Title: Phase 2b open label randomized controlled trial to evaluate the adverse drug reaction protective efficacy, safety and tolerability of N-acetylcysteine in combination with second-line in adult people treated for multidrug-resistant tuberculosis in Tanzania. (Trial Acronym NAC Trial).

Names protocol contributors

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Abstract

Trials guidance: The Abstract should not exceed 350 words. Please minimize the use of abbreviations and do not cite references in the abstract. The abstract must include the following separate sections:

- **Background**: the context and purpose of the study
- **Methods**: how the study will be performed
- **Discussion**: a brief summary and potential implications
- **Trial registration**: If your article reports the results of a health care intervention on human participants, it must be registered in an appropriate registry and the registration number and date of registration should be in stated in this section. If it was not registered prospectively (before enrollment of the first participant), you should include the words 'retrospectively registered'. See our editorial policies for more information on trial registration.

Background: Adverse drug reactions (ADRs) frequently occur in patients using second-line anti-tuberculosis medicine for treatment of multidrug resistant tuberculosis (MDR-TB). ADRs contribute to treatment interruptions which can compromise treatment response and risk acquired drug resistance to critical newer drugs such as bedaquiline, while severe ADRs carry considerable morbidity and mortality. N-acetylcysteine (NAC) has shown promise in reducing ADRs for medications related to TB in case series or randomized controlled trials in other medical conditions. We therefore designed a pilot clinical trial to study the protective effect of NAC among people treated for MDR-TB with second-line anti-TB medications.
Methods: This is a phase 2b randomized open label pilot clinical trial with 3 treatment arms including a control arm, an interventional arm of NAC 900mg daily, and an interventional arm of NAC 900mg twice-daily administered during the intensive phase of MDR-TB treatment. Patients initiating MDR-TB treatment will be enrolled at Kibong'oto National Center of Excellence for MDR-TB in the Kilimanjaro region of Tanzania. The minimum anticipated sample size is 66; with 22 participants in each arm. ADR monitoring will be performed at baseline and regular follow-up over 24 weeks including blood and urine specimen collection for hepatic and renal function and electrolyte abnormalities, electrocardiogram and hearing function by pure tone audiometry. Sputum will be collected at baseline and monthly thereafter and cultured for mycobacteria as well as assayed for other molecular targets of *Mycobacterium tuberculosis*. Adverse drug events will be analyzed over time using mixed effect models. Mean differences between arms in change of the ADRs from baseline (with 95% confidence intervals) will be derived from the fitted model.
Discussion: Given that NAC promotes synthesis of glutathione, an intracellular antioxidant that combats the impact of oxidative stress, it may protect against medication induced oxidative damage in organs such as liver, pancreas, kidney and cells of the immune system. This randomized controlled trial will determine if NAC leads to fewer ADRs, and if this protection is dose dependent. Fewer ADRs among patients treated with MDR-TB may significantly improve treatment outcomes for multidrug regimens that necessitate prolonged treatment durations.
Trial registration: PACTR202007736854169

Keywords

Trials guidance: Three to ten keywords representing the main content of the article.

Multidrug resistant tuberculosis, adverse drug reactions, N acetylcyteine, drug Induced liver injury, clinical trial,

Administrative information

Trials guidance: please include this text in your submitted protocol just above the Administrative information table:

Note: the numbers in curly brackets in this protocol refer to SPIRIT checklist item numbers. The order of the items has been modified to group similar items (see http://www.equator-network.org/reporting-guidelines/spirit-2013-statement-defining-standard-protocol-items-for-clinical-trials/).

Title {1}	SPIRIT guidance: Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym.
Trial registration {2a and 2b}.	SPIRIT guidance: Trial identifier and registry name. If not yet registered, name of intended registry. Item 2b is met if the register used for registration collects all items from the World Health Organization Trial Registration Data Set.
Protocol version {3}	SPIRIT guidance: Date and version identifier.
Funding {4}	SPIRIT guidance: Sources and types of financial, material, and other support.
Author details {5a}	SPIRIT guidance: Affiliations of protocol contributors.
Name and contact information for the trial sponsor {5b}	SPIRIT guidance: Name and contact information for the trial sponsor.
Role of sponsor {5c}	SPIRIT guidance: Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities.

Administrative information

Title	Phase 2b double blind randomized controlled trial to evaluate the efficacy, safety and tolerability of N-acetylcysteine in reducing adverse drug reactions among adults treated for multidrug-resistant tuberculosis in Tanzania

Trial registration	https://pactr.samrc.ac.za/Researcher/TrialRegister.aspx?TrialID=12163 (PACTR202007736854169)
Protocol version	Version 3.0
Funding	European Union through the European and Developing Countries Clinical Trial Partnerships 2 Project (TMA2016SF-1463)
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Role of sponsor	This is an investigator-initiated study. The funder has no role in the study design, conduct or writing the report. The corresponding author had full access to the data and final decision to submit for publication.

Introduction

Background and rationale {6a}

SPIRIT guidance: Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention.

The crisis of multidrug-resistant tuberculosis (MDR-TB), defined as point mutations of *Mycobacterium tuberculosis* (MTB) chromosomes resulting in strains that are resistance to isoniazid and rifampicin, key medicines for the treatment of TB, continues to worsen [1]. In 2019, the World Health Organization (WHO) estimated 484,000 cases of MDR-TB, while approximately 156,000 were enrolled to initiate treatment. This marked a 21% increase in enrolment compared to the 2016 report [2]. With the growing epidemic of MDR-TB, further exacerbated by the COVID-19 pandemic [3], and a greater proportion of people accessing treatment, there is further urgency to assure treatment regimens are tolerable and completed without interruption which poses risk to individual outcomes and *M. tuberculosis* strains with amplified drug resistance[4]. Despite the priority for tolerable regimens, medicines used to treat MDR-TB are referred to as "second-line" because of reduced potency and a worse side effect profile [5]. Regimens are constructed of 4-6 drugs based on the susceptibility profile of the infecting *M. tuberculosis* strain, drug-drug interactions or individual contraindications, and supply chain availability [6]. New and repurposed drugs have improved the efficacy and shortened the duration of MDR-TB treatment but those medications have significant and often overlapping toxicities [7]. Based on data from controlled trials and large operational studies, the WHO has recently prioritized bedaquiline, linezolid, newer generation fluoroquinolones (moxifloxacin or levofloxacin), and clofazimine for the treatment of MDR-TB as well as consideration of other new agents such as delamanid and conventional anti-TB drugs such as cycloserine, pyrazinamide, ethionamide/prothionamide and ethambutol to complete a multi-drug regimen [8]. While medications requiring intravenous or intramuscular injection are no longer preferred, there are some uncommon clinical scenarios where TB treatment programs or patients themselves may opt to include an injectable aminoglycoside (amikacin or streptomycin) or a carbapenem with beta-lactamase inhibitor (meropenem/clavulanate or imipenem-cilastin/clavulanate).

