensitivity ranges from 53% to 91% and specificity ranges from 53% to 100% depending on whether speculum examination or laboratory investigations are used. In the syndromic approach of cervical infections due to *N. gonorrhoeae* and *Chlamydia trachomatis*, sensitivity ranges from 23% to 90% and specificity ranges from 35% to 74% according to the different flowcharts.[12 15]

Genital HSV infection is the predominant cause of GUD that affects the outcome of syndromic management of GUD.[16] A study conducted in eight STD clinics in India reported the overall sensitivity and specificity of the syndromic algorithm for genital ulcer syndrome to be 68% and 52%, respectively, and the positive predictive value 50%. The sensitivity of the algorithm for herpetic GUD was better than that of non-herpetic GUD (74% vs 33%), whereas specificity was lower (33% vs 56%).[12 14 17]

Recruitment and randomisation

A community engagement plan was created to inform patients about STDs and the importance of reducing their spread, and a formal information session is organised prior to providing informed consent. Patients who present with UDS, AVD or GUD and are eligible will be adequately informed about the study by members of the research team in English and the local language (Luganda) and asked to participate by signing the informed consent form (online supplemental material file 1).

Random allocation stratified by gender according to a complete block scheme will be used to assign participants to the experimental group (arm A) or control group (arm B) and the research team will treat patients according to the randomisation code written in a sheet contained in progressively numbered and sealed opaque envelopes. Participants will be randomised into arm A and arm B with a 1:1 ratio through a randomisation list made by a computer program (SAS V.14.2).

Intervention

Patients with UDS, AVD or GUD randomised to arm A will undergo a NAAT. After having obtained the result of the molecular test, patients will be prescribed a targeted treatment. Patients randomised to arm B will undergo a NAAT, but they will be treated according to the current guidelines and the best practice using the clinical syndromic approach.[8] So, patients randomised to arm B and their physician also will be blinded of the results of the NAAT.

Sample collection will be performed in the STD clinic of the Mulago National Referral Hospital in Kampala. The patient will be placed in a comfortable position in a private room. Samples will be collected by research assistants properly trained in the preanalytical procedures described in the protocol, and they will take care to avoid any contaminations of the sample. The samples will be delivered immediately to the microbiology laboratory, taking advantage of its proximity to the clinic. All the patients randomised to arm A or to arm B will be asked to return after 2 weeks (or three if the prescribed therapy has a longer duration) for a control visit. Patients will be advised to have their partners take the same therapy and to abstain from sexual intercourse until they and their sex partners have been adequately treated and any

Table 3 Pathogen list, clinical sensitivity (urine/swab) and specificity (urine/swab) (%)

Pathogens	Sensitivity	Specificity
Herpes simplex virus-1	100/96.5	100/99.5
Herpes simplex virus-2	100/92.2	100/99.7
Chlamydia trachomatis	83.3/86.5	100/100
Haemophilus ducreyi	100/100	100/100
Mycoplasma genitalium	86.7/81.3	99.5/100
Mycoplasma hominis	93.8/91.5	100/100
Neisseria gonorrhoeae	100/93.7	100/100
Treponema pallidum	90/87.5	100/100
Ureaplasma urealyticum	90.9/88.4	100/99.1
Trichomonas vaginalis	100/90.6	100/100

symptoms have resolved. Patients will be asked to answer a short questionnaire regarding their experience with trial procedures (online supplemental material file 2). This questionnaire will help to assess the acceptability of NAAT from a qualitative point of view.

Microbiological methods

The NAAT will be performed with *Bosch Vivalytic STI test*. It is a qualitative PCR-based assay for simultaneous detection of 10 common sexually transmitted pathogens (table 3).[18]

The analytical phase will be performed on *Vivalytic* analyser that is a portable, fully automated device for molecular diagnostics, able to perform PCR procedures. Molecular tests will be performed on urine sample or genitourinary swab. The samples will be processed as soon as possible to allow the test-and-treat approach to be performed for patients included in arm A; test turnaround time is 150 min. In any case, even for patients included in arm B, sample processing will be performed in the same way. Laboratory staff will be adequately trained in the use of the *Vivalytic* analyser device and analytical procedures. The type of sample collected for each syndrome is summarised in table 4.

