Dated 18/09/2024

**Study Protocol**

An Insight on the Relationship Between the Variation in Medication Adherence Level and the Clinical Outcomes among Tuberculosis and Human Immunodeficiency Virus Co-Infected Patients

## 1. Background and Significance

Tuberculosis (TB) is a leading cause of death in young people and adults (1). The control of tuberculosis remains a major public health challenge in all parts of the world. In some geographical areas, particularly in low and middle-income countries, TB incidence is increasing rapidly and is often associated with a high prevalence of human immunodeficiency virus (HIV) (2). DOTS (directly observed treatment, short course) is the internationally recommended control strategy for TB. This strategy includes the delivery of a standard short course of drugs, lasting 6 months for new patients and 8 months for retreatment patients diagnosed with TB. The delivery includes the direct observation of therapy (DOT), either by a health worker or by someone nominated by the health worker and the patient for this purpose (sometimes called a DOT supporter). The strategy has been promoted widely and implemented globally (3). A qualitative approach and pharmacist-led intervention are needed to improve the adherence level of medication for TB patients.

## 2. Literature review

In 2019, out of 10 million people who had TB, 8.2% were diagnosed with HIV (4). According to the Ministry of Health, 87,581 persons were living with HIV in Malaysia as of 2019. Only 77,903 people are aware of their situation. According to the United Nations, Malaysia is one of the 10 nations responsible for over 95% of all new HIV infections in the Asia-Pacific area in 2016. TB is a disease with high prevalence and high mortality in many developing nations. Antiretroviral therapy (ART) has improved in effectiveness over the last few decades, allowing PLWH to live longer. Though antiretroviral medication adherence remains a significant predictor of treatment success, there is currently no gold standard tool for reliably measuring ART adherence, and scientists and researchers are still working on new methods to achieve this aim. Current anti-TB therapy (ATT) consists of a cocktail of drugs taken over at least 6-8 months. Because of the long duration of the therapy, there is a risk of treatment interruption or default, a phenomenon that contributes to prolonged infectiousness, relapse, and drug resistance.

The difficulty experienced by patients in following treatment regimens has raised the awareness of adherence as a complex behavioral issue. Efforts to improve treatment outcomes require a better understanding of particular barriers to and facilitators of patient adherence. Although the available anti-TB drugs are effective and supplied free of charge in many national TB programs (NTPs), non-adherence to ATT is a major barrier to global control. It may result in persistent infectiousness and higher rates of treatment failure, relapse, and drug resistance, requiring more prolonged and expensive treatment that is less likely to be successful than treatment for drug-susceptible TB.

The prolonged treatment period can have detrimental effects on the patient's health-related quality of life (HRQoL). Reduced HRQoL has been attributed to symptoms of TB, side effects of drug therapy, societal stigma, economic costs, anxiety, and depression during the treatment process. Adherence is measured by outcome-oriented (e.g. cure rate) and process-oriented (e.g. visit attendance, pill counts) methods (5). It can also be measured by direct and indirect methods. The direct method involves directly observed therapy (DOT), detection of drugs and metabolites inurine and blood, and involves ingestible sensor-based system. DOT is founded on the idea that a trained and monitored agent – such as a health worker, a community volunteer, or a family member

– directly supervises patients ingesting their anti-TB drugs.

Anti-TB medications or their metabolites can be measured to provide objective proof of drug intake by the patient. This, together with its ability to be conducted at the point-of-care level, should be considered the key advantage of the Direct method of adherence measurement.

Clinical pharmacists providing pharmaceutical care services have been shown to improve adherence to therapy and reduce potential prescribing errors. Pharmacists should educate patients about the treatment, potential side effects, and drug-drug interactions. Patient awareness and education regarding treatment regimens improve the rate of adherence. One objective measure of adherence is a biochemical test called the Arkansas method, where a chemical reaction with urinary INH metabolites produces a visible blue color change. This method has a high sensitivity (>99%) and specificity (>96%) (6). Their low cost, ease of use, and accuracy make them an ideal evaluative component of TB control programs. As an objective test, it might be used repeatedly as a follow-up method to better characterize adherence and potentially increase adherence among patients receiving treatment for active TB. *IsoScreen* is a new colorimetric point-of-care diagnostic for detecting isoniazid (INH) metabolites in urine.