No other infectious disease requires treatment strategies of such quantity of drugs, and drug class diversity as MDR-TB, and combined toxicities are not dissimilar to treatment of hematological malignancy. As described elsewhere, the severe adverse drug reactions (ADRs) from medications used to treat MDR-TB include hepatotoxicity, cytopenias, QTc prolongation and cardiac arrhythmia, nephrotoxicity, ototoxicity, peripheral and optic neuropathy, and psychosis [9, 10]. Given the combination of drug-classes, including broad spectrum classes such as the fluoroquinolones, gastrointestinal side effects are common, and can even confuse the presentation of more serious ADRs including hepatoxicity and lactic acidosis [11]. For example, despite the efficacy of bedaquiline, up to 20.8% of people experience bedaquiline related ADRs, with 7.4% serious. ADRs related to bedaquiline were most frequently gastrointestinal (14%), followed by metabolic disorders (8.5%) and nervous system disorders (8.5%)[12]. Likewise, the inclusion of linezolid in MDR-TB regimen has a significant positive effect in improving treatment outcome and reducing mortality, yet the occurrence of ADRs include myelosuppression (33%), neuropathy (30%) whereas other

less common ADRs are vomiting, hyperpigmentation and transient visual impairment[13]. Clofazimine is considered safe since the ADRs requiring discontinuation or withdrawal reported have been as low as 0.1% common ADRs are skin discolouration and gastrointestinal side effects with a pooled proportion of 22%[14]. Furthermore, several drugs have overlapping ADRs for instance the fluoroquinolones, bedaquiline and delamanid all may cause QTc interval prolongation, and hepatotoxicity [15].

We have designed the following trial to study a promising compound, N-acetylcysteine (NAC), for reduction of ADRs during the course of MDR-TB treatment, which is to our knowledge the first of its kind with this primary objective for MDR-TB. NAC is a thiol compound and the acetylated form of L-cysteine with the chemical formula C5H9NO3S and molecular weight of 163.2 g/mol. NAC is efficiently absorbed and metabolized primarily by the liver. Absorption provides a large amount of NAC for cellular uptake, deacetylation of cysteine and synthesis of glutathione (GSH). GSH is an intracellular antioxidant that combats the impact of oxidative stress thus protecting the vital cellular components against the dangerous effect of peroxidation. The free sulfyhydryl group in GSH readily scavenges harmful radicals such as reactive oxygen species, peroxides and superoxides to thiyl radical, which rapidly dimerises to form glutathione disulfide [16, 17]. GHS released from NAC then carriers out its putative protective effect both enzymatically and non-enzymatically. The benefit of NAC on oxidative damage extend to organs such liver, pancreas, kidney, inner ear hair cells, and cells of the immune system. Furthermore, NAC demonstrates anti-inflammatory properties by limiting pro-inflammatory cytokine release, particularly through a NF-kappa beta pathway [18].

NAC has been employed in clinical practice for several decades. It has been used as a mucolytic agent and for the treatment of numerous disorders including paracetamol intoxication, doxorubicin cardiotoxicity, ischemia–reperfusion cardiac injury, acute respiratory distress syndrome, bronchitis, chemotherapy-induced toxicity, heavy metal toxicity and psychiatric disorders[19]. Most relevant to prevention of ADRs from MDR-TB treatment, previous NAC interventions have been trialed in drug susceptible (DS)-TB among primarily Asian populations [20, 21]. Baniasadi et al conducted a clinical trial in an older age ($\geq$ 60 years) population at higher risk of drug induced liver injury (DILI). The hepatoprotective effect of NAC was significant since anti-TB DILI occurred in 12 patients (37.5%) in the control group but none in the NAC group [20]. Farazi et al conducted another NAC trial in 85 patients with age $\geq$ 50 years and treated for DS-TB. Eligible participants were randomly selected to receive NAC 600mg or placebo with standard rifampicin, isoniazid, pyrazinamide and ethambutol (RHZE) treatment. DILI occurred in 14.3% of the placebo group as signified by raised serum aspartate transaminases (AST), alanine transaminases (ALT) and bilirubin. Interestingly, the group receiving NAC also had reduced levels of AST, ALT and bilirubin compared to their baseline values [21]. A further meta-analysis by Kranzer et al conducted a review on the efficacy and safety of NAC in preventing aminoglycoside induced ototoxicity and found the weight of evidence supporting the safety and otoprotective effect of NAC when co-administered with an aminoglycoside, even for durations shorter than those used for MDR-TB [22]. Thus, the prior studies in non-MDR-TB populations and among people with drug-susceptible TB provide a strong justification for a clinical trial to investigate the effect of concomitant NAC treatment in patients receiving MDR-TB treatment. If NAC allows uninterrupted use of the most efficacious dose and duration while limiting the long-term sequela of MDR-TB treatment, then the trial may indeed contribute to the current ambitious goals set by the global health community for achieving a 90% TB treatment success rate by 2035 [23]. Hence conducting this randomized controlled trial within a MDR-TB programme

from a TB endemic country will provide actionable programmatic data with which to determine the eventual role of a NAC intervention.

Objectives {7}

SPIRIT guidance: Specific objectives or hypotheses.

Hypothesis I
Administration of oral NAC at a dose of 900mg daily in combination with second-line during treatment of multidrug-resistant tuberculosis (MDR-TB) will protect against occurrence of serious advent events without interfering with the effect of second line anti-TB regimen.
Hypothesis II
Administration of oral NAC at a dose of 900mg twice a day in combination with second-line during treatment of multidrug resistant tuberculosis (MDR-TB) will protect against occurrence of serious adverse events without interfering with the effect of second line anti-TB regimen
Primary Objective
To assess the development of clinical or laboratory-based adverse drug reactions (ADRs) at any frequency during the first six months of local standard of care MDR-TB treatment among patients receiving NAC at a dose of 900mg daily or 900mg twice a day compared to those patients treated with the standard of care regimen and placebo.

Trial design {8}

SPIRIT guidance: Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory).

NAC trial is a phase 2b randomized double blind superiority trial with three parallel groups and a primary end point of occurrence of ADRs at any time during treatment for MDR-TB.

Methods: Participants, interventions and outcomes

Study setting {9}

SPIRIT guidance: Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained.

Patients will be recruited at one centre, Kibong'oto Infectious Diseases Hospital (KIDH) in Tanzania. The site is experienced in recruitment, hospitalization, safety and efficacy measurement and has the capacity to receive more than 100 MDR-TB patients per year [24]. KIDH has also participated in the International Collaboration for Infectious Diseases Research (ICIDR) consortium to build capacity for MDR-TB trials recruitment and follow-up of a longitudinal cohort of MDR-TB participants through 96 weeks (NCT 03559582). In addition, the KIDH research team has demonstrated the capacity of recruiting DS-TB patients for other trials in the Pan African Consortium for Evaluating Anti-TB Antibiotics (PanACEA) with a remarkably high proportion of retention [25].

Eligibility criteria {10}

SPIRIT guidance: Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study

centres and individuals who will perform the interventions (eg, surgeons, psychotherapists).