Table 4 Syndromes and type of sample collected for each syndrome

Syndromes	Sample
UDS	► First catch urine or ► Urethral swab (if first catch urine collection is not possible or based on researcher's assessment)
AVD	► Vaginal swab or ► First catch urine (if vaginal swab is not possible)
GUD	► Genital ulcer swab ► Urethral/vaginal swab (if genital ulcer swab is not possible)

AVD, abnormal vaginal discharge; GUD, genital ulcer disease; UDS, urethral discharge syndrome.

Outcomes

Primary research outcome

Appropriateness of therapy

Primary research outcome will be therapeutic appropriateness defined as (either as study intervention during consultation in arm A or *post-hoc* in arm B) the use of a recommended drug or drug combinations which are recommended against the pathogen(s) diagnosed by the molecular test according to the current National STD Management Guidelines 2016[8] and Centre for Disease Control and Prevention sexually transmitted infections guideline, 2021.[19] The primary endpoint is not the clinical or microbiological cure to avoid the study being unpowered because it is difficult to foresee the proportion of patients who will be missed to the follow-up (ie, not presenting for the test of cure). However, the chosen primary endpoint is relevant both for individual patient and in terms of Global Health (optimisation of antimicrobial stewardship and containment of antimicrobial resistances spreading in the population).

Secondary research outcomes

1. Percentage of patients who will achieve success at the test of cure performed after 2–3 weeks from the end of therapy in both arms. For the clinical outcome, we will consider the percentage of patients who will recover from signs and symptoms of the STDs in both arms at the same time-point.
2. Percentage of concordant results between the syndromic approach and the NAAT. The diagnosis is considered concordant when at least one pathogen responsible for a specific syndrome (table 1) diagnosed through the syndromic approach is detected by molecular testing. The diagnosis is considered not concordant when one or more pathogens responsible for syndromes other than those identified by the syndromic approach are detected by molecular testing.
3. Percentage of patients who will be sent home the same day with the treatment prescribed according to the molecular test result (arm 'A') or with the treatment according to the syndromic approach (arm 'B'). Patients will not be able to wait for the result of the test and for the targeted therapy will be considered as failure for the primary endpoint. In addition, patients who will drop out from the study will be considered as failure.
4. Prevalence of detected pathogens' genome at the molecular test (overall population).

Data collection, monitoring and management

The collected data will be entered into a database specifically created for this study to be able to export data tables and managed by the principal investigators and coinvestigators.

The study consists of two stages shown in figure 1.