Indirect methods for the measurement of drug adherence are self-interview, use of electronic monitoring devices, pill counts, and prescription review. In the clinical setting, patient self-reports are frequently used. Questionnaires, scales, interviews, and patient diaries are among them. The most common questionnaires for adherence measurement are The Morisky Medication Adherence Scale (MMAS), the Brief Medication Questionnaire (BMQ), the Medication Adherence Rating Scale (MARS), and Visual Analogue Scales (VAS) (7).

It is well established that providers' adherence figures are often unreliable, leading to the incorrect presumption of good adherence. The adherence metrics used in research studies and clinical practice are medication electronic monitoring system (MEMS), pill counts, biological markers of adherence, Pharmacy refill data, and verbal self-report [9]. Virologic, immunologic, and therapeutic results are all linked to adherence to ART. Also, small gains in adherence can have a big impact on these outcomes. However, near-perfect adherence is required to increase the chances of long-term clinical success. People from all classes who have been treated often struggle to sustain such a high degree of medication adherence. To ensure that patients get the most out of ART, clinicians must inquire about and encourage medication adherence.

**Table 2.1.** Studies reporting the significant predictors

| Color | 1 minute & 5 minutes | |
|---|---|---|
| Color | Status | Interpretation |
| Yellow | Negative | Urine color remains unchanged. |
| Purple<br>Blue | Positive | INH was taken recently. |
| Green | Equivocal | Drugs have been taken recently but not within the last 30 hours |

## 3. Research aim and objectives

The study is aimed to investigate the degree of adherence and evaluate its impacts on clinical outcomes in TB and HIV co-infected patients.

### 3.1 Specific Objectives

1. To perform a systematic review and meta-analysis on evidence-based TB drug adherence.

2. To determine the association between the pharmacist-led intervention and medication adherence level among TB and HIV co-infected patients.
3. To assess the effects of different levels of adherence with therapeutic impact and its possible clinical outcomes.
4. To assess the survival trend with the study population and health-related quality of life (HRQoL) by using the EQ5Dv3 tool.
5. To assess the defaulter patients' approach towards TB and its treatment outcomes. (Qualitative Approach)

## 4. Ethical Approval

The study protocols will be approved by the Ethical and Research Committee Ministry of Health, Malaysia. The protocol will also be registered at clinicaltrials.gov

## 5. Methodology

### 5.1 Study Design

A longitudinal study design will be adopted to achieve the present study objectives. The study population will be classified with one intervention and one control arm, using a 1:1 allocation ratio, to evaluate whether the intervention is superior to the control treatment will be adopted to achieve the study objectives at General Hospital Penang.

**Intervention arm:** Enrolled participants will be counseled and educated by a pharmacist on the therapy and benefits of adherence.

**Control arm:** Enrolled patient will be counseled and educated by the material provided on the therapy and benefits of adherence.

### 5.2 Study Setting

The study will be conducted at Hospital Pulao, Penang from November 2022 to November 2023. We will enroll patients with culture-confirmed TB. Additionally, the enrolled PLHIV with any TB symptoms will be investigated with sputum smear microscopy, mycobacterial culture (BACTEC MGIT), Xpert Ultra, and Chest X-ray (CXR). Patients will be eligible for enrolment if they are ≥18 years of age, diagnosed with active TB according to national TB program guidelines, had not been previously treated for these diseases, agreed to start treatment for TB, and could give informed consent in either English or Malay.

In Malaysia, a DOT program has been implemented in two phases, “intensive” and “continuation”. In the intensive phase (daily DOT) the current standard anti-TB regimen recommended by WHO

Consists of a 2-month treatment with INH, rifampicin (RIF), pyrazinamide (PZA), and ethambutol(EMB), followed by a 4-month continuation phase with INH and RIF (2EHRZ/4HR) as daily regimen. In the intensive phase (daily DOT), a patient takes their medication in front of the healthcare worker every day at a health facility. In the continuation phase (non-daily DOT), a patient is given the responsibility to take the medication at home and come to a health facility weekly for health checking and medication refilling.