MDR-TB participants will be eligible if they are able and willing to provide a written informed consent prior to participation, with age range between 18 and 65 years. They should be newly diagnosed with MDR-TB without a history of using or being on MDR-TB treatment. Likewise, they should be eligible for receiving second line anti-TB medicine and Karnofsky score of ≥ 50 defined as individuals requiring less considerable and frequent medical care. Female participants should not be pregnant as confirmed by urinary pregnant test
Participants that will be excluded includes those with previous existing pathology which will preclude testing such as auditory, or central nervous system (i.e. major head trauma, meningitis, encephalitis, brain metastasis,). Likewise, participants with mental disorder such as schizophrenia, schizoaffective disorder or psychotic disorder will be excluded. Similarly, participants with comorbid conditions such as severe liver or renal diseases will be exclude

Who will take informed consent? {26a}

SPIRIT guidance: Who will obtain informed consent or assent from potential trial participants or authorized surrogates, and how (see Item 32).

Potential participants for recruitment will be individuals admitted patients at KIDH for MDR-TB treatment. Upon identification of a potentially eligible participant, study staff (research nurse or research doctor) will provide information about the study to the participant. As described in greater detail in the study-specific standard operating procedure, the informed consent process will include detailed review of the study informed consent form (ICF), and will allow time to address any questions or concerns each participant may have, and an assessment of each participant's understanding will be performed before proceeding to the informed consent decision. The process will be fully documented and only participants who are able to demonstrate understanding will be asked to provide written informed consent to take part in the study. Written informed consent for study participation must be obtained before any study related procedures are performed. Screening evaluations must be performed within 7 days of entry. Participants screening and enrolment registers will be used to assist with tracking the screening and enrolment process. When informed consent is obtained for the study, a participant identification screening (PID) number will be assigned and eligible participants for trial drugs will receive enrolment PID number. For participants who are found to be ineligible for the study, or who do not enrol in the study for any reason, an electronic case report form (eCRF) will be completed to record the screening outcome.

Additional consent provisions for collection and use of participant data and biological specimens {26b}

SPIRIT guidance: Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable.

Additional consent will be administered for phlebotomy for full pharmacokinetic sampling, which will be performed at 2 weeks after initiation of treatment. The consent will cover week 2 venous blood collection that will be drawn at 1, 2, 6, and 10-12 (late sample per site feasibility) hours and week 8 drawn at 2 and 6 hours after medication administration with attempt by single venipuncture for peripheral IV insertion. Such sampling allows for adequate calculation of C_{max} and AUC. A maximum of 7 ml will be obtained in heparinized tubes at each draw as up to 4 drugs will be required to be assayed for determination of drug concentration within a multidrug anti-TB regimen. Additional consent will be obtained for other baseline specimens like stool, and saliva for future studies.

Interventions

Explanation for the choice of comparators {6b}

SPIRIT guidance: Explanation for choice of comparators.

Intervention description {11a}

SPIRIT guidance: Interventions for each group with sufficient detail to allow replication, including how and when they will be administered.

We will use n-acetylcysteine 900mg an effervescent table as an intervention. A similar tablet containing the same ingredient as the active tablet but zero NAC as a placebo. Both NAC and placebo are manufactured by BioAdventex Pharma Inc -Canada based on good manufacturing practice standards. One intervention group will receive NAC at a dose of 1800mg divided twice daily and another intervention group will receive NAC at a dose of 900mg provided during the evening hours and placebo during the morning hours. The control group will receive placebo both during the morning and evening hours. There will be no restrictions that will be imposed on standard treatment for MDR-TB while NAC is administered.

Criteria for discontinuing or modifying allocated interventions {11b}

SPIRIT guidance: Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease).

Participants will stop the use of trial medications if there will be any serious adverse events related to the use of NAC for instance allergic reactions or participant opt to withdraw from continuing with research participation.

Strategies to improve adherence to interventions {11c}

SPIRIT guidance: Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests).

Study nurses will be responsible for observing the participants taking investigational medicinal product (IMP) during the treatment phase. Study nurses will be providing the participants daily IMP and will document in the IMP treatment adherence chart. Besides, the site trial pharmacist/delegated dispenser will be responsible for dispensing the IMP. Accurate accountability records will be kept by the site to assure that the IMP will not be dispensed to any person who is not a subject under the terms and conditions set forth in this protocol i.e. delivery to site, inventory at site, use by subject, destruction etc. The investigator/designee will immediately inform the sponsor of any quality issues arising with respect to the IMP. The sponsor will take whatever action is required should such a situation arise. The investigator undertakes to use the IMP only as indicated in this protocol.

Relevant concomitant care permitted or prohibited during the trial {11d}

SPIRIT guidance: Relevant concomitant care and interventions that are permitted or prohibited during the trial.

NAC interacts with nitroglycerin resulting in formation of S-nitroso-NAC, which strongly inhibit platelet aggregation whereas the free sulfhydryl donated from NAC, potentiate the systemic, and coronary vasodilator effects of nitroglycerin in patients with acute myocardial infarction or angina pectoris [24, 25]. This effect increases the risk of hypotension. Likewise, the sulfhydryl NAC modifies the renin-angiotensin II, possibly by inhibition of angiotensin converting enzyme inhibitors, thus reduces conversion of angiotensin I to angiotensin II [26, 27].Through different mechanisms, NAC modulates glutamate and may results in clinically relevant psychopharmacological properties. In animal model, Costa-Campos et al investigated the combination of NAC with antidepressant drugs [28]. Findings show NAC reduced the potency of imipramine and escitalopram but not those of desipramine and bupropion. Conversely, in the same model NAC potentiates fluoxetine.
Although there is no concrete evidence on the interaction of NAC with second line anti-TB drugs for MDR-TB, a number of antimicrobial drugs have the potential to exert central nervous system (CNS) effects and many are associated with stimulant, psychotomimetic and epileptogenic properties[29]. For example, quinolone CNS undesired effect is mediated by gamma-aminobutyric acid (GABA) antagonism while cycloserine and aminoglycosides undesired effect is mediated by N-Methly-D-Aspartate (NMDA) agonism. Besides, the effect of linezolid and isoniazid exerts through monoamine oxidase (MAO) inhibition.

Provisions for post-trial care {30}

SPIRIT guidance: Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation.

N/A

Outcomes {12}

SPIRIT guidance: Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance

of chosen efficacy and harm outcomes is strongly recommended.

Primary endpoint
Development of clinical or laboratory-based or both adverse events at any frequency during the first six month of MDR-TB treatment
Primary end-point is based on Safety and Tolerability
Patients will be regularly assessed for adverse events including physical examinations, vitals and routine clinical laboratory tests such as complete metabolic panel, complete blood count and urinalysis

- Proportion of adverse events
- Proportion of adverse events related to experimental treatment

Secondary endpoints
Efficacy

- Proportion of MDR-TB patients requiring second-line anti-TB regimen modifications due to ADRs
- TB Symptoms profile (Appendix 1)
- Time to first negative culture on solid/liquid media
- Proportion of patients converting to negative sputum culture on solid culture at 4 or 6 or 8 months after treatment initiation
- Rate of change of sputum MTB RNA in MBL assay during treatment

Pharmacokinetics endpoints of second-line anti-TB drugs

- Area under the curve (AUC)
- Observed C_{max}
- Volume of distribution
- Elimination half life

Participant timeline {13}

SPIRIT guidance: Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see figure at http://www.spirit-statement.org/publications-downloads/).