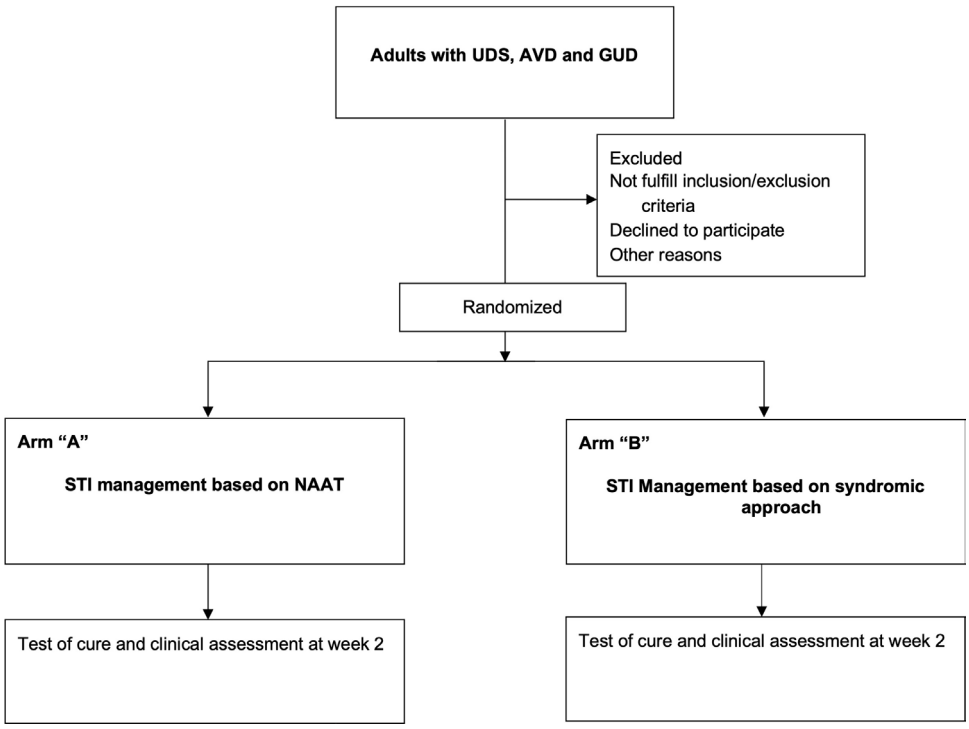

Figure 1 Study flow chart. AVD, abnormal vaginal discharge; GUD, genital ulcer disease; NAAT, nucleic acid amplification techniques; UDS, urethral discharge syndrome.

First stage: patients with symptoms of UDS, AVD and GUD coming to the STD clinic at Mulago Hospital will be evaluated according to inclusion/exclusion criteria; if eligible and after signing informed consent, they will be randomised into arm A or B. Secondarily, according to randomisation, they will be treated according to an aetiological approach or a syndromic approach. During this first stage, the enrolment form and the first part of the case record form (CRF) will be completed by the research team members. Finally, patients randomised into both two arms will be asked to return for a follow-up visit 2–3 weeks after the first visit.

Second stage: during follow-up visit, microbiological and clinical cure will be assessed; so, patients will be clinically re-examinate and undergo a second NAAT. In this second phase, the last part of CRF will be completed and patients will then be referred to normal follow-up.

The following data will be collected: demographics, HIV status, type of syndrome (UDS, AVD and GUD), symptoms and signs, type of sample collected, result of molecular test, treatment prescribed (both to patients and to the sexual partner), result of speculum examination (if performed), any other laboratory investigations, clinical outcome, result of test of cure and adherence to the treatment prescribed. Data will be collected in an anonymised form (an ID number will be assigned to each patient) by filling in the CRF in which all the variables object of our study have been reported. CRF will be stored assuring protection of the privacy of the patients.

Confidentiality

According to Uganda Data Protection and Privacy Act (2019), documentation, data and all other information generated will be held in strict confidence. The name and any other identifying information will not be included in any electronic data. No information concerning the study, or the data will be released to any unauthorised third party, without prior approval of the principal investigators, except as necessary for monitoring by the Mulago Hospital Research Ethics Committee and Uganda National Council for Science and Technology (UNCST) who can assess the data or records during the inspections and audit of records.

Statistical analysis

Descriptive statistics will be calculated for quantitative variables (mean, SD, median and IQR together minimum and maximum) and for qualitative variables (absolute and per cent frequencies). 95% CI will be calculated for the main clinically relevant variables. The two groups will be compared by means of Fisher's exact test for qualitative variables (primary and secondary endpoints) and by means of Student's t-test or its non-parametric counterpart Mann-Whitney test in the case of quantitative variable with no or skewed sampling distribution. The agreement between the syndromic approach and the NAAT will be tested by means of Cohen's kappa statistic weighted or not weighted on the number of the categorical classes.

Multivariable methods (logistic regression or linear regression in the case of a dependent variable qualitative or quantitative, respectively) will be carried out

for considering the role of some baseline variables as confounders.

The statistical analysis will be carried out according with the intention-to-treat principle. A sensitive analysis will be performed on a per-protocol population (patients without major protocol deviations to be defined in an ad hoc meeting before database freezing).