**5.3. Participants**

Adult TB and HIV co-infected patients who are receiving treatment at specific public health facilities in Penang, Malaysia will be prospective participants for the study.

**5.4. Data Collection and Management**

A trained healthcare professional team will collect and supervise the data collection. Enrolled participants will answer a brief questionnaire focused on recent isoniazid adherence. Patients will be asked to report their recent adherence, including the date of their most recent dose. They will be also asked to estimate how many of the last thirty doses they had missed or how many doses they missed in a typical week and the reasons for any missed doses. Data will be collected from the primary source through interviews and sample urine collection, and a secondary source by a review of the health facility attendance registration book to confirm the patient's medication refill visit attendance. A trained pharmacist and nurses will collect urine specimens in a clean and dry container. Urine collected at any time of the day may be used. The sample does not have to be freshly voided as the sample can be stored at room temperature for 1 to 10 days and thereafter at -20°C and retain its suitability for testing. The urine sample will be collected on day 0, and then after every 2 months i.e., 2 months, 4 months, and 6 months. The urine sample will be discarded immediately after the analysis. A health worker will be trained to administer the EQ-5D. Enrolled patients will be explained the purpose and duration of the study. All participants included in the study provided informed consent and completed the EQ-5D facilitated by the trained health worker in a face-to-face interview at the clinic. The interview will be conducted on the premises of the hospital. Interviews will be conducted at the start of the study, after 2 2-month visits, and then at the end of the study ($6^{th}$ month). To ensure the security of the patients' information, a double-blind randomization technique will be adopted. The computer gives each patient a code number, and thecode numbers will then be allocated randomly to the treatment groups. All data will be kept in a database, and backup file with password protectors. All identifiable information will be removed

And replaced by code. Code breaking will be done by the external personnel who are not involved inthe study so the biasedness couldn't be possible. At the end of the trial, the patients can check their records on request. The study record shall be destroyed after the research is completed.

**5.5. Study End Point:**

The study will be ended after 6$^{th}$ month, but still, if the patients are non-adherent to the therapy the study period will be extended to 1 year.

**5.6. Inclusion Criteria**

Patients have to meet the following criteria,

- Men and women aged 18 years or more.
- Newly bacteriologically confirmed TB case (less than a month since diagnosis). This restriction (not more than one month of treatment) does not refer to patients whose most recent treatment outcome was a failure and who were assigned to a new treatment regimen.
- No plans to move out of the clinical settings of the participating TB program sites within 9 months of enrollment.

**5.7 Exclusion Criteria**

- Pregnancy and any medical condition unrelated to TB will be considered on exclusion grounds.
- Currently enrolled in a clinical trial that prohibits enrollment in another study.
- Patients who are leaving the area within the next six months.
- Previous history of TB, MDR, or extensively drug-resistant (XDR) TB.

**5.8 Data Analysis**

All analyses were performed using SPSS software, version 28.0 (SPSS Inc., Chicago, IL, USA). *T-test*, $\cong\chi^2$ test, Fisher's exact test, and a logistic regression model were used to determine differences between the intervention group and the control group. According to log-likelihood values, the best fit for the correlation structure was determined to be a compound symmetry structure and random effects were applied to the slope as well as to the intercept. In addition, analysis of variance (ANOVA) was used to determine differences between the two groups based on each monthly accumulated drug adherence rate. Statistical significance was set at $p < 0.05$ (16)

**5.9 Statement that collection of specimens**

A urine sample is collected for *ISoScreen* Urine analysis to know whether either patient is taking his medication regularly or not. The patient's consent is of utmost important. If patients are comfortable with the collection of samples, then the sample will be collected otherwise the sample will not be collected. An informed consent form translated into both English and Malay will be provided to the patient.