Screening may be initiated after a written informed consent is obtained. Screening procedures may be performed on multiple days, including on the date of enrollment. For potential participants who do not meet the eligibility criteria, screening may be discontinued once ineligibility is determined. The information will be entered in the case record form to record the screening outcome. Subjects, who, following the screening assessments, are eligible for the trial and willing to participate, will be randomized/enrolled into the trial and assigned a Randomization Number. Participant will be followed according to the schedule of events as described in Figure 1.

Sample size {14}

SPIRIT guidance: Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations.

Sample size is calculated on the basis of the primary hypothesis “NAC protects against the occurrence of severe ADRs such as ototoxicity with injectable based regimen.” In the longitudinal cohort of 58 MDR-TB patients from the proposed study site in Tanzania, hearing thresholds were measured monthly by pure tone audiometry up to the end of the intensive phase and completion of kanamycin treatment (8 months) (manuscript on preparation). The number of MDR-TB patients that developed hearing loss and substituted kanamycin to capreomycin or decrease of the injectable doses from 5 to 3 times per week was 25%. In general the mean pre-post difference was 16.1 decibels (dB), while a threshold shift of $\geq$ 20dB at high frequency of 8KHz or 4 KHz accord with other evidence that this is clinically significant and important difference [36]. [The mean hearing threshold prior to start of the kanamycin based MDR-TB regimen was 30.5dB, SD (25.1) while the hearing threshold after 8 months of treatment with aminoglycosides was 46.46, SD (27.3).
The sample size estimation has considered event rate of ototoxicity at any grade (mild, moderate, severe and profound) was ~ 60% based in our previous reports instead of clinically significant ototoxicity that resulted in changes of the treatment. To achieve 90% power to detect this difference with a significance level of 5%, it is estimated that a reduction from 60% to 15% is considered the minimum significant and therefore the sample size will be 20-per arm and thus 60 participants will be enrolled for three arms. However, previously we have found a withdrawal/non-evaluable subject rate from our previous TB clinical trials estimated at 10% [37]. Therefore a total of 66 MDR-TB patients will be recruited for the clinical trial [38].

Recruitment {15}

SPIRIT guidance: Strategies for achieving adequate participant enrolment to reach target sample size.

At the on-site, the research coordinators will immediately review newly diagnosed or referred MDR-TB patients and administer processes for potential eligibility and assessment. Likewise, communication with Physicians managing MDR-TB in different centre will eventually sensitize potential participants to facilitate recruitment.

Assignment of interventions: allocation

Sequence generation {16a}

SPIRIT guidance: Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions.

All patients who give consent for participation and those that fulfil the inclusion criteria will be randomized to either the control or the experimental group of 1:1:1 allocation as per a computer-generated random schedule using permuted blocks of randomization (Figure 2).

Concealment mechanism {16b}

SPIRIT guidance: Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned.

The randomized block size will not be disclosed to ensure concealment to avoid selection bias. In addition, block size will randomly vary to avoid the person who assigned randomization to deduce some of the next treatment allocation. Details of the randomization block size will not be included in the protocol, but will be provided in a separate document with restricted access.

To prevent selection bias by facilitating enrolment of the comparable participants in each arm, allocation concealment will be ensured. Participants will complete all baseline measurements prior to release of the randomization code. Opaque sealed envelope with printed randomization numbers will be prepared. For every randomization number a code for a treatment will be assigned.

Primary endpoint (ADRs) will be measured by both clinical assessment and laboratory measurements. To avoid ascertainment bias in the measurement of other endpoints such as adverse events, or performance bias in decision to discontinue or modify treatment, or exclusion/attrition bias in the decision to withdraw from the trial or exclude a participant from analysis, blinding will be implemented. Blinding will be at two levels; the trial participants and health care providers. Health care providers include outcomes assessors (Physicians and Audiologists), data collectors, laboratory staff, nurses and pharmacists.

Implementation {16c}

SPIRIT guidance: Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions.

Allocation sequence will be generated by the sponsor representative pharmacist, site investigators will enroll participants while site research pharmacist will assign participants to interventions

Assignment of interventions: Blinding

Who will be blinded {17a}

SPIRIT guidance: Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how.

Procedure for unblinding if needed {17b}

SPIRIT guidance: If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial.

This is a double-blind trial executing two level of blinding (i) Research participants and (ii) the investigators team. The later includes clinicians and data collectors. Participants will complete all baseline measurements prior to release of the randomization code. Randomization will be generated in the computer with a specific randomization software. For every randomization number a code for a treatment will be assigned. Blinding will be broken, if the research participants experience unexpected serious adverse event. The investigator and the sponsor will communicate and decide appropriately. The standard operating procedure for breaking the blind will be followed.

Data collection and management

Plans for assessment and collection of outcomes {18a}

SPIRIT guidance: Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol.

Data collection

All case record forms (CRF) both electronic and hard copy pages will be completed for every participant receiving any amount of IMP. For screening failure participants, a screening failure CRF will be completed. For subjects who are prematurely withdrawn, the visits up to withdrawal plus the withdrawal and follow-up visits need to be completed.

Source documents

Source documents are defined as all information in original records and certified copies of original records of clinical findings, observations, or other activities in a clinical trial necessary for the reconstruction and evaluation of the trial. Source documents will include, but are not limited to, progress notes, electronic data, screening logs, and recorded data from automated instruments.
All source documents pertaining to this trial will be maintained by the investigators. Source documents will be available for trial-related monitoring, audits, Institutional Review Board (IRB) review and regulatory inspections providing authorized persons direct access to source documents.

File management at the Trial Site

It is the responsibility of the investigators to ensure that the trial center files are maintained in accordance with International Good Clinical Practice Guidelines and the ethical principles that have their origin in the Declaration of Helsinki.

Records retentions at the Trial Site

The investigator will retain records and data from the trial for safety reasons and for audit and inspection subsequent to trial completion. The essential documents should be retained according to ICH-GCP guideline. The sponsor will make financial provisions for the investigator to deposit the documents at an external site for safekeeping for as long as required by regulations and the sponsor.

Plans to promote participant retention and complete follow-up {18b}

SPIRIT guidance: Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols.

Although the site has experience with retaining up to 100% of research participants, refresher training of research staff will be conducted to maintain the enthusiasm and perseverance but also,

high morale, and compassion to bond with patients. This is one of the important strategies in achieving high retention rates

Data management {19}

SPIRIT guidance: Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol.

The CRFs will be filled in a timely, accurate and legible manner. CRF entries will be verifiable to source documentation other than the CRF, as described in the applicable trial instructions. The CRFs will be filled electronically, in a timely, accurate and legible manner. CRF entries will be verifiable to source documentation other than the CRF. Likewise, double data entry will be performed as described in the applicable trial instructions. Site Standard Operating Procedures will be adhered to for all clinical and bioanalytical activities relevant to the quality of the trial. Subject compliance will be monitored throughout the trial.
The investigator will sign and date any analysis results example laboratory test results to verify that the results have been reviewed. The investigator may appoint other sub-investigators to assist with the trial. However, the investigator maintains responsibility for the trial and will supervise the sub-investigators. Written IEC/IRB approval will be obtained prior to involvement in the trial. The investigator will ensure that all site personnel are adequately trained in GCP, the protocol, IB and all trial procedures and requirements.
The study will be monitored to verify that the rights and well-being of human subjects are protected; that trial data are accurate, complete and verifiable with source data; and that the trial is conducted in compliance with the protocol, International GCP, the ethical principles that have their origin in the Declaration of Helsinki and the applicable regulatory requirements.
Monitors assigned by the sponsor will conduct regular site visits for the purpose of monitoring various aspects of the trial. Visits will take place usually within a predetermined interval, but this may vary during the course of the trial. The investigator and site staff will allow the trial monitor and authorized representatives of the sponsor to
(1) Inspect all CRFs, written informed consent documents and corresponding source documents (e.g. original medical records), subject records and laboratory raw data, and
(2) Access clinical supplies, dispensing and storage areas. The investigator and site staff should also (1) agree to assist with monitoring activities if requested and (2) provide adequate time and space for monitoring visits.
The monitor will query any missing, confusing, spurious, or otherwise ambiguous data with the investigator. All queries should be resolved in a timely manner. A monitoring log will be maintained recording each visit, the reason for the visit, the monitor's signature and investigator or designee's confirmation signature.

Confidentiality {27}

SPIRIT guidance: How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial.

All site staff, the sponsor, and any sponsor representatives will preserve the confidentiality of all subjects taking part in the trial, in accordance with International GCP, applicable local legislation/regulations. Subject to the requirement for source data verification by the trial personnel by reference to the subject's notes, confidentiality of all subject identities will be maintained. Only subject trial number and initials will be used on the CRF and in all trial correspondence, as permitted. No material bearing a subject's name will be kept on file by the sponsor. The written informed consent will contain a clause granting permission for review of the participants' source data

Plans for collection, laboratory evaluation and storage of biological specimens for genetic or molecular analysis in this trial/future use {33}

SPIRIT guidance: Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable.

NA

Statistical methods

Statistical methods for primary and secondary outcomes {20a}

SPIRIT guidance: Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol.

The primary efficacy analysis will be conducted using laboratory measurement. Both Modified Intent to Treat (MITT) and a Per Protocol (PP) analysis will be performed.
The primary efficacy endpoint is the change of grade of liver enzyme to the higher one during the intensive phase of MDR-TB treatment. The primary efficacy analysis in the treatment groups will include two comparisons, a superiority comparison of each of the 2 experimental NAC arms with the standard treatment arm.
The difference in the proportion of patients with drug induced liver injury (DILI) (as defined by the primary efficacy endpoint) between each treatment arm and control arm will be calculated with 95% confidence interval using standard methods [39].
In addition, hearing threshold level will be analyzed over time using mixed effect models to account for the longitudinal nature of the data. Mean differences between arms in change from baseline (with 95% confidence intervals) will be derived from the fitted model.

Secondary Efficacy Endpoint Analysis

- Incidence of severe adverse drug reactions (Yes/No) as defined by Common Terminology Criteria for Adverse Event (CTCAE).
- Rate of MDR-TB patients requiring drug modifications due to adverse drug reactions
- Time to first negative culture on solid media
- Rate of patients converting to negative sputum culture on solid culture at 4 or 6 or 8 months after treatment initiation
- Rate of change of sputum MTB RNA in MBL assay during treatment

Safety and Tolerability Analysis

- Incidence of adverse events
- Incidence of adverse events related to experimental treatment

Pharmacokinetics Analysis

Plasma concentration will be used to build a population PK Model to evaluate the effects of NAC on the distribution, metabolism and excretion of MDR-TB drugs

Mycobacterial characterization

Descriptive summary statistics of Mycobacterium characteristics will be presented.

Interim analyses {21b}

SPIRIT guidance: Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial.

There will be no interim analysis. The analysis will be performed when the last participant has completed the last trial procedure. There will be database lock, data analysis and trial reports generated from this trial

Methods for additional analyses (e.g. subgroup analyses) {20b}

SPIRIT guidance: Methods for any additional analyses (eg, subgroup and adjusted analyses).

Methods in analysis to handle protocol non-adherence and any statistical methods

to handle missing data {20c}

SPIRIT guidance: Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation).

We will deploy multiple imputations under the missing at random assumptions for the missing outcomes or covariates

Plans to give access to the full protocol, participant level-data and statistical code {31c}

SPIRIT guidance: Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code.

Data will be available and will be accessed in appropriate data management portals, yet privacy and personal data will not be shared.

Oversight and monitoring

Composition of the coordinating centre and trial steering committee {5d}

SPIRIT guidance: Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee).

There will be a Clinical Trial Management Group which will undertake all sponsorship responsibilities to ensure that the conduct of the clinical trial comply with Medicines for Human use Regulations of 2004 and subsequent amendments for regulated trials. The CTMG will ensure the right, safety, dignity and well-being of the participants are protected and take priority over other interests while the data generated are reliable and robust.

Composition of the data monitoring committee, its role and reporting structure {21a}

SPIRIT guidance: Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed.

A DSMB will be appointed with a primary responsibility of an act in an advisory capacity to the sponsor to safeguard the interests of trial subjects by monitoring subject safety, assess subject risk versus benefit, assess data quality and general evaluation of the trial progress. It's activities will be delineated in a DSMB terms of references that will define the membership, responsibilities and the scope and frequency of data reviews. The DSMB will operate on a conflict-free basis independently of the sponsor and the trial team. It will comprise at least 3 voting members.

The DSMB may have an organisational meeting prior to commencement of the trial. The DSMB will meet approximately every six months and at least annually when it will review unblinded data during a closed session. The sponsor or the DSMB may convene ad hoc meetings if safety concerns arise during the trial. After its assessment, the DSMB will recommend to the sponsor continuation, modification or termination of the clinical trial.

Adverse event reporting and harms {22}

SPIRIT guidance: Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct.

Adverse events will be collected by the investigator from the time a subject sign the Informed Consent Form through to their end of follow-up visit. The exception to this is early withdrawal subjects who will only have SAEs collected from their time of early withdrawal to their end of the follow-up visit. Any AE (serious or non-serious) observed by the investigator or reported by the subject will be recorded on the Adverse Event Case Report Form. The investigator will review each AE and assess its relationship to drug treatment based on all available information at the time of the completion of the case report form.
In the case where an overall diagnosis cannot be made, each specific sign and/or symptom will be recorded as individual AEs. Documentation of the date of onset, and stop date (duration) if applicable will be done. The AEs will also be described in severity, and action taken with IMP while concurrently describing the action taken to the participant. The outcome and relationship to IMP will be recited. Also, the occurrence and seriousness of the AEs will be documented.