Timelines of the study and expected results

Enrolment began in January 2024. The first results of the study will be reported in the second half of 2024. The results of this research will assess the feasibility of implementing a larger scale intervention. In addition, this initial study will serve as a pioneering investigation of the impact of using NAAT in STD management in LMICs. If the results confirm the beneficial effects on outcomes, this investigation will help provide further evidence to support the effectiveness of NAAT use even in LMICs. The trial will assess the acceptability of NAAT in an LMIC by calculation of the percentage of patients retained into the study with the prescribed treatment and through a questionnaire although will not assess acceptability through psychometrically assessed new measures,[20] but this can be evaluated in future studies if our study will provide positive results. This study will serve as a basis for extrapolating to more centres, thus being able to carry it out in larger samples, which will allow us to be able to assess the utility and acceptability of NAAT not only in Uganda but also in other LMICs.

This research project originated within the ERASMUS+KA107 'International Credit Mobility' project[21] through the University of Catanzaro which provided the support to the fellow who took part to the mutual exchange programme between this University and the University of Makerere, Kampala, Uganda.[22]

Ancillary studies

1. In patients without symptom improvement or microbiological cure after receiving therapy, we will propose a study to determine the prevalence of antibiotic-resistant infections due to NG and *M. genitalium*. For NG, we will test for drug resistance genes using rapid methods and assess concordance with phenotypic resistance results.
2. Pharmacoeconomic evaluation of the use of NAAT in the management of STDs. With this objective in mind, external advisors with skills in pharmacoeconomic will be part of the research team.

Patient and public involvement

None.

ETHICS AND DISSEMINATION

This project will adhere to the guidelines outlined in the Declaration of Helsinki (64th World Medical Association General Assembly, Fortaleza, Brazil, October 2013) and the Standards of Good Clinical Practice (ICH E6). Only researchers involved in the project will be permitted to access the research data. Each subject's information will be linked with a unique numerical identification code and will be the sole means of identifying the patient for the purposes of data processing and analysis. This trial was approved by the Mulago Hospital Research and Ethical Committee (MHREC2023-97) and the UNCST (HS31000ES).

All subjects participating in the study will provide signed informed consent prior to their inclusion. To do so, participants will be asked to read and sign the patient information sheet and consent form. They will also be informed of their right to withdraw their consent at any time without having to provide a reason and without any consequences.

The results of this study will be published in scientific peer-reviewed journals and presented in both the academic and public domain, including at scientific conferences and in the media in public engagement forums. Patient confidentiality will be maintained in all the above. All protocol documents are available in Uganda on reasonable requests at the STD clinic, Mulago National Referral Hospital, Kampala.

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Contributors CT designed the study, wrote the manuscript and helped with methodological and statistical design. RS and PM designed the study and wrote the manuscript. BMC wrote the manuscript and helped with methodological and statistical design. VO, HLM wrote the manuscript. PB-K wrote the manuscript and made suggestions and helped in the experimental design. GGM, MC, LSB, PK and EMT made suggestions and helped in the experimental design. FM made suggestions and helped in the experimental design and microbiological methods. All authors have read and approved the manuscript.

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Competing interests None declared.

Patient and public involvement Patients and/or the public were not involved in the design, or conduct, or reporting, or dissemination plans of this research.

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terminology, drug names and drug dosages), and is not responsible for any error and/or omissions arising from translation and adaptation or otherwise.

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REFERENCES

1 Masanja V, Wafula ST, Ssekamatte T, *et al*. Trends and correlates of sexually transmitted infections among sexually active Ugandan female youths: evidence from three demographic and health surveys, 2006-2016. *BMC Infect Dis* 2021;21:59.
2 WHO. n.d. Guidelines for the management of symptomatic sexually transmitted infections. Available: https://www.who.int/publications/i/item/9789240024168
3 WHO. n.d. Sexually transmitted infections (Stis). Available: https://www.who.int/news-room/fact-sheets/detail/sexually-transmitted-infections-(stis)
4 WHO. n.d. Target product profiles for improved antimicrobial stewardship for Gonococcal infection. Available: https://www.who.int/news/item/01-09-2019-target-product-profiles-for-improved-antimicrobial-stewardship-for-gonococcal-infection
5 Workneh M, Hamill MM, Kakooza F, *et al*. Antimicrobial resistance of Neisseria Gonorrhoeae in a newly implemented surveillance program in Uganda: surveillance report. *JMIR Public Health Surveill* 2020;6:e17009.
6 Kakooza F, Musinguzi P, Workneh M, *et al*. Implementation of a standardised and quality-assured enhanced Gonococcal antimicrobial surveillance programme in accordance with WHO protocols in Kampala, Uganda. *Sex Transm Infect* 2021;97:312–6.
7 Melendez JH, Hardick J, Onzia A, *et al*. Retrospective analysis of Ugandan men with Urethritis reveals Mycoplasma Genitalium and associated macrolide resistance. *Microbiol Spectr* 2022;10:e02304-21.
8 Ministry of Health Knowledge Management Portal. n.d. Publications. Available: http://library.health.go.ug/publications/guidelines/uganda-clinical-guidelines-2016
9 Chan A-W, Tetzlaff JM, Gøtzsche PC, *et al*. SPIRIT 2013 explanation and elaboration: guidance for protocols of clinical trials. *BMJ* 2013;346:e7586.
10 Hu X-M, Xu J-X, Jiang L-X, *et al*. Design and evaluation of a novel Multiplex real-time PCR melting curve assay for the simultaneous detection of nine sexually transmitted disease pathogens in Genitourinary secretions. *Front Cell Infect Microbiol* 2019;9:382.
11 Tann CJ, Mpairwe H, Morison L, *et al*. Lack of effectiveness of Syndromic management in targeting vaginal infections in pregnancy in Entebbe, Uganda. *Sex Transm Infect* 2006;82:285–9.
12 Wi TE, Ndowa FJ, Ferreyra C, *et al*. Diagnosing sexually transmitted infections in resource-constrained settings: challenges and ways forward. *J Int AIDS Soc* 2019;22 Suppl 6:e25343.
13 Pettifor A, Walsh J, Wilkins V, *et al*. How effective is Syndromic management of Stds?: A review of current studies. *Sex Transm Dis* 2000;27:371–85.
14 Gupta V, Sharma VK. Syndromic management of sexually transmitted infections: A critical appraisal and the road ahead. *Natl Med J India* 2019;32:147–52.
15 Zemouri C, Wi TE, Kiarie J, *et al*. The performance of the vaginal discharge Syndromic management in treating vaginal and Cervical infection: A systematic review and meta-analysis. *PLoS One* 2016;11:e0163365.
16 Makasa M, Buve A, Sandøy IF. Etiologic pattern of genital ulcers in Lusaka, Zambia: has Chancroid been eliminated *Sex Transm Dis* 2012;39:787–91.
17 Prabhakar P, Narayanan P, Deshpande GR, *et al*. Genital ulcer disease in India: Etiologies and performance of current syndrome guidelines. *Sex Transm Dis* 2012;39:906–10.
18 BOSCH. n.d. The innovative PCR test for sexually transmitted infection. Available: https://www.bosch-vivalytic.com/en/tests/sti/
19 CDC's sexually transmitted infections (STI) treatment guidelines. 2021 Available: https://www.cdc.gov/std/treatment-guidelines/default.htm
20 Weiner BJ, Lewis CC, Stanick C, *et al*. Psychometric assessment of three newly developed implementation outcome measures. *Implement Sci* 2017;12:108.
21 UMG. n.d. "Università Degli Studi "Magna Graecia" Di Catanzaro". Available: https://web.unicz.it/it/
22 Serraino R, Owachi D, Byekwaso SN, *et al*. From the global north to the global south: preparing students for away rotations. *BMC Med Educ* 2023;23:102.