### 5.10 Rationale for collection of specimens

Treatment adherence for coinfection diseases is difficult due to the complexity, low tolerability, and extended duration of current treatment regimens, especially for both drug-susceptible and drug-resistant TB. As a result, poor adherence raises the chance of treatment failure, relapse, and the development or amplification of drug resistance. Studies have shown rates of non-adherence to be typically 8–33% [5]. The adherence metrics used in research studies and clinical practice have a big impact on the clinical outcomes of the disease. However, near-perfect adherence is required to increase the chances of long-term clinical success. More research is needed on the factors that influence treatment adherence in TB and HIV patients receiving concurrent treatment. As a result, we will undertake a study to learn more about evidence-based adherence and treatment outcomes in TB and HIV patients.

### 5.11 Specimen Storage for Future Use

Urine analysis will be discarded immediately after the analysis. No sample will be reused in any further research.

## 6. Sample Size

The sample size for the current study is based on a statistical superiority trial (continuous data) design of a randomized control trial. Based on the nature of the relevant amount, a superiority design contains statistical superiority trials and clinical superiority trials (9).

$$N = 2 \times \frac{Z_{1-\alpha=2}\, Z_{1-b}^{2} \times S^{2}}{d}$$

Where N is the size per group, *p* is the response rate of the standard treatment group; *p* 0 is the responserate of the treatment group; *z is* the standard normal deviate for a 1 or 2-sided *x*; *d is* the real difference between 2 treatment effect; a clinically acceptable margin; and S is the Polled standard deviation of both comparison groups. By following this design and keeping in mind the prevalence of TB

And HIV co-infection in Malaysia, i.e., is 5.9%, the calculated sample size calculated is 382 individuals (191 subjects per arm). We'd need a minimum final sample size of 420 people if we assume a 20% attrition rate (210 per group). All estimates are based on a two-sided test with a significance level of $p < 0.05$.

**7. Privacy and Confidentiality**

Subject's names will be kept on a password-protected database and will be linked only with a study identification number for this research. The identification number instead of patient identifiers will be used on subject data sheets. All data will be entered into a computer that is password-protected. On completion of the study, data in the computer will be copied to CDs, and the data in the computer erased. CDs and any hardcopy data will be stored in a locked office of the investigators and maintained for a minimum of three years after the completion of the study. The CDs and data willbe destroyed after that period of storage. Subjects will not be allowed to view their studydata, as the data will be consolidated into a database. Subjects can write to the investigators to request access to study findings.

**8. Termination of Study**

The researcher/ sponsor may decide to terminate the study at any time. Subjects will be informed if the study is terminated and follow-up visits will be arranged if needed. The criteria for termination depend on unseen circumstances and, the incorporation of the patients.

**9. Withdrawal Criteria**

Subjects can choose to withdraw at any time. Subjects may be withdrawn if the investigator deems that it is detrimental or risky for the subject to continue. As this is a prospective observational study and data in the form of information are only required, the literature suggests about 10-20% dropout or withdrawal has been observed in the past so 20% dropout is already included in the sample size calculation thus the withdrawn subjects will not be replaced with new subjects.

**10. Selection of subjects to be included in the analysis**

All the subjects who will participate until the end of this study will be included in the final analysis ofthis study. All subjects who withdraw from the study at any point in time will not be included inthefinal results analysis.

**11. Risk-Benefit Assessment**

As stated above, there is no risk from the investigational process and study procedures. No harm could be caused to the patient as there is no invasive procedure involved in the study nor any

Medication is given to the patient. Only non-clinical intervention is given in the study. Study findings shall potentially improve treatment outcomes. The expected benefit outweighs the minimal risk to subjects and thus this study should be supported.

If still any injuries do occur as a direct result of participating in the study, treatment for such injuries shall be provided or paid for by the researcher/ sponsor.

## 12. Ethics of Study

The study will be conducted in compliance with ethical principles outlined in the Declaration of Helsinki and the Malaysian Good Clinical Practice Guideline.

All aspects of the study protocol, including access to and the use of clinical information of patients, and demographics will get approval by the institutional medical ethics committee locally as well as central health authorities before initiation of this study. Information on individuals will be strictly protected and used for clinical research only.