Serious Adverse Event (SAE)
Any AE that occurs which is serious must be reported by the investigator to the sponsor within 24 hours of the site first being aware of the SAE, whether or not the serious event is deemed associated with the use of the drug. In addition, the investigator will provide a detailed, signed, written, and complete SAE report form that addresses the investigator's estimates of the attribution/causality of the AE to the trial drug and the seriousness of the AE in question to the trial monitor and sponsor medical monitor within 24 hours of becoming aware of the SAE.The timing of reporting to the sponsor, institutional review board and regulatory authorities will follow the ICH guideline for GCP. The follow up of AE will be followed until there is either satisfactory clinical resolution or stabilization; or until the end of the follow-up period; and or until all queries on these AEs have been resolved.

Frequency and plans for auditing trial conduct {23}

SPIRIT guidance: Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor.

Plans for communicating important protocol amendments to relevant parties (e.g. trial participants, ethical committees) {25}

SPIRIT guidance: Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators).

Any change to the protocol will be effected by means of a protocol amendment. Any changes, which affect subject safety or welfare, will be submitted to the IRB and Regulatory Authorities prior to implementation.Besides information will be incorporated in the Partnership for Access to Clinical Trial Registry

Dissemination plans {31a}

SPIRIT guidance: Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions.

Results of this research will be submitted for publication as soon as feasible upon completion of the trial. An integrative knowledge translation will be used through involving the policy makers. Findings of this study will be presented to different audiences including the community through media coverage and policy briefs. Findings will also be shared in the international conference egnion meeting

Discussion

Trials guidance: This should include a discussion of any practical or operational issues involved in

performing the study and any issues not covered in other sections.

This is the first registered trial to examine the protective effect of NAC on occurrence of adverse drug reactions such as DILI in MDR-TB patients using the WHO injectable free second line anti-TB regimen. Although, the recent priority second line anti-TB regimen include drugs with high effectiveness yet have the potential of causing severe adverse drug reactions and treatment interruptions. `NAC has been recommended as an adjuvant therapy but currently few programmes have adapted this indispensable option perhaps due to lack of evidence. Exploring the benefits of NAC in injectable free MDR-TB regimen, may pave a way of maximizing the benefit of the novel drugs such as bedaquiline, pretomanid and others in use for MDR-TB treatment. NAC trials conducted in DS-TB showed not only a protective effect in DILI but also delayed the onset and fasten the resolutions of DILI{Cheng, 2016 #339}.Likewise a large cohort study revealed that patients that received pulmonary TB treatment and NAC had a substantial reduction in 90 day all-cause mortality NAC{Jeeraaumponwat, 2019 #340}
While NAC may provide an opportunity of halting severe drug reaction events, still the drug may contribute to the occurrence of adverse events. For example in a pooled analysis of 83 studies (N=9988) described the administration of NAC for >6 weeks. Important adverse events included the gastrointestinal intolerance such as nausea, vomiting, abdominal pain and arthralgia increased 1.4–2.2 times [15]. These findings show clinical equipoise with MDR-TB drugs such as bedaquiline, pyrazinamide, flouroquinolone, and may be worse with co-administration with NAC.
Although there is no concrete evidence on the interaction of NAC with second line anti-TB drugs for MDR-TB, a number of antimicrobial drugs have the potential to exert central nervous system (CNS) effects and many are associated with stimulant, psychotomimetic and epileptogenic properties[29]. For example quinolone CNS undesired effect is mediated by gamma-aminobutyric acid (GABA) antagonism while cycloserine and aminoglycosides undesired effect is mediated by N-Methly-D-Aspartate (NMDA) agonism. Besides, the effect of linezolid and isoniazid exerts through monoamine oxidase (MAO) inhibition.

Trial status

Trials guidance: Authors should report the protocol version number and date, the date recruitment began, and the approximate date when recruitment will be completed.

Recruitment has not started

Abbreviations

Trials guidance: If abbreviations are used in the text they should be defined in the text at first use, and a list of abbreviations should be provided.

Abbreviations

ADR	Adverse Drug Reaction
AE	Adverse Event
ALT	Alanine Transaminase
AST	Aspartate Transaminase
AUC	Area Under the curve
CTCAE	Common Terminologies Criteria for Adverse Events
CTMG	Clinical Trial Monitoring Group
DILI	Drug Induced Liver Injury
DSMB	Data Safety Monitoring Board
EDCTP	European and Developing Countries Clinical Trials Partnerships
GCP	Good Clinical Practice
GSH	Glutathione
ICF	Informed Consent Form
ICH	International Conference of Harmonization
ICIDR	International Consortium for Infectious Diseases Research

KIDH	Kibong'oto Infectious Diseases Hospital
MDRTB	Multidrug resistant tuberculosis
MITT	Modified Intention to Treat
MTB	*Mycobacterium tuberculosis*
NAC	N acetylcysteine
PACTR	Pan-African Clinical Trial Registry
PanACEA	Pan-African Consortium for Evaluating Anti-TB Antibiotics
PID	Patients Identification
PK	Pharmacokinetics
PP	Per-Protocol
REDCap	Research Electronic Data Capture
SADR	Severe Adverse Drug Reaction
TB	Tuberculosis
WHO	World Health Organization

Declarations

Trials guidance: All manuscripts must contain the following subheadings:

- Acknowledgements
- Authors' contributions
- Funding
- Availability of data and material
- Ethics approval and consent to participate
- Consent for publication
- Competing interests
- Authors' information (optional)

Acknowledgements

Trials guidance: Please acknowledge anyone who contributed towards the article who does not meet the criteria for authorship including anyone who provided professional writing services or materials. Authors should obtain permission to acknowledge from all those mentioned in the Acknowledgements section. See our editorial policies for a full explanation of acknowledgements and authorship criteria. If you do not have anyone to acknowledge, please write "Not applicable" in this section.

Group authorship (for manuscripts involving a collaboration group): if you would like the names of the individual members of a collaboration Group to be searchable through their individual PubMed records, please ensure that the title of the collaboration Group is included on the title page and in the submission system and also include collaborating author names as the last paragraph of the "Acknowledgements" section. Please add authors in the format First Name, Middle initial(s) (optional), Last Name. You can add institution or country information for each author if you wish, but this should be consistent across all authors. Please note that individual names may not be present in the PubMed record at the time a published article is

initially included in PubMed as it takes PubMed additional time to code this information.

We would like to acknowledge the Kibong'oto Infectious Diseases Hospital Administration and the Ministry of Health, Community Development Gender Elderly and Children for the continuous administration support. We are grateful to the European and Developing Countries Clinical Trial Partnership (EDCTP) for agreeing to support the clinical trial capacity through EDCTP-2 programme

Authors' contributions {31b}

SPIRIT guidance: [31b] - Authorship eligibility guidelines and any intended use of professional writers.

Trials guidance: The individual contributions of authors to the manuscript should be specified in this section. Guidance and criteria for authorship can be found in our editorial policies. Please use initials to refer to each author's contribution in this section, for example: "AB is the Chief Investigator; she conceived the study, led the proposal and protocol development. CD contributed to study design and to development of the proposal. EF was the lead trial methodologist. All authors read and approved the final manuscript."

SGM is the Chief Investigator, she conceived the study led the proposal and protocol development. SKH designed the study and co-led proposal development. MB, SHG and GSK contributed in the proposal development. PJP was the lead trial methodologist. AAL, PMM, HHS and HCM involved in protocol development. All authors have read and approved the manuscript

Funding {4}

SPIRIT guidance: Sources and types of financial, material, and other support.

Trials guidance: All sources of funding for the research reported should be declared. You will be required to include a copy of the original funding document and an English translation of this document as an additional file on submission, which will be checked against this declaration. The role of the funding body in the design of the study and collection, analysis, and interpretation of data and in writing the manuscript should be declared.

This study receives financial supports from the European and Developed Countries Clinical Trials Partnership (EDCTP2) program supported by the European Union project through the Senior Fellowship Scheme TMA1463 awarded to Stellah G Mpagama

Availability of data and materials {29}

SPIRIT guidance: Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators.

Trials guidance: Please do not include any baseline or pilot data in your study protocol. The Editorial Office will ask you to remove this if it is included. Please declare here who will have access to the final trial dataset and disclose contractual agreements that limit such access for investigators.

Not applicable

Ethics approval and consent to participate {24}

SPIRIT guidance: Plans for seeking research ethics committee/institutional review board (REC/IRB) approval.

Trials guidance: Trials do not consider study protocols for studies without ethical approval. You will be required to provide a copy of the original ethical approval document and an English translation of this document as an additional file on submission, which will be checked against this declaration. The name of the ethics committee that approved the study and the committee's reference number (if applicable) should be declared. Details of authors' intentions to obtain consent to participate in the study from participants (or their parent or legal guardian in the case of children under 16) should be declared. "eg. ABC Ethical Review Board ABC123456. Written, informed consent to participate will be obtained from all participants"

Our proposal has been approved by the Research and Ethics Review Committee (RERC) for three collaborative institutions namely Kibong'oto Infectious Diseases Hospital, Nelson Mandela African Institution of Science and Technology and Center for Educational Development in Health -Arusha acronym KNCHREC. Likewise, the proposal has been approved by the National Health Research Ethics Sub-Committee (NatHREC) of the National Medical Research Institute (NIMR) of Tanzania. Received regulatory approval from the Tanzania Medicines and Diagnostics Agency. Informed consent will be obtained from all study participants.

Consent for publication {32}

SPIRIT guidance: Model consent form and other related documentation given to participants and authorised surrogates.

Trials guidance: Please do not include any baseline or pilot data in your study protocol. The Editorial Office will ask you to remove this if it is included. If you have included any details, images or videos relating to an individual person, written informed consent for the publication of these details must be obtained from that person (or their parent or legal guardian in the case of children under 18) and declared in this section. Please also state whether you will be willing to provide a model consent form on request. If this section does not apply, please state "Not applicable".

Not applicable

Competing interests {28}

SPIRIT guidance: Financial and other competing interests for principal investigators for the overall trial and each study site.

The authors declare that they have no competing interests.

Trials guidance: All financial and non-financial competing interests must be declared in this section. See our editorial policies for a full explanation of competing interests. If you are unsure whether you or any of your co-authors have a competing interest please contact the editorial office. Please use the authors initials to refer to each authors' competing interests in this section. If you do not have any competing interests, please state: "The authors declare that they have no competing interests" in this section.

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References

References

1. World Health Organization (WHO): **Global Tuberculosis Report**. WHO/HTM/TB/2016.13; 2016.
2. World Health Organization (WHO): **Global Tuberculosis Report**. 2019.
3. Hogan AB, Jewell BL, Sherrard-Smith E, Vesga JF, Watson OJ, Whittaker C, Hamlet A, Smith JA, Winskill P, Verity R *et al*: **Potential impact of the COVID-19 pandemic on HIV, tuberculosis, and malaria in low-income and middle-income countries: a modelling study**. *The Lancet Global Health* 2020.
4. Brigden G, Nyang'wa BT, du Cros P, Varaine F, Hughes J, Rich M, Horsburgh CR, Jr., Mitnick CD, Nuermberger E, McIlleron H *et al*: **Principles for designing future regimens for multidrug-resistant tuberculosis**. *Bull World Health Organ* 2014, **92**(1):68-74.
5. Mpagama SG, Houpt ER, Stroup S, Kumburu H, Gratz J, Kibiki GS, Heysell SK: **Application of quantitative second-line drug susceptibility testing at a multidrug-resistant tuberculosis hospital in Tanzania**. *BMC Infect Dis* 2013, **13**:432.
6. WHO: **Guidelines for the programmatic management of drug-resistant tuberculosis Emergency Update**; 2008.
7. Ahmad N, Ahuja SD, Akkerman OW, Alffenaar J-WC, Anderson LF, Baghaei P, Bang D, Barry PM, Bastos ML, Behera D *et al*: **Treatment correlates of successful outcomes in pulmonary multidrug-resistant tuberculosis: an individual patient data meta-analysis**. *The Lancet* 2018, **392**(10150):821-834.
8. WHO: **WHO Consolidated Guidelines on Drug Resistant Tuberculosis Treatment**. 2019.
9. Nunn AJ, Phillips PPJ, Meredith SK, Chiang CY, Conradie F, Dalai D, van Deun A, Dat PT, Lan N, Master I *et al*: **A Trial of a Shorter Regimen for Rifampin-Resistant Tuberculosis**. *N Engl J Med* 2019, **380**(13):1201-1213.
10. Mpagama SG, Heysell SK, Ndusilo ND, Kumburu HH, Lekule IA, Kisonga RM, Gratz J, Boeree MJ, Houpt ER, Kibiki GS: **Diagnosis and interim treatment outcomes from the first cohort of multidrug-resistant tuberculosis patients in Tanzania**. *PLoS One* 2013, **8**(5):e62034.
11. Malik AA, Fuad J, Siddiqui S, Amanullah F, Jaswal M, Barry Z, Jabeen F, Fatima R, Yuen CM, Salahuddin N *et al*: **Tuberculosis Preventive Therapy for Individuals Exposed to Drug-resistant Tuberculosis: Feasibility and Safety of a Community-based Delivery of Fluoroquinolone-containing Preventive Regimen**. *Clin Infect Dis* 2020, **70**(9):1958-1965.
12. Mbuagbaw L: **Review of available evidence on the use of bedaquiline for the treatment of multidrug resistant tuberculosis: Data analysis report. Prepared for The World Health Organization**. *McMaster University: Health Research Methods, Evidence and Impact* 2017.

13. Agyeman AA, Ofori-Asenso R: **Efficacy and safety profile of linezolid in the treatment of multidrug-resistant (MDR) and extensively drug-resistant (XDR) tuberculosis: a systematic review and meta-analysis**. *Ann Clin Microbiol Antimicrob* 2016, **15**(1):41.
14. Hwang TJ, Dotsenko S, Jafarov A, Weyer K, Falzon D, Lunte K, Nunn P, Jaramillo E, Keshavjee S, Wares DF: **Safety and availability of clofazimine in the treatment of multidrug and extensively drug-resistant tuberculosis: analysis of published guidance and meta-analysis of cohort studies**. *BMJ Open* 2014, **4**(1):e004143.
15. Ferlazzo G, Mohr E, Laxmeshwar C, Hewison C, Hughes J, Jonckheere S, Khachatryan N, De Avezedo V, Egazaryan L, Shroufi A *et al*: **Early safety and efficacy of the combination of bedaquiline and delamanid for the treatment of patients with drug-resistant tuberculosis in Armenia, India, and South Africa: a retrospective cohort study**. *The Lancet Infectious Diseases* 2018, **18**(5):536-544.
16. Winterbourn C, Metodiewa D: **Reactivity of Biologically Important Thiol Compounds with Superoxide and Hydroen Peroxide**. *Free Radical Biology & Medicine* 1999, **27**(3/4):322-328.
17. Pompella A, Visvikis A, Paolicchi A, Tata VD, Casini AF: **The changing faces of glutathione, a cellular protagonist**. *Biochemical Pharmacology* 2003, **66**(8):1499-1503.
18. Elbini Dhouib I, Jallouli M, Annabi A, Gharbi N, Elfazaa S, Lasram MM: **A minireview on N-acetylcysteine: An old drug with new approaches**. *Life Sci* 2016, **151**:359-363.
19. Ershad M, Naji A, Vearrier D: **N Acetylcysteine** *NCBI Bookshelf A service of the National Library of Medicine, National Institutes of Health:StatPearls Publishing LLC* 2020.
20. Baniasadi S, Eftekhari P, Tabarsi P, Fahimi F, Raoufy MR, Masjedi MR, Velayati AA: **Protective effect of N-acetylcysteine on antituberculosis drug-induced hepatotoxicity**. *European journal of gastroenterology & hepatology* 2010, **22**(10):1235-1238.
21. Farazi A, Sofian M, Jabbariasl M: **Efficacy of N-Acetylcysteine on Prevention of Antituberculosis Drug-Induced Hepatotoxicity**. *World Journal of Medical Sciences* 2015, **12**(4):413-418.
22. Kranzer K, Elamin WF, Cox H, Seddon JA, Ford N, Drobniewski F: **A systematic review and meta-analysis of the efficacy and safety of N-acetylcysteine in preventing aminoglycoside-induced ototoxicity: implications for the treatment of multidrug-resistant TB**. *Thorax* 2015, **70**(11):1070-1077.
23. Raviglione M: **Vision and proposed framework for a Post-2015 TB Elimination Strategy and Targets; Consultation on "Elimination the catastrophic economic burden of TB" Universal Health Coverage and Social Protection Opportnities** In*: 2013; Sao Paulo, Brazil,29April - 1 May 2013*; 2013.
24. Nyaki FS, Taksdal M, Mbuya AW, Sariko M, Lekule IA, Kisonga RM, Kibiki GS, Mmbaga BT, Heysell SK, Mpagama SG: **Predictors of Nutritional Status in Patients Treated for Multidrug-Resistant Tuberculosis at a Referral Hospital in Tanzania**. *Journal of Clinical Infectious Diseases & Practice* 2016, **01**(02).
25. Boeree MJ, Heinrich N, Aarnoutse R, Diacon AH, Dawson R, Rehal S, Kibiki GS, Churchyard G, Sanne I, Ntinginya NE *et al*: **High-dose rifampicin, moxifloxacin, and SQ109 for treating tuberculosis: a multi-arm, multi-stage randomised controlled trial**. *The Lancet Infectious Diseases* 2017, **17**(1):39-49.
26. Horowitz J, Henry C, Syrjanen M, Louis W, Fish D, Smith T, Anman E: **Combined use of nitroglycerin and N-acetylcysteine in the management of unstable angina pectoris**. *Circulation* 1988, **77**(4):787-794.
27. Ignarro LJ, Gruetter CS: **Requirement of thiols for activation of coronary arterial guanylate cyclase by glyceryl trinitrate and sodium nitrite possible involvement of S-nitrosothiols**. *Biochimica et Biophysica Acta (BBA) - General Subjects* 1980, **631**(2):221-231.

28. Barrios V, A. C, Navarro-Cid J, V. L, Ruilope L: **N Acetylcysteine Potentiates the Antihypertensive Effect of ACE Inhibitors in Hypertensive Patients.** *Blood Pressure* 2009, **11**(4):235-239.
29. Boesgaard S, Aldershvile J, Poulsen HE, S. C, H. D-P, J. G: **N-Acetylcysteine inhibits Angiotensin Converting Enzyme in Vivo**. *The J Pharmacol and Experiment Theraeup* 1993, **265**(3):1239-1244.
30. Costa-Campos L, Herrmann AP, Pilz LK, Michels M, Noetzold G, Elisabetsky E: **Interactive effects of N-acetylcysteine and antidepressants**. *Prog Neuropsychopharmacol Biol Psychiatry* 2013, **44**:125-130.
31. Zareifopoulos N, Panayiotakopoulos G: **Neuropsychiatric Effects of Antimicrobial Agents**. *Clin Drug Investig* 2017, **37**(5):423-437.
32. Boeree MJ, Heinrich N, Aarnoutse R, Diacon AH, Dawson R, Rehal S, Kibiki GS, Churchyard G, Sanne I, Ntinginya NE *et al*: **High-dose rifampicin, moxifloxacin, and SQ109 for treating tuberculosis: a multi-arm, multi-stage randomised controlled trial**. *The Lancet Infectious Diseases* 2016.
33. Ltd SE: **Power calculator for continuous outcome superiority trial.** . In. Edited by 2016]. OAfhwscpc-sAWD; 2012.
34. Zhang Y, Wu S, Xia Y, Wang N, Zhou L, Wang J, Fang R, Sun F, Chen M, Zhan S: **Adverse Events Associated with Treatment of Multidrug-Resistant Tuberculosis in China: An Ambispective Cohort Study**. *Med Sci Monit* 2017, **23**:2348-2356.
35. Cheng S-L: **Protective effect of N-acetylcysteine on antituberculosis drug-induced hepatotoxicity**. In: *102 Tuberculosis.* 2016.
36. Jeeraaumponwat T: **N-acetylcysteine and mortality in hospitalized pulmonary tuberculosis infection**. In: *Tuberculosis.* 2019.
37. Campochiaro PA, Iftikhar M, Hafiz G, Akhlaq A, Tsai G, Wehling D, Lu L, Wall GM, Singh MS, Kong X: **Oral N-acetylcysteine improves cone function in retinitis pigmentosa patients in phase I trial**. *J Clin Invest* 2020, **130**(3):1527-1541.
38. Mullins ME, Vitkovitsky IV: **Hemolysis and hemolytic uremic syndrome following five-fold N-acetylcysteine overdose**. *Clin Toxicol (Phila)* 2011, **49**(8):755-759.
39. Bailey B, Blais R, Letarte A: **Status epilepticus after a massive intravenous N-acetylcysteine overdose leading to intracranial hypertension and death**. *Ann Emerg Med* 2004, **44**(4):401-406.
40. Cassidy N, Tracey JA, Drew SA: **Cardiac arrest following therapeutic administration of N-acetylcysteine for paracetamol overdose**. *Clin Toxicol (Phila)* 2008, **46**(9):921.