## 13. Informed Consent/Assent Process

Patients shall be informed of the study during their usual clinic visits. They will be requested to contact investigators if they are interested. An appointment will be made where the patient information sheet will be provided and explained to them. If they are willing to participate, the consent forms will be signed and dated. If they need to, they are allowed to take the information sheet home to consult with their family members, and another day for getting consent arranged.

## 14. Conflict of Interest

The investigators declare they have no conflict of interest.

## REFERENCES

1. Elziny MM, Ghazy A, Elfert KA, Aboukamar M. Case report: development of miliary pulmonary tuberculosis in a patient with peritoneal tuberculosis after COVID-19 upper respiratory tract infection. The American Journal of Tropical Medicine and Hygiene. 2021;104(5):1792.
2. Sulaiman SAS, Khan AH, Ahmad N, Iqbal MS, Muttalif AR, Hassali MA. Impact of diabetes mellitus on treatment outcomes of tuberculosis patients in tertiary care setup. The American journal of the medical sciences. 2013;345(4):321-5.
3. Munro SA, Lewin SA, Smith HJ, Engel ME, Fretheim A, Volmink J. Patient adherence to tuberculosis treatment: a systematic review of qualitative research. PLoS medicine. 2007;4(7):e238.
4. Chaudhry M, Alessandrini M, Pepper MS. Pharmacogenomics for infectious diseases in sub-Saharan Africa: successes and opportunities. Applied & Translational Genomics. 2016;9:3.
5. Laghari M, Talpur BA, Sulaiman SAS, Khan AH, Bhatti Z. Assessment of adherence to anti-tuberculosis treatment and predictors for non-adherence among the caregivers of children with tuberculosis. Transactions of the Royal Society of Tropical Medicine and Hygiene. 2021;115(8):904-13.

6. Ellis SL, Billups SJ, Malone DC, Carter BL, Covey D, Mason B, et al. Types of interventions made by clinical pharmacists in the IMPROVE study. Pharmacotherapy: The Journal of Human Pharmacology and Drug Therapy. 2000;20(4):429-35.
7. Nicolau I, Tian L, Menzies D, Ostiguy G, Pai M. Point-of-care urine tests for smoking status and isoniazid treatment monitoring in adult patients. 2012.
8. Hanifa Y, Mngadi K, Lewis J, Fielding K, Churchyard G, Grant A. Evaluation of the Arkansas method of urine testing for isoniazid in South Africa. The international journal of tuberculosis and lung disease. 2007;11(11):1232-6.
9. Zhong B. How to calculate sample size in randomized controlled trials? Journal of thoracic disease.2009;1(1):51.

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**INFORMED CONSENT FORM**

Title of Study: ***(****An Insight on the Relationship Between the Variation in Medication Adherence Level and the Clinical Outcomes among Tuberculosis and Human Immunodeficiency Virus Co-Infected Patients****)***

By signing below, I confirm the following:

- I have been given oral and written information for the above study and have read and understood the information given.
- I have had sufficient time to consider participation in the study and have had the opportunity to ask questions and all my questions have been answered satisfactorily.
- I understand that my participation is voluntary and I can at any time freely withdraw from the study without giving a reason and this will in no way affect my future treatment. I am not taking part in any other research study at this time. I understand the risks and benefits, and I freely give my informed consent to participate under the conditions stated. I understand that I must follow the study doctor's (investigator's) instructions related to my participation in the study.
- I understand that study staff, qualified monitors and auditors, the sponsor or its affiliates, and governmental or regulatory authorities, have direct access to my medical record to make sure that the study is conducted correctly and the data are recorded correctly. All personal details will be treated as STRICTLY CONFIDENTIAL
- I will receive a copy of this subject information/informed consent form signed and dated to bring home.

**Subject:**

Signature: I/C number:

Name: Date: Phone Number:

**Investigator conducting informed consent:**

Signature: I/C number:

Name: Date:

**Impartial witness:** *(Required if subject is illiterate and contents of participant information sheet is orally communicated to subject)*

Signature: I/C number:

Name: Date: