TECHNOLOGY REVIEW (MINI-HTA)

BREATH TEST FOR TUBERCULOSIS

Malaysian Health Technology Assessment Section (MaHTAS)
Medical Development Division
Ministry of Health Malaysia
002/2024

IPT Mark
Mini-HTA
INAHTA

DISCLAIMER

This technology review (mini-HTA) is prepared to assist health care decision-makers and health care professionals in making well-informed decisions related to the use of health technology in health care system, which draws on restricted review from analysis of best pertinent literature available at the time of development. This technology review has been subjected to an external review process. While effort has been made to do so, this document may not fully reflect all scientific research available. Other relevant scientific findings may have been reported since the completion of this technology review. MaHTAS is not responsible for any errors, injury, loss or damage arising or relating to the use (or misuse) of any information, statement or content of this document or any of the source materials.

Please contact htamalaysia@moh.gov.my if further information is required.

EVIDENCE INFORMED DELIBERATIVE PROCESS

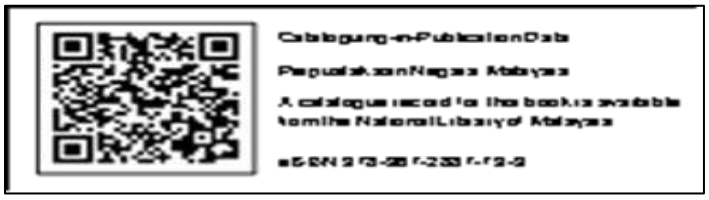
This Technology Review underwent an evidence informed deliberative process during the Health Technology Assessment (HTA) Technical Advisory Committee meeting, chaired by the Director, Medical Development Division, MOH. Then it was endorsed in the HTA-CPG Council meeting chaired by the Director General of Health.

Published by:

Malaysian Health Technology Assessment Section (MaHTAS)

Medical Development Division, Ministry of Health Malaysia, Level 4, Block E1, Precinct 1
Government Office Complex
62590, Putrajaya
Tel: 603 8883 1229
Available online via the official Ministry of Health Malaysia website: http://www.moh.gov.my

eISBN: 978-967-2887-79-9



SUGGESTED CITATION: Nur Hazlinda K, Roza S and Izzuna MMG. Breath Test to Diagnose Tuberculosis. Technology Review. Ministry of Health Malaysia: Malaysian Health Technology Assessment Section (MaHTAS); 2024. 28 p. Report No.: 002/2024

DISCLOSURE: The author of this report has no competing interest in this subject and the preparation of this report is entirely funded by the Ministry of Health Malaysia.

Prepared by

Nur Hazlinda binti Khalidi
Biochemist
Principal Assistant Director
Health Technology Assessment Section (MaHTAS)
Medical Development Division
Ministry of Health Malaysia

Reviewed by

Dr. Roza binti Sarimin
Public Health Physician
Head of HTA Unit
Health Technology Assessment Section (MaHTAS)
Medical Development Division
Ministry of Health Malaysia

Dr. Izzuna Mudla Mohamed Ghazali
Public Health Physician
Deputy Director
Malaysian Health Technology Assessment Section (MaHTAS)
Medical Development Division
Ministry of Health Malaysia

External reviewer(s)

Y.M. Dr Tengku Norita Tengku Yazid
Consultant Pathologist
Hospital Selayang
(Head of Pathology Specialty, Ministry of Health)

Dato' Dr Mat Zuki bin Mat Ja'eb
Consultant Respiratory Physician
Hospital Raja Perempuan Zainab II, Kota Bharu
(Head of Respiratory Medicine Specialty, Ministry of Health Malaysia)

EXECUTIVE SUMMARY

Background

Tuberculosis (TB) is a preventable disease caused by *Mycobacterium tuberculosis*, primarily affecting the lungs and spreading through the air. Despite being curable, TB was the second leading cause of death from a single infectious agent worldwide in 2022, after COVID-19, causing almost twice as many deaths as HIV/AIDS. Southeast Asia accounted for the largest proportion of TB cases in 2021. Malaysia is categorised as an intermediate TB burden country, and detection rates are still below WHO estimates, indicating a need for more proactive case finding, especially among high-risk groups.

Breath test analyse breath specimen in two ways, chemically and physically. Chemical technique in the analysis which identify chemical interaction between volatile organic compounds (VOCs) and sensor surface. Physical method quantify a molecule's physical characteristics such as size, shape and charge of the breath molecules. There is a need for a rapid, low-cost, easy-to-use diagnostic method to ensure effective screening and control. There is growing interest in the use of breath tests as a screening method for TB. This technology review aims to evaluate the effectiveness, safety of breath test in diagnosing TB following a request from a Pathologist (Medical Microbiologist) in Hospital Sultan Abdul Halim, Sungai Petani.

Objective/ aim

The objective of this technology review was to assess the accuracy, effectiveness, safety, and economic implication of breath test in diagnosing tuberculosis in patients suspected of having TB.

Results and conclusions:

Search results

A total of **301** records were identified through the Ovid interface, PubMed and other method. Two hundred and sixty-one duplicate references were found; **41** potentially relevant titles were screened using the inclusion and exclusion criteria. Of these, **29** relevant abstracts were retrieved in full text. After reading, appraising and applying the inclusion and exclusion criteria to the **29** full text articles, **seven** were included. **Eight articles** were excluded as those primary studies were already included in systematic review and meta-analysis (n=8), seven articles irrelevant study design (n=7) and seven articles found as irrelevant intervention (n=8). All full text articles finally selected for this review comprised of one systematic review and meta-analysis and six diagnostic accuracy studies. The studies were conducted mainly in South America (Paraguay, Venezuela), Africa (South Africa, Cameroon, Mali), Asia (Indonesia, India, China, Vietnam, Philippines), Bangladesh, Egypt and United Kingdom (UK).

Efficacy/ effectiveness

There was fair level of retrievable evidence on breath test showing its varying performance in diagnosing tuberculosis.

Breath test (electronic nose)
Breath test using electronic nose showed good pooled diagnostic accuracy. However, the performance of electronic nose was varied in four diagnostic accuracy studies included in this review. Sensitivity and specificity ranging from 52.3% to 90.8% and 36.4% to 99% respectively. The studies included in this review varies in terms of type of electronic nose used and population tested.

Breath test (other than electronic nose)
The performance of breath test other than electronic nose was good in diagnosing TB with sensitivity and specificity ranging from 80.4% to 95.7% and 80.3% to 91.3% respectively, and AUC performance ranged from 0.867 to 0.935.

Safety

There was limited evidence retrieved on the safety of breath test. Breath tests was not associated with treatment-related adverse events during the sessions or the follow-up. The United States Food and Drug Administration (US FDA) has not yet approved breath testing for tuberculosis. The electronic nos [redacted] device is claimed as having CE approval, indicating compliance with European regulatory standards.

Organisational

All the practices regarding the test analysis including the pre-examination, examination, post-examination, safety and training of staff or operator shall follow international or local guidelines such as Malaysian Standard, MS ISO 15189:2014, Specific Technical Requirement for Accreditation of Medical Microbiology Laboratories (STR 2.5) and National Policy and Guidelines for Point of Care Testing by Ministry of Health Malaysia.

Economic implication

No cost-effectiveness study retrieved on breath test to diagnose Tuberculosis. Nevertheless, the estimated price for laboratory-based automatic thermal desorption-gas chromatography-mass spectrometer [redacted] whereas a more accessible point-of-care device, using a portable gas chromatograph coupled to a surface acoustic wave detector (ATD-GC-SAW), is priced lower at around [redacted]

Conclusion

A fair level of retrieved evidences has demonstrated that point-of-care (POC) breath test has moderate to good diagnostic accuracy in detecting TB in high incidence setting.

No safety issue has reported with the use of this device. No cost effectiveness study retrieved on this device.

Methods

A systematic review was conducted. Following PRISMA, search strategy was developed by the main author while systematic search was conducted by an *Information Specialist* who searched for published articles on breath test to diagnose Tuberculosis. The following electronic databases were searched through the Ovid interface: MEDLINE(R) and Epub Ahead of Print, In-Process, In-Data-Review & Other Non-Indexed Citations, Daily and Versions 1946 to January 08, 2024, HTA Full-text Journals, EBM Reviews - Cochrane Database of Systematic Reviews 2005 to March 6, 2024, EBM Reviews - ACP Journal Club 1991 to February 2024, EBM Reviews - Cochrane Clinical Answers February 2024, EBM Reviews - Cochrane Central Register of Controlled Trials February 2024. Parallel searches were run in MISR (PubMed), US FDA and INAHTA database while additional articles were retrieved from reviewing the bibliographies of retrieved articles. The search was limited to articles on human. There was no language limitation in the search. The last search was conducted on 13th March 2024. Among the tools used to assess the risk of bias and methodological quality of the articles retrieved is the ROBIS and Critical Appraisal Skill Programme (CASP) cheklist. All full text articles were then graded based on guidelines from the US/Canadian Preventive Services Task Force.

TABLE OF CONTENTS

ABBREVIATION

AEs	Adverse events or adverse effects
AUC	Area under the ROC curve
AFB	Acid Fast Bacilli
BMI	Body mass index
CASP	Critical Appraisal Skills Programme
CI	Confidence interval
COPD	Chronic obstructive pulmonary disease
DM	Diabetes mellitus
ENose	Electronic nose
EPTB	Extra Pulmonary TB
GCMS	Gas chromatography mass spectrometry
HPPI-TOFMS	High-pressure photon ionization time-of-flight mass spectrometry
INAHTA	International Network of Agencies for Health Technology Assessment
LTBI	Latent TB Infection
LR	Likelihood ratio
MaHTAS	Malaysian Health Technology Assessment Section
MD	Mean difference
MOH	Ministry of Health
Mtb	*Mycobacterium tuberculosis*
NPV	Negative predictive value
NTB	Non-PTB
OR	Odds ratio
PCR	Polymerase Chain Reaction
POC	Point-of-care
PPV	Positive predictive value
PTB	Pulmonary TB
PTBX	Subjects with TB
QUADAS	Quality Assessment of Diagnostic Accuracy Studies
RCT	Randomised controlled trial
RD	Risk difference
RoB	Cochrane Risk of Bias Tool
ROBIS	National Collaborating Centre for Methods and Tools
TB	Tuberculosis
UI	Uncertainty Interval
US FDA	United States Food and Drug Administration
VOC	Volatile Organic Compound
WHO	World Health Organization

1.0 BACKGROUND

According to the World Health Organization (WHO), Tuberculosis (TB) is a preventable and usually curable disease. Tuberculosis (TB) is caused by bacteria (*Mycobacterium tuberculosis*) and it most often affects the lungs. [1] It spreads easily by air, with inhalation of infected droplets exhaled, coughed or sneezed by individuals affected with contagious forms of pulmonary TB. [1,2,3] Globally in 2022, the reported number of people newly diagnosed with TB was 7.5 million with an estimated 10.6 million (95% uncertainty interval [UI]: 9.9 to 11.4 million) people fell ill with TB worldwide, of which 5.8 million were men, 3.5 million were women and 1.3 million were children. There is still a large global gap between the estimated number of people who fell ill with TB and the number of people newly diagnosed, with approximately 3.1 million people not diagnosed with the disease, or not officially reported to national authorities in 2022. [8]

In 2021, the largest proportion of individuals who were affected by TB were concentrated in Southeast Asia, accounting for 45% of the cases based on geographical distribution. The number of TB cases in Malaysia has remained high for the past 30 years despite high cure rates that are achievable with a timely diagnosis and an appropriate antibiotic treatment. Malaysia falls into the intermediate category, with an estimated total TB incidence of 97 (range from 79 to 106) per 100,000 population in the year 2021 and this figure has increased to 113 cases per 100, 000 population in 2022. [13] Throughout the past few decades, the containment of TB has been an integral component of the strategic agenda outlined by the MOH Malaysia. The detection of TB cases in Malaysia is still below the estimated cases by the World Health Organization (WHO). Estimated incidence rate for Malaysia (2014) was 103 per 100,000 population, whilst actual achievement was 81 per 100,000 population, hence about 6000 cases were still undetected. Using predicted data between 1990-2010 and notification data between 1990-2014, the annual mean difference or underrepresentation is 13.5% (95% CI: 10.4 to 15.8). Therefore, more pro- active intensified case finding activities need to be planned. Attention should be given to ensure an effective systematic screening of high-risk group of getting TB and early diagnosis are implemented. [14]

Diagnostic delay is a major concern in TB control. There is an average loss of one to three months delay between the first day that patients present to the health care system, and the moment of diagnosis. [4,5]

The WHO criteria for pulmonary TB (PTB) diagnosis include clinical symptoms and isolation of *M. tuberculosis* from sputum by culture or by molecular assays as the line probe assays (LPAs), or detection of acid-fast bacilli by sputum smear microscopy (SSM) if culture or LPAs is unavailable, or for smear-negative PTB patients, with chest radiography (CXR) showing abnormalities consistent with active PTB.[16] Meanwhile, for extra-pulmonary TB (EPTB), the diagnosis is based on at least one specimen with confirmed *M. tuberculosis* or histological, or clinical evidence consistent with active EPTB, followed by a clinician's decision to treat with TB chemotherapy. Culture currently is the reference standard but the laboratory turn-around time is long (two to eight weeks). [16,17] The existing tests have high sensitivity, such as culture and PCR, or very high specificity, such as Gene Xpert, or moderate sensitivity, such as sputum smear microscopy. [12,17,18] However, these tests are not point-of-care, requiring sputum collection, trained staff and prone to laboratory error.

In this regard, it is imperative to create quick, affordable, and user-friendly techniques for diagnosing tuberculosis (TB) that can guarantee efficient field screening, prompt isolation of infected individuals, and management of treatment efficacy. Given its high sensitivity, ease of use, portable, point-of-care design, and portability, the breath test has gained interest to be employed as a screening for TB. [19]

Hence, this Technology review (TR) was requested by a Pathologist (Medical Microbiology), Hospital Sultan Abdul Halim, Sungai Petani to assess the feasibility and evidence on its accuracy to be used as one of the modalities in diagnosing Tuberculosis in Ministry of Health facilities.

2.0 OBJECTIVE / AIM

The objective of this technology review was to assess the accuracy, effectiveness, safety, and economic implication of breath test in diagnosing tuberculosis in patients suspected of having TB.

3.0 TECHNICAL FEATURE

Breath analysis provides potentially point-of-care, non-invasive sampling with minimal biosecurity risk, easy-to-perform, fast and broad applicability across various global contexts, including resource-constrained settings like community or primary care settings.
[27] Certain volatile organic compounds (VOCs) are produced by infections that alter the host metabolism, and *Mycobacterium tuberculosis* (MTB) generates many VOCs that are detectable by breath. [28–31] Breath specimen analysis can be done in two ways, chemically and physically. Chemical approaches include, for example, gas chromatography coupled with mass spectrometry (GC/MS), immunosensor and bio-optical technologies, and electronic-nose with sensors that detect chemical reactions between volatile organic compounds (VOCs) and the sensor surfaces. [36–37] Physical methods quantify a molecule's physical characteristics. For example, Field Asymmetric Ion Mobility Spectrometry (FAIMS) tracks the movement of breath's ionized molecules. [38] However, GC/MS requires complex equipment, operation skills, and a well-conditioned environment, especially to record concentration differences of VOCs specific for TB and different studies report different VOCs. [29-31, 39-41]

Volatile organic compounds (VOCs) are gaseous organic compounds that can arise from infections, host-pathogen interactions, and host metabolism. Both laboratory-based and real-time methods are available for detecting them. In the former case, the exhaled air is gathered, preserved, and pre-processed before being brought into the measurement device. [42]

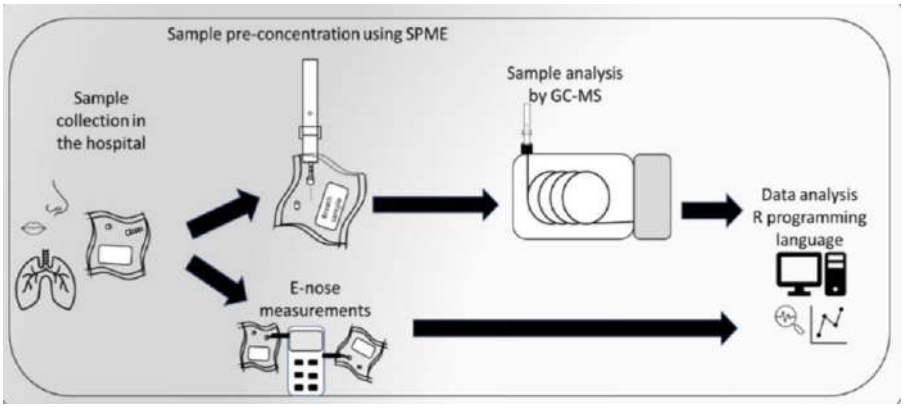


Figure 1: Overview of sample collection and analysis [48]

Analysers for exhaled breath can be separated in two groups:

Systems for detecting and measuring a set of specific VOCs (laboratory-based). Usually, these systems comprise of a mobile unit for collecting the gas sample and another (fixed) unit (analyser) for analysing it and separating the individual components. Separation techniques typically include GC-MS or IMS.

MASS SPECTROMETRY TECHNIQUES (Chemical technique)
Gas chromatography mass spectrometry (GC-MS) is considered the gold standard in breath research and uses preconcentrated breath samples for offline analysis. GC-MS is one type of breath analysis that use chemical technique in the analysis which identify chemical interactions between VOCs and the sensor surfaces. [19] GC-MS can be used for an untargeted approach, to provide chemical identification of the exhaled VOCs.[42,43] Other MS techniques, for example, selected ion flow tube MS, can perform online untargeted or targeted analysis of VOCs, following chemical ionization. They have the potential to be developed into point-of-care tests. [42] Unfortunately, its clinical use is not feasible since it is not real-time, needs a long time for sample processing, relies on non-portable devices, and needs constant calibration for specific analytes. Other MS-hybrid methodologies were also proved to be useful. [56,57]

Figure 2: [redacted]

ION MOBILITY SPECTROMETRY (IMS) (Physical Technique)
The IMS is a type of breath test that apply physical technique in the analysis. This technique measures the physical property of the breath's molecule which is it separates and identifies ionised molecules of breath based on their size, shape and charge and works by measuring the time it takes for ions to travel through a gas-filled chamber when pulled by an electric field.[19,42] Figure 3 illustrates a breath test that utilizes IMS technology, namely the [redacted] Breath Analyser. This device uses a dual polarity ion mobility spectrometer.

The procedure that the technology works is as follows: [50]

i. Breath samples are collected by seven to ten forced expirations onto a sample card
ii. The sample card is inserted into the desorber of Tuberculosis Breath Analyzer for data analysis.
iii. Results are displayed within 20 s as a Green Screen (TB negative subjects) or a Red Screen (TB positive subjects) on the display panel.

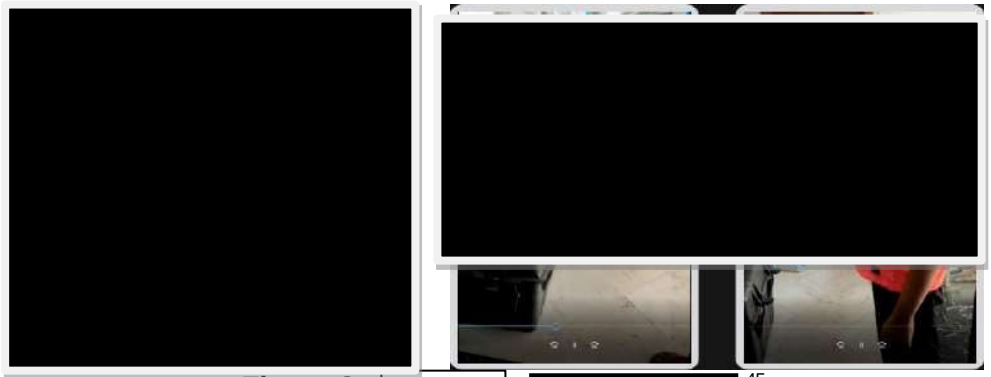


Figure 3: [redacted] 45

Electronic noses (real-time method and pattern-based technique), used for online breath analysis, often small, portable, potentially rapid diagnostic 'point-of-care' device which comprises an array of electronic chemical sensors and an appropriate pattern-recognition system, capable of recognizing simple or complex odors or gases in human breath. [20,25] This technology was designed to detect tuberculosis (TB) works by mimicking the human sense of smell to detect specific odor profiles or volatile organic compounds (VOCs) produced by *Mycobacterium tuberculosis*. Usually, electronic nose will come with nose clip and mouthpiece with filter and will connect to a laptop or computer with installed software for data analysis and result. The exhaled breath will be collected either in bags or tube or direct to the device lik[redacted][0,12,19] Electronic nose can identify different complex odors by comparing the incoming odor with previously learnt patterns by creating so called breathprints. [21] Readings occur when VOCs react at the surfaces of the eNose sensors, causing a change in conductivity of the sensors. [22] These are then detected by transducers and converted into electrical signals that create specific VOC signatures or in other word, eNose will compares vectors derived from VOCs from exhaled breath samples (breath data) to a machine learning algorithm to diagnose a patient with TB. [20, 25] For example, after measuring the breath using [redacted] the sensors are regenerated for 10 minutes and breath data are downloaded into a laptop or ipad with installed software, and uploaded onto the website of eNose for analysis. [12] The e-nose could provide results in just 15 min after the patient starts exhaling through the device and if the device is connected to an internet network. [54] Example of result showed were "TB yes" or "TB No" or "sick" or "healthy". [10,12] Several distinct eNose technologies have been developed such as the Aeonose, which uses micro hotplate metal-oxide sensors, the Common Invent eNose using metal oxide semiconductor sensors and the [redacted]sing cross-reactive metal-oxide semiconductor sensors. [22-24]

The simplified explanation of the mechanism was:

i. Sampling: The e-nose starts with sampling the air or breath of a person. In the case of TB detection, breath samples are commonly used because the bacteria can affect lung tissue and release specific VOCs into the breath.
ii. Sensor Array: The core of the e-nose is its sensor array, which consists of various sensors, each slightly different in its chemical sensitivity. These sensors react with the VOCs present in the sample. The reaction is usually a change in electrical properties (resistance, capacitance, or potential) that the sensor can detect. The sensors are designed to be broadly responsive to a range of compounds rather than highly specific to one molecule.
iii. Detection and Signal Processing: When the VOCs from the sample interact with the sensor array, they cause a change in the electrical properties of the sensors. This change is measured and converted into a digital signal for processing. The pattern of reactions across the sensor array creates a "fingerprint" of the sample.

iv. Pattern Recognition and Analysis: The digital signal is then analyzed using pattern recognition algorithms. These algorithms compare the fingerprint against a database of known profiles associated with TB infection. Machine learning techniques can be employed to improve the accuracy and reliability of detection over time, as the e-nose is exposed to more samples.

v. Output: Finally, the e-nose provides an output, often indicating whether the VOC profile matches that of a TB infection. This information can be used as a preliminary screening tool, suggesting further clinical testing if TB is indicated.

Figure 4: Examples of exhaled breath test : electronic nose [9,10,11]

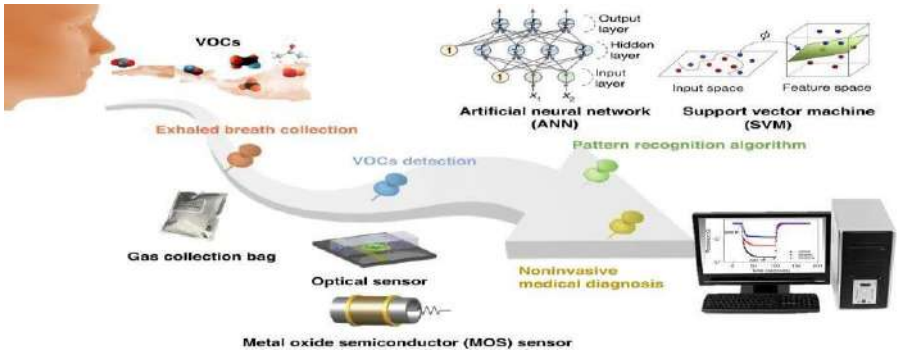


Figure 5: Schematic diagram of the noninvasive breath detection via the eNose system [49]

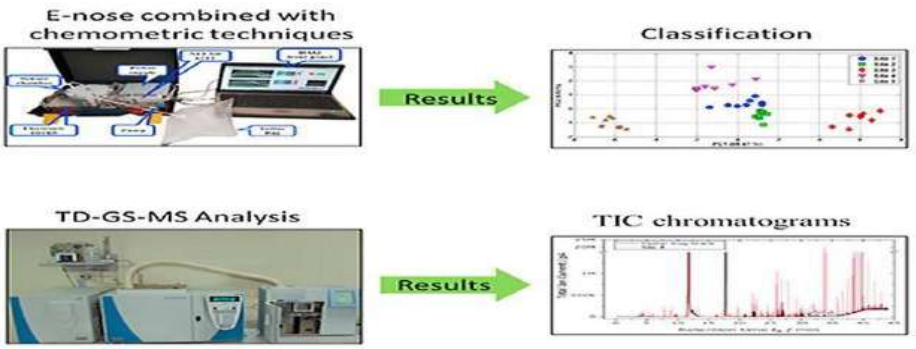


Figure 6: Example of electronic nose device and GC-MS analyser and the results diagram [44]

4.0 METHODS

A systematic review was conducted. Search strategy was developed by the main author and an Information Specialis.

4.1 SEARCHING

The following electronic databases were searched through the Ovid interface:

- MEDLINE(R) and Epub Ahead of Print, In-Process, In-Data-Review & Other Non-Indexed Citations, Daily and Versions <1946 to January 08, 2024>
- HTA Full-Text Journals
- EBM Reviews - Cochrane Database of Systematic Reviews <2005 to March 6, 2024>
- EBM Reviews - ACP Journal Club <1991 to February 2024>
- EBM Reviews - Cochrane Clinical Answers <February 2024>
- EBM Reviews - Cochrane Central Register of Controlled Trials <February 2024>

Other databases: PubMed, US FDA, INAHTA

General databases such as Google were used to search for additional web-based materials and information. Additional articles retrieved from reviewing the bibliographies of retrieved articles. The search was limited to articles on human. **Appendix 1** showed the detailed search strategies. The last search was conducted on 1st March 2024.

4.2 SELECTION

A reviewer screened the titles and abstracts against the inclusion and exclusion criteria. Relevant articles were then critically appraised depending on the type of the study design. Studies were graded according to *US/ Canadian Preventive Services Task Force* (**Appendix 2**). All data were extracted and summarised in evidence table as in **Appendix 3**.

The inclusion and exclusion criteria were:

Inclusion criteria:

a.	**Population**	Patients suspected of having TB
b.	**Intervention**	Breath test for TB
c.	**Comparator**	**1- Culture** **2- Line probe assays LPAs** **3- Detection of acid-fast bacilli by sputum smear microscopy** **4- Chest radiography (CXR)** **5- Rapid diagnostic test for TB –** for example: Xpert MTB, RIF

d.	**Outcomes**	**Diagnostic accuracy**: Sensitivity, specificity, accuracy **Effectiveness:** easy-to-perform, fast, feasible, convenient **Safety:** Adverse events (AEs) related to breath testing **Organisational issues:** procedural time, training **Economic implications:** Cost, cost-effectiveness, cost-utility analysis
e.	**Study design**	HTA reports, systematic review with/out meta-analysis, randomised controlled trial (RCT), cohort, diagnostic, case-control, economic evaluation studies
f.	Full text articles published in English	

Exclusion criteria:

a.	**Study design**	Case report, case series, animal study, laboratory study, narrative review
b.	Non-English full text articles	

5.0 RESULTS

Search results

An overview of the search is illustrated in **Figure 10**. A total of **301** records were identified through the Ovid interface, PubMed and other method. Two hundred and sixty-one duplicates references were found; **41** potentially relevant titles were screened using the inclusion and exclusion criteria. Of these, **29** relevant abstracts were retrieved in full text. After reading, appraising and applying the inclusion and exclusion criteria to the **29** full text articles, **seven** were included. **Eight articles** were excluded as those primary studies were already included in systematic review and meta-analysis (n=8), seven articles irrelevant study design (n=7) and seven articles found as irrelevant intervention (n=8). All full text articles finally selected for this review comprised of one systematic review and meta-analysis and six diagnostic accuracy studies. The studies were conducted mainly in mainly in South America (Paraguay, Venezuela), Africa (South Africa, Cameroon, Mali), Asia (Indonesia, India, China, Vietnam, Philippines), Bangladesh, Egypt and United Kingdom (UK).

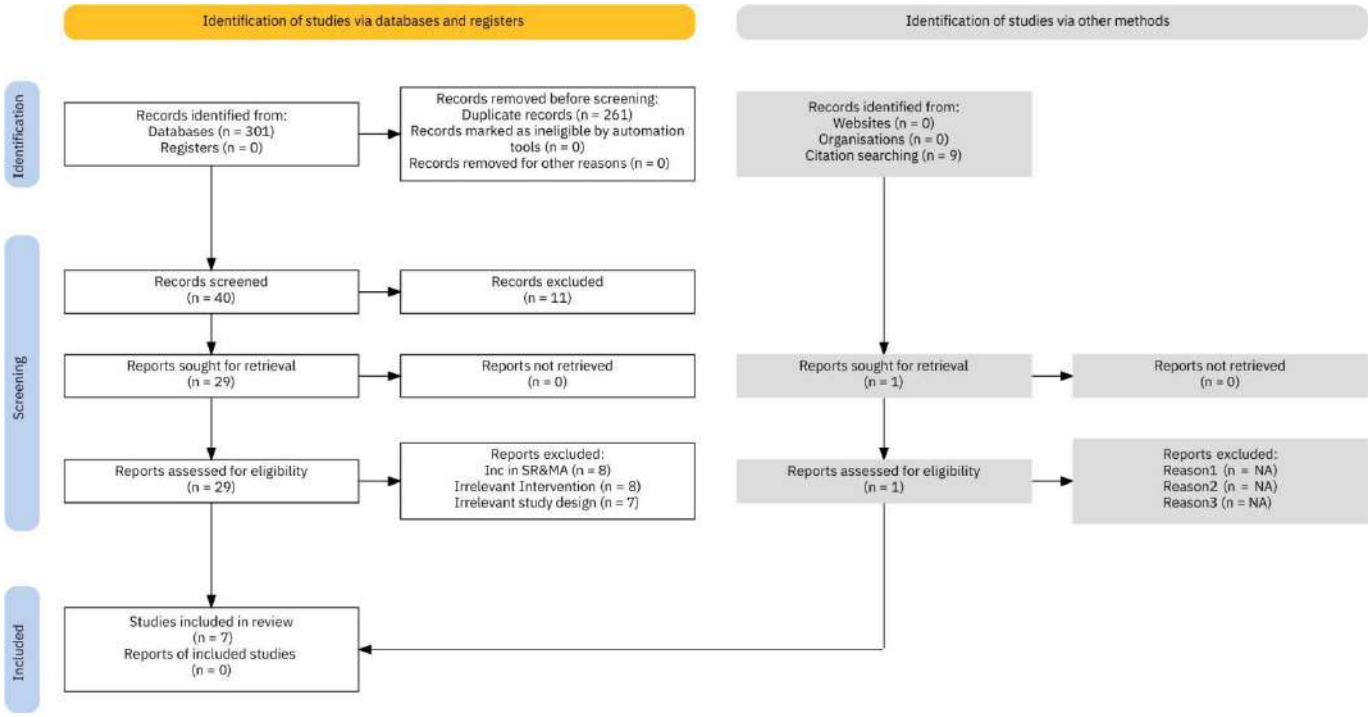


Figure 7: Flow chart of retrieval of articles used in the results [68]

Quality assessment of the studies

The risk of bias or quality assessment (methodology quality) of all retrieved literatures was assessed depending on the type of the study design. These assessments involved answering a pre-specified question of those criteria assessed and assigning a judgement relating to the risk of bias: using the relevant checklist of National Collaborating Centre for Methods and Tools (ROBIS) for systematic review and meta-analysis and Critical Appraisal Skill Programme (CASP) checklist for diagnostic test study. [59,60] The risk of bias of the included studies was assessed independently by two reviewers, NHK and RS. Any disagreements were resolved through discussion until consensus was reached. All full text articles were graded based on guidelines from the *U.S. / Canadian Preventive Services Task Force*. [61]

Risk of bias assessment for included systematic review and meta-analysis

One study was included in this assessment and was judged to have an overall low risk of bias following uncertainty in the data collection or risk of bias assessment process. (**Figure 7.1**).

Review	Risk of bias domains				
	1-STUDY ELIGIBILITY CRITERIA	2-IDENTIFICATION AND SELECTION OF STUDIES	3-DATA COLLECTION AND STUDY APPRAISAL	4-SYNTHESIS AND FINDINGS	RISK OF BIAS IN THE REVIEW
Saktiawati AMI et al. 2019 [19]	☺	☺	☺	☺	☺

Figure 7.1: Risk of bias assessment for systematic review and meta-analysis using ROBIS

Risk of bias assessment for included diagnostic accuracy study using CASP

Based on the CASP checklist, two studies had low risk of bias, two studies had unclear risk which is the studies did not clearly mention the blinding and the study protocol followed and other two studies had high risk of bias because of the studies did not involve all participants in index test and reference test analysis (**Figure 7.2**).

Studies	Risk of bias domains					
	D1	D2	D3	D4	D5	Overall
	Comparison with an appropiate reference standard	All patients get the diagnostic test and reference standard	Blinding	Disease status	Protocol followed	
Saktiawati AMI et al. 2019 [12]	☺	☺	☺	☺	☺	☺
Coronel Teixeira R et al. 2023 [62]	☺	☺	☺	☺	?	?
Ketchanji Mougang YC et al. 2023 [11]	☺	☹	?	☺	☺	☹
Coronel Teixeira R et al. 2021 [10]	☺	☹	☺	☺	?	☹
Badola M et al. 2023 [49]	☺	☺	?	☺	?	?
Fu L et al. 2023 [66]	☺	?	☺	☺	?	☺

Figure 7.2: Risk of bias assessment for diagnostic accuracy study using CASP

☺ low risk; ☹ high risk; ? unclear risk

5.1 EFFICACY/ EFFECTIVENESS

5.1.1 Diagnostic performance of breath test (electronic nose) to diagnose TB

A systematic review of the evidence and meta-analysis regarding breath test to diagnose TB was undertaken by Saktiawati AMI et al. (2019) to examine the accuracy of electronic-nose and other devices in diagnosing TB from patients' breath. The authors reviewed the diagnostic test accuracy (DTA) studies that assessed the sensitivity and specificity of electronic-nose and other devices in diagnosing TB in patients with TB or suspected of having TB. A total of 14 articles included in this SR and with that, six articles were based on electronic nose, eight articles use other than electronic nose as breath test to diagnose TB with a total of 1715 patients were included. Number of subjects analysed ranged from 40 to 251 per study. A total of four studies used PTB patients, PTB suspects and healthy controls as study subjects, three studies included newly diagnosed PTB patients, other studies involved asthma or chronic obstructive pulmonary disease (COPD) patients, tuberculous pleural effusion (TPE) suspects, human immunodeficiency virus (HIV) patients, patients with respiratory complaints and patients suspected to have EPTB. Only three studies included children (aged ≥ 13 years). These studies were performed by different medical centres in different countries. As the number of studies using breath test devices other than electronic- nose was insufficient for grouping, the statistical analysis was only calculated for the electronic-nose. The overall meta-analysis of the data revealed that the pooled sensitivity and specificity of electronic-nose in diagnosing TB were high and there was no heterogeneity among studies that diagnose TB with electronic nose. The **pooled sensitivity and specificity of electronic-nose** in diagnosing TB were 0·92 (95% CI: 0·82 to 0·97) and 0·93 (95% CI: 0·88 to 0·96). Respectively, the positive likelihood ratio (PLR), negative likelihood ratio (NLR), and diagnostic odd ratio (DOR) were 13·4 (95% CI:7·2 to 24·9), 0·08 (95% CI: 0·03 to 0·20), and 162 (95% CI: 41 to 634) respectively. The area under the curve (AUC) of electronic-nose was 0·97 (95% CI: 0·19 to 1·00). Meanwhile, the sensitivity and specificity of breath test devices other than electronic nose ranged from 0·62 to 1·00, and 0·11 to 0·84, respectively. The Q of electronic-nose in diagnosing TB were p = 0·23 indicating no statistically significant heterogeneity among the studies and I^2, 0 (95% uncertainty intervals: 0 to 100) suggesting no observed heterogeneity with a 95% uncertainty interval from 0% to 100%. [19, level II-2]

In a diagnostic study conducted by Saktiawati AMI et al. (2019), the author investigated the diagnostic potential of **electronic nose (Aeonose)** to identify PTB among patients with **suspected PTB** and factors associated with the sensitivity and specificity of the breath test. A cohort of 327 patients (≥18 years old) with suspected tuberculosis from the public Lung Clinics and Dr. Sardjito Hospital, Yogyakarta, Indonesia and healthy controls recruited from the neighboring area of subjects who were diagnosed with PTB were included. The study was conducted between October 2013 and December 2015. All patients were characterised by clinical symptoms, three sputum smear microscopic examinations, CXR and sputum culture and follow-up for 1.5 to 2.5 years after diagnosis. Participants were classified into active PTB, probably active PTB, no PTB or Healthy controls and probably no PTB. Demographic data were recorded. Participants breathed normally through the Aeonose for five minutes and measured by the device. Then, the sensors were regenerated for 10 minutes and the breath data were downloaded into a laptop and uploaded onto the website of eNose for analysis. The author started with a calibration phase to build breath model, involving participants in “active PTB” (n=85), “no PTB” (n=57), and “Healthy controls” (n=40)

groups. Since the electronic nose can only classify unknown patients correctly if the patient characteristics are similar to the ones in the calibration set to have fair classification, the author kept blinded the participants from the “Probably active PTB” (n=43) and “Probably no PTB” (n=102) groups during breath samples collection for validation purposes. The results of the calibration phase showed that the diagnostic accuracy was 82% (95% CI: 75 to 88), with sensitivity and specificity of 85% (95% CI: 75 to 92) and 55% (95% CI: 44 to 65), respectively. The result obtained in validation phase were, sensitivity of 78% (95% CI: 70 to 85), specificity of 42% (95% CI: 34 to 50), PPV of 52% (95% CI: 48 to 56), NPV of 71% (95% CI: 62 to 78) and AUC of 72% (95% CI: 66 to 78). This study revealed that the sensitivity was modest and specificity was low for Aeonose to diagnose TB among patients with suspected TB in Yogyakarta, Indonesia. The author also evaluated several factors that may associate with the breath prints, such as the physiological factors (age, sex, food, beverages) or pathological and disease-related conditions and found that the sensitivity and specificity of the breath test were lower in men (AUC=0.76) compared to women (AUC=0.93). The diagnoses for patients who turned out to have no PTB included asthma, pneumonia, bronchiectasis, chronic bronchitis, COPD, Obstructive Syndrome Post TB, lung fibrosis, lung abscess, empyema, and polycystic lung disease also reported in this study. [12, level II-3]

Another diagnostic study by Coronel Teixeira R et al. (2023) assess the accuracy of the **eNose** to classify individuals with **signs and symptoms compatible with TB** (in and outpatient clinic) referred to the Paraguayan National Reference Center for respiratory diseases and TB (INERAM). The study included a total of 107 participants aged ≥15 years, presenting with respiratory symptoms (cough, fever, dyspnoea, night sweat, lymphadenopathy and/or haemoptysis) for more than 15 days or having started anti-TB treatment already (<3 days before inclusion). Patients demographic and clinical data were recorded. Every participant underwent a physical examination, a CXR, a microbiological examination of their sputum or biopsies, and an eNose breath sample. Breath tests and microbiological examinations carried out on the same day. The gold standard for establishing a PTB diagnosis was a positive culture of the MTB complex. When a culture test yielded a negative result, the diagnosis was established by further compelling evidence (such as ZN positivity or GeneXpert detection of MTB). A diagnosis was referred to as a "clinical diagnosis" if no bacteriological evidence was discovered. In the last category, the first two months of anti-TB medication were used to assess how well clinical symptoms had improved. Patients with extra-pulmonary tuberculosis (EPTB) were identified either by histological or chemical evidence of tuberculosis (TB) in biopsies, or by microbiological confirmation (positive culture of MTB). The analysis of the breath samples was done by The eNose company using machine learning classifier, artificial neural network (ANN) or Neural Network (NN) and the result produced as “TB yes” or “TB no”. Univariate and multivariate logistic regression analyses were performed to assess potential risk factors for erroneous classification results by the eNose. The study found that TB was diagnosed in 91/107 (85%) of the participants which was 74% from that was established by gold standard, mycobacterial culture and another 26% was diagnosed by other microbiological examination and clinical diagnosis. The most common TB characteristic was cough (87.9%), fever (77.6%), dyspnoea (73.8%) and night sweat (72.0%). The **‘blind data set’** (n=76 participants), analysis resulted in low accuracy (50%), with sensitivity of 52.3% (CI 95%: 39.6 to 64.7%) and a specificity of 36.4% (CI 95%: 12.4 to 68.4%). The result obtained for PPV of 83.0% (CI 95%: 67.4 to
92.3%) with NPV of 11.4% (CI 95%: 3.7 to 27.7%). Risk factors for erroneous classifications by the eNose were **older age** (multivariate analysis: OR 1.55; 95% CI:1.10 to 2.18, p=0.012) and **antibiotic use** (multivariate analysis: OR 3.19; 95% CI: 1.06 to 9.66, p=0.040). The author reported that analysing larger cohorts will increase the accuracy of the eNose by

establishing a more robust neural network algorithm and the gold standard to establish TB diagnosis was not accomplished in all patients (mainly patients with EPTB) and it is possible that these patients in fact do not have active TB and the eNose classification was correctly made. [62, level II-3]

Ketchanji Mougang YC et al. (2023) conducted an interventional case control diagnostic study in the Center of Respiratory Disease of the public Laquintinie Hospital in Douala (Cameroon) involving 46 participants with **diagnosed TB**, 16 **suspected TB but affected by different diseases** and 38 healthy controls and all of them were over 18 years old. The author evaluated the use of an electronic nose made of porphyrinoid-functionalized quartz microbalance sensors. The study involved diagnosed TB patients, patients suspected to have TB but affected by different diseases, and a group of healthy controls. Using a sputum test with TB-LAMP and further clinical studies such chest X-rays and SMEAR, all suspected TB patients were identified as **non-PTB (had TB symptoms but negative TB- LAMP results)** or **subjects with TB (PTBX) (in spite of negative TB-LAMP results, the TB was confirmed by symptoms, chest X-ray, and physician outcome)**. After breath test analysis, the study participants were followed-up in time to confirm TB diagnosis**.** Measurements were collected in a period of two months. The healthy and TB subjects were assesed in a random sequence to avoid any effect of a possible drift of sensor signals. For each participant, demographic and clinical data and blood or urine laboratory tests were also collected. The non-parametric Kruskal-Wallis rank sum test was used to assess the statistical significance of sensor signals. Principal component analysis (PCA) and linear discrimination analysis (LDA), supervised algorithms that need to be trained and tested, were used in multivariate analysis. To allocate each measured sample to the TB and non-TB group in the machine learning model, classification performance metrics for LDA were obtained, including sensitivity, specificity, accuracy, and area under the ROC curve (AUC). The study found that from 100 breath samples were measured, there were **46 patients proven to be positive for pulmonary TB (PTB) with TB-LAMP** and **38 healthy controls (HC)**. Additional groups of **10 non-PTB (NTB)** subjects and **six subjects with TB (PTBX)** were measured to test the predictivity of EN on uncertain cases. Based on demographic and clinical data collected, all subjects of the PTB group presented cough symptoms (more than two weeks) and an average low BMI (19.5). Among the PTB group, 41.5% were patients who relapsed to PTB and who had been treated for six months with healing confirmation. All the recruited subjects were negative for antigenic COVID-19 tests. Based on distribution of the sensor signals in the two classes of PTB and HC, all sensors showed the mean of the frequency shift for the group of the PTB group is larger than that for the HC subjects. Hence, for all sensors the two classes are statistically different. The results from machine learning analysis show that the tested EN can detect all positive TB patients with respect to healthy controls with an accuracy result as in the table 1.

Table 1: Performance of the classification of PTB with respect to HC

	Accuracy	Sensitivity	Specificity	AUC
Training	89.8%	92.6 ± 10%	87.5 ± 11%	0.90
Test	88.0%	90.8 ± 17%	85.7 ± 18%	0.88

The model generated with the data collected from TB cases and healthy controls was applied to those patients who showed **symptoms** but had a **negative** TB-LAMP test and the performance achieved by the classifier between PTB and HC is confirmed with the cases for which the current diagnostic tools provided contradictory results.

Table 2: Results of the application of the LDA classifier to the negative TB-LAMP of symptomatic patients

	PTBX	NTB
Predicted as PTB	5	2
Predicted as HC	1	8

Based on sensitivities shown by all diagnostic tools utilised in the study, sensitivity of this **electronic nose** made of porphyrinoid-functionalized quartz microbalance sensors is comparable to TB-LAMP and chest-Xray, and largely outperforms SMEAR. [11, level II-3]

Another study conducted by Coronel Teixiera R et al. (2021) determined the **electronic nose** utility and accuracy as a **screening tool to detect TB** in an **indigenous population** in a **remote area**. This study has been conducted from November 2015 to February 2016 to identify TB in an isolated indigenous population in Livio Farina, Department of Alto Paraguay and a total of 131 of isolated indigenous population refer to Maskoy ethnic group adults (>15 years) were included. After signing informed consent, clinical data, medication, smoking habits and the last meal/drink schedule were recorded and then all included participants had a physical examination, CXR and five-minute breath sampling with electronic nose (Aeonose). Participants with anamnesis and/or abnormal CXR were asked to provide sputum samples for microbiological examinations. The analysis of the VOCs of individual breath using artificial neural network (ANN) was done by eNose company. The eNose analysis was performed in two stages: first, the training with a combination of a previous study population plus 47 participants from the new cohort (total n=153), and second, the 'blind prediction' of 84 participants. The results reported was the mean of age was 37.3 with 45.8% was male. Twenty-seven participants (21%) had symptoms and/or CXR abnormalities. All sputum samples from these participants were examined with ZN, GeneXpert MTB/RIF and culture- negative. One person was currently receiving TB treatment (pulmonary TB, third month). Three subjects were on antibiotic treatment (two with amoxicillin and one with azithromycin). Six subjects had just finished antibiotics from the penicillin group (ended > 24 hours before participating in the study). In the 12 months after the study was completed, no cases of active TB were diagnosed. The new ANN 'training model' yielded an area under the curve (AUC) of 0.90, a sensitivity of 87% (95% CI: 71 to 95) and a specificity of 92% (95% CI: 85 to 96). After training, the ANN was fixed and the remaining 84 blind 'smell prints' were then classified (blind prediction model). The specificity of the eNose was 99% (95% CI: 93 to 99) and the NPV was 100%, as only one participant was falsely classified as positive; this participant had no respiratory symptoms or CXR abnormalities. As **not a single community member in the blind prediction group had active TB**, the **sensitivity** and **positive predictive value** of the eNose **could not be calculated** from this cohort. [10, level II-3]

5.1.2 Diagnostic performance of breath test (other than electronic nose) to diagnose TB

Badola M et al. (2023) conducted a quasi-experimental pilot study in Pulmonology OPD of the Government Doon Medical College, India. This study assessed the diagnostic accuracy of a VOC-identification-based TB screening tool (**Breath Analyzer TSI-3000 (I),** Technoscan, Vaughan, Canada) against the gold standard of PCR-based TRUENAT/CBNAAT in terms of sensitivity, specificity, as well as positive and negative predictive values and examine the potential socio behavioral and epidemiological correlates that influenced the test results. The

study was conducted between July to December 2022 with 334 TB suspects (≥10 years) included. All TB suspects identified were recruited and were offered the index test after obtaining informed consent. All participants were further subjected to a diagnostic test, PCR-based TrueNat/CBNAAT procedure. The clients whose samples were **PCR-based TrueNat positive were considered as reference positive** subjects (gold standard), and those who were **PCR-based TrueNat negative** were considered **healthy subjects**. Breath samples were collected by seven to ten forced expirations onto the sample card, and then the sample card was inserted into the desorber of Tuberculosis Breath Analyzer for data analysis and results were displayed within 20 seconds as a Green Screen (TB negative subjects) or a Red Screen (TB positive subjects) on the display panel. The author reported the mean age of the study subjects was 35.8 years, 187 (55.9%) among them were males and 147 (44%) were females; total number of eligible participants was 334 and 139 (41.6%) of them were disease positive; 103 (30.83%) of the study subjects had some lesion present on chest Xray, 21 (6.28%) each were HIV positive and diabetics and 34 (10.1%) were currently smoking. The breath test result showed that 133 study subjects found as disease positive out of a total of 139 study subjects. These disease positives were determined by the gold standard; furthermore, the breath test identified 178 study subjects as disease negative out of the 195 disease negative subjects that were determined by the gold standard. The Tuberculosis Breath Analyzer identified active pulmonary TB with a **sensitivity of 95.7%** (95% CI: 90.8 to 94.8), the **specificity was 91.3%** (95% CI: 86.45 to 94.8), **the positive predictive value was 88.7%**, the **negative predictive value was 96.7%**, and the **ROC area was 0.935**. The **area under curve (AUC)** in the study findings was **0.935**. The sub-analysis data of diagnostic accuracy test (Table 3) showed that the tests showed considerable uniformity in terms of accuracy indicators (sensitivity, specificity, and predictive values) across the different age groups. However, the study analysis showed a relatively low indicator status in the age group of 30 to 44 years, and as much as 67.2% of the age group of 45 to 59 were negative for TB. The diagnostic accuracy of the Tuberculosis Breath Analyzer was found to be high for TB detection. [49, level II-3]

Table 3: Result of Diagnostic accuracy Tests.

Total	n	Sensitivity (95% CI)	Specificity (95% CI)	PPV	NPV	ROC Area
TP= 133 **FP = 17**	334	95.7% (90.8-98.4)	91.3% (86.4–94.8)	88.7%	96.7%	0.935
Age group (15-29) TP = 68 FP = 8 FN = 2 TN = 70	148	97.1% (90.1–99.7)	89.7% (80.8–95.5)	89.5%	97.2%	0.934
Age group (30-44) TP = 33 FP = 6 FN = 4 TN = 52	95	89.2% (74.6–97)	89.7% (78.8–96.1)	84.6%	92.9%	0.894
Age group (45-59) TP = 17 FP = 2 FN = 0 TN = 39	58	100% (80.5–100)	95.1% (83.5–99.4)	89.5%	100%	0.976

Fu L et al. (2023) conducted a diagnostic cross-sectional study to explore a novel, rapid, and simple LTBI detection method via breath test on **high pressure photon ionization time-of-flight mass spectrometry (HPPI-TOFMS**). The study was conducted at the Third People's Hospital of Shenzhen in Shenzhen, China, between March 2020 and November 2022 and 435 participants with inform consent signed involved. The **LTBI** group included the

participants who were contacts of ATB patients and had a positive IGRA result, with normal chest imaging and no evidence of ATB (n=185). The **control group** consisted of two main subcategories: 1) **ATB group** (n=121): ATB subjects in whom *M.tb* culture or GeneXpert TB-DNA was positive, and chest imaging was suggestive of ATB; 2) **healthy control (HC) group** (n=129): healthy subjects who came for physical examination and had no known contacts with ATB patients, with a negative IGRA result and a normal chest imaging. All participants were enrolled in the queue. The median age was significantly different (p<0.001) between the case and control groups; 41 and 28, respectively. There was no significant difference (p=0.397) in gender between the case and control groups. Breath samples were collected from 435 participants using a predefined protocol and sampling apparatus comprised a disposable gas nipple and a sampling bag made of polyether-ether- ketone (PEEK) then tested in developed High-pressure photon ionization time-of-flight mass spectrometry (HPPI-TOFMS) within twenty-four hours. All the enrolled participants were randomly split into three groups, 50% of them for model construction, 20% for internal validation and 30% for model blinded testing. A total of 92 LTBI patients and 123 controls were randomly selected as the discovery data set for Random Forest (RF) based LTBI detection model training, which was evaluated on internal validation dataset (37 LTBI patients and 51 controls) and blinded test dataset (56 LTBI patients and 76 controls). From the mass spectrum data produced by HPPI-TOFMS, the model-based feature selection was executed based on training and validation datasets, and the top ten VOC ions were selected according to the ranked feature importance. The study found that, in internal validation data set, with cut-off value of 0.5 (over 0.5 is considered LTBI), the LTBI detection model achieved good discrimination performance with sensitivity and specificity of 78.4% (95% CI: 63.4 to 94.3)
and 84.3% (95% CI: 74.3 to 94.3), AUC (0.91). While in the **test dataset (model blinded testing)**, the model performance slightly dropped, with the AUC decreasing from 0.91 (95% CI: 0.85 to 0.97) to 0.87 (95% CI: 0.81 to 0.93). The sensitivity and specificity achieved were
80.4% (95% CI: 69 to 92) and 80.3% (95% CI: 71 to 89) respectively. Since there were two subgroups in the controls, the author also evaluated the performances in discriminating LTBI with ATB and HC, respectively. The LTBI model performed better in discriminating LTBI and HC with an AUC of 0.95 (95% CI: 0.91 to 0.99) than in discriminating LTBI and ATB with an AUC of 0.78 (95% CI: 0.69 to 0.86). Besides, to evaluate the selected VOC ions in LTBI detection, the author trained the LTBI detection model on each single VOC ion and evaluated it in the test dataset. The discrimination of a single VOC ion was also good but limited (0.64 < AUC < 0.80), which is much inferior to the performance (AUC=0.87) of the combination of all ten VOCs. The study found that there were significant differences (p<0.001) among LTBI, HC, and ATB groups for almost all ten VOC ions, except for the VOC with mass-to-charge ratio (m/z) of 129 between LTBI and ATB (p=0.589). Figure 8 illustrates the patterns of these ten VOC ions that were visually different in ATB, LTBI, and HC groups. [66, level II-3]

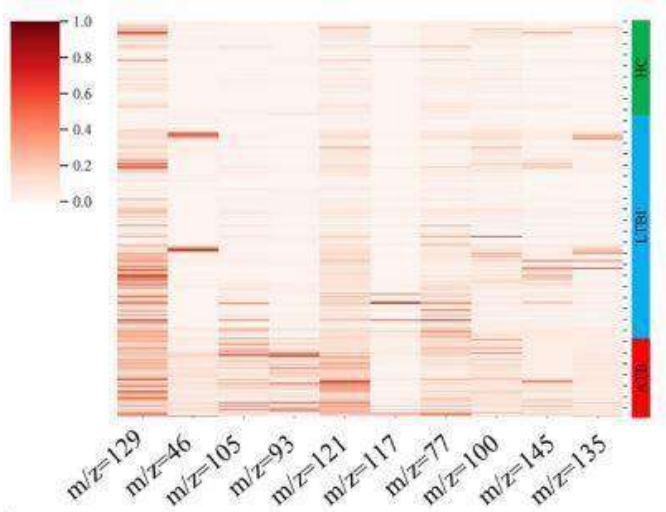


Figure 8: The heatmap of peak area distribution in all HC, LTBI, and ATB samples. [66]

Summary of studies related to the efficacy/ effectiveness of breath test to diagnose TB are shown in **Table 4**.

Table 4: Efficacy/ effectiveness of breath test to diagnose Tuberculosis reported by the included studies

Study	Patient characteristic/ disease	Follow-up duration	Intervention		Findings
			Treatment	**Comparison**	
Saktiawati AMI et al. 2019 SR & MA Bangladesh, South Africa, Egypt, Paraguay, Venezuela, Ethiopia, Vietnam, Philippines, United Kingdom, India, Mali	1715 PTB patients, PTB suspects and healthy controls, newly diagnosed PTB patients, involve asthma or COPD patients, TPE suspects, HIV patients, patients with respiratory complaints and suspect EPTB patients	-	Breath tests by electronic nose or other devices	Culture SSM Histopathology of the pleural biopsy CXR Gene Expert PCR Histology	▪ There is no heterogeneity among studies that diagnose TB with electronic nose (Q= p=0.23, I^2= 0; 95% uncertainty intervals: 0-100) ▪ The pooled sensitivity and specificity of electronic-nose in diagnosing TB were high (Sen= 92%, Spec= 93%, PLR= 13.4, NLR= 0.08, DOR= 162, AUC= 0.97) ▪ The sensitivity and specificity of breath test devices other than electronic nose ranged from 62% to 100% and 11% to 84%. (no pooled statistical data because of insufficient number for grouping)
Saktiawati AMI et al. 2019 Diagnostic Study Yogyakarta, Indonesia	327 Suspected PTB, Healthy controls	1.5 - 2.5 years	Electronic Nose (Aeonose ™) Sensor: metal oxide	Clinical diagnosis sputum smear CXR and sputum culture	▪ The sensitivity was modest and specificity was low for Aenose to diagnose TB among patients with suspected TB in Yogyakarta, Indonesia. (Sen= 78%, Spec= 42%, PPV= 52%, NPV= 71%, AUC= 72%) ▪ The sensitivity and specificity of the breath test were lower in men (AUC: 0.76) than women (AUC: 0.93) ▪ **Safety**: There were no adverse events associated with the study intervention.
Coronel Teixeira R et al. 2023 Diagnostic Study Paraguay, South America	107 aged ≥15 years, presenting with respiratory symptoms (cough, fever, dyspnoea, night sweat, lymphadenopathy and/or haemoptysis) for more than 15 days or having started anti-TB treatment already (< 3 days before inclusion)	Two months after anti-TB treatment (only for the cases which diagnosed by clinical diagnosis)	Electronic Nose (Aeonose ™) Sensor: metal oxide	Culture of MTB complex ZN staining GeneXpert MTB/RIF Histopathological or chemical evidence (biopsies, pus or pleural fluid) Clinical diagnosis	▪ TB was diagnosed in 91/107 (85%) of the participants which is 74% from that was established by gold standard, mycobacterial culture ▪ The most common TB characteristic was cough (87.9%), fever (77.6%), dyspnoea (73.8%) and night sweat (72.0%). ▪ The accuracy (AUC) of breath test was 50%, sensitivity was 52.3%, specificity was 36.4%, PPV and NPV was 83% and 11.4% respectively ▪ Older age and the use of antibiotics are significant risk factors for an incorrect prediction by the eNose with a very high OR for antibiotics use before TB diagnosis. (OR for older age; univariate= 1.50, multivariate= 1.55; OR for the use of antibiotics (other than anti- TB treatment) for more than 24 hours; univariate= 2.89,multivariate= 3.19) ▪ **Safety**: all false positive participants were admitted to the in patient clinic and later discharged with a final diagnosis of pneumonia ▪ **Safety**:The breath test measurements were done in the morning, always in the same place, in a room free of odours of gasses and alcohol and without dust. Cleaning of the eNose device on the outside was not performed unless it was needed because of stains on the device from a previous user. After every breath sampling a clean burn (approximately 10 minutes) was performed inside the device by heating of the metal sensors to 280 degrees Celsius, as part of the complete cycle process. Without this clean burn the eNose device cannot be used for another participant

Ketchanji Mougang YC et al. 2023 Diagnostic Study Cameroon, Central of Africa	100 TB patients, suspected TB (NTB & PTBX) and healthy controls, all over 18 years old	Two months	Electronic Nose (EN) Sensor: porphyrinoid-functionalized quartz microbalance sensors with corrole-based sensors along with a mass flow controller	TB-LAMP CXR } Suspected TB SMEAR } Suspected TB	▪ 46 patients proven to be positive for pulmonary TB (PTB) with TB-LAMP and 38 healthy controls (HC). Additional groups of 10 non-PTB (NTB) subjects and 6 subjects with TB (PTBX) were measured to test the predictivity of EN on uncertain cases ▪ For all sensors, the mean of the frequency shift for the group of the PTB is larger than that for HC subjects. All sensors show a *p*-value lower than 0.001. Hence, for all sensors, the two classes are statistically different ▪ The results show that the tested EN can detect all positive TB patients with respect to healthy controls with an accuracy of 88%, specificity and sensitivity of 85.7% and 90.8% and AUROC 0.88 respectively.
Coronel Teixeira R et al. 2021 Diagnostic Study Paraguay, South America	131 Isolated indigenous population refer to Maskoy ethnic group adults.	1 year	Electronic Nose (Aeonose ™) Sensor: metal oxide	Ziehl-Neelsen (ZN) stain, GeneXpert MTB/RIF (Cepheid) Mycobacterial Culture	▪ Twenty-seven participants (21%) had symptoms and/or CXR abnormalities. All sputum samples from these participants were ZN, GeneXpert MTB/RIF and culture-negative. One person was currently receiving TB treatment (pulmonary TB, third month). Three subjects were on antibiotic treatment (2 with amoxicillin/1 azithromycin). Six subjects had just finished antibiotics from the penicillin group (ended > 24 hours before participating in the study). In the 12 months after the study was completed**, no cases of active TB were diagnosed** ▪ As not a single community member in the blind prediction group had active TB, the sensitivity and positive predictive value of the eNose **could not** be calculated from this cohort. (Specificity= 99%, NPV= 100%)
Badola M et al. 2023 Diagnostic Study India	334 TB suspects	-	Breath test (Breath Analyzer TSI-3000 (I) (IMS method))	PCR-based TRUENAT/CBNAAT	▪ The diagnostic accuracy of the Tuberculosis Breath Analyzer was found to be high for TB detection (Sensitivity=95.7%, Specificity=91.3%, PPV=88.7%, NPV=96.7%, ROCAUC=0.935) ▪ The sub-analysis of the data showed that the tests showed considerable uniformity in terms of accuracy indicators (sensitivity, specificity, and predictive values) across the different age groups. However, the study analysis shows a relatively low indicator status in the age group of 30–44 years, and as much as 67.2% of the age group of 45–59 were negative for TB.
Fu L et al. 2023 Diagnostic Study Shenzhen, China	435 LTBI suspects and Controls group (ATB and healthy controls)	-	Breath test (High-pressure photon ionization time-of-flight mass spectrometry (HPPI-TOFMS))	Interferon-gamma release assays (IGRAs) CXR	▪ The age is significantly ($p<0.001$) different between the case and control groups (median= 41 and 28) ▪ In the internal validation dataset, the LTBI detection model achieved good discrimination performance with SEN and SPE of 78.4 (95% CI: 63.4%, 94.3%) and 84.3% (95% CI: 74.3%, 94.3%) , AUC (0.913). ▪ In blinded test dataset, the AUC decreasing from 0.913 (95% CI: 0.854, 0.972) to 0.867 (95% CI: 0.809, 0.925). The SEN and SPE achieved were 80.4% (95% CI: 68.7%, 92.0%) and 80.3% (95% CI: 71.3%, 89.2%) ▪ The LTBI model performed better in discriminating LTBI and HC with an AUC of 0.952 (95% CI: 0.909, 0.995) than in discriminating LTBI and ATB with an AUC of 0.777 (95% CI: 0.692, 0.861) ▪ For evaluation of selected VOC ions (ten ions) in LTBI detection, the box plot of peak area in all LTBI, HC and ATB samples show that the patterns of ten VOC ions were visually different in ATB, LTBI, and HC groups and there were significant differences ($p < 0.001$) among LTBI, HC, and ATB groups for almost all ten VOC ions, except for the VOC with m/z of 129 between LTBI and ATB ($p = 0.589$)

5.2 SAFETY

A study by Saktiawati AMI et al. (2019) reported that breath tests was not associated with treatment-related adverse events (e.g.breathless, infection, or bleeding) during the sessions or the follow-up. [12]

In a study conducted by Coronel Teixeira et al. (2021), participants with false-positive (n=9) and false-negative results (n=5) were analysed in depth. Two false positives had respiratory symptoms < 15 days: one of them with CXR with infiltrates and cavities and the other presented with overweight. The remaining seven false positives showed comorbidities varying from asthma, arterial hypertension and overweight. False negatives included two with culture-negative pleural TB (both used ceftriaxone before TB diagnosis was established) and two others had microbiologically proven pulmonary TB, also treated with antibiotics (penicillin group) for ≤ 3 days. The last one was obese. [10]

Several studies which using electronic nose as index test were use the disposable mouthpiece equipped with an antibacterial or antiviral filter such as HEPA filter during breath sampling to protect the electronic nose from getting contaminated by bacteria and viruses. [10,12,37] Saktiawati AMI et al (2019) in a diagnostic study reported that to prevent interference by VOCs in the environment such as ethanol and isopropanol, the Aeonose was equipped with a valve and carbon filter, thus the breath prints were not biased by the room air and inaccurate measurement can be avoided. [12]

Despite the clinical research and the science behind this simple, rapid, and point-of-care breath test that has been supported by several studies that have turned up with encouraging results, the FDA has not yet approved a breath test to diagnose TB, this means it is presently still seen as an investigational or experimental technology. Since TB has been recognized as one of the important infectious occupational disease affecting health care workers (HCWs), the safety procedure for healthcare worker during handling specimen and analysis should be included in study conducted. However, for Aeonose brand, the company claim that it was CE approved (IVDD 98/79/EC and MDD 93/42/EEC). [53, 58]

5.3 ORGANISATIONAL ISSUES

5.3.1 Treatment protocol

Turn around time

In a diagnostic accuracy study conducted by Badola M et al. (2023), which use VOC-identification-based TB screening tool (Breath Analyzer TSI-3000 (I) against the gold standard of PCR-based TRUENAT/CBNAAT, the author reported that once the data were analysed by the Tuberculosis breath analyzer the results were displayed within 20 s on the display panel. In another study by Phillips M et al. (2012) which evaluate breath VOC biomarkers in subjects with active pulmonary TB in Philipines, UK and India, using an internet-linked rapid point-of-care breath test (ATD-GC-MS), the breath test was completed in six minutes. Besides, in several studies of electronic nose such as study conducted by Saktiawati AMI et al. (2019) and Coronel Teixeira R et al. (2021,2023) using the electronic nose (Aeonose) as breath test to diagnose TB found that the measurement of electronic nose takes, in total about 15 minutes which include five minutes spent on

respiration procedure and 10 minutes are used for sensor regeneration and detecting possible low-concentrated VOCs. [10,12,54,63] These studies showed that the breath tests took less time for analysis than the current test practices such as culture's lab turnaround time (LTAT) which is one to eight weeks and PCR test's LTAT is one week. [19,62]

5.3.2 Training

No reagent needed for breath test electronic nose and breath test analyser TSI 3000(I) as compared to gold standard test like culture or PCR and chemical-analytical method. [50,63] However, the operation and handling of analysis software such as the solution model, graph and Artificial Intelligence programme in machine learning algorithm by breath analysis especially electronic nose may need access of internet and special training to the operator. [9,10,12,32,37,62]

According to the diagnostic study conducted by Saktiawati AMI et al. (2019), the author revealed that the electronic nose (Aeonose) was user-friendly and required no extensive training to provide non-invasive diagnosis in just a few of minutes. However, to operate and make sure more accurate result provided by an electronic nose, more participants are needed in the calibration phase to get comparable performance between calibration and validation phase. When an artificial neural network should predict a breath profile it hasn't 'seen' during the calibration phase, a false prediction is more likely. A larger calibration group would improve the blind predictions. Once the VOC-markers for TB are adequately determined, the use of highly selective sensors that target these VOC-markers may also add the sensitivity and specificity. [12]

Saktiawati AMI et al. (2019) compared the enose with chemical-analytical method (mass spectrometry). Although the enose offers less detailed information compared to spectroscopic or chemical-analytical method (mass spectrometry) in identifying volatile organic compounds (VOCs), it is still feasible to classify the data. Furthermore, there are a number of drawbacks to the chemical-analytical or spectroscopic techniques including the requirement of a well-conditioned environment, particularly when recording concentration differences of biomarkers, larger analyzer size prevents this method from being utilised as a point-of-care diagnostic test and operating the devices requires staff with the necessary training. [19] Moreover, Oliveira LF et al (2022) in a pilot study of breath analysis using electronic nose and gas chromatography-mass spectrometry on bronchial infections in bronchiectasis stated that the signal processing pipeline for GC-MS data is more complex than that of e-nose data. The large number of peaks, sometimes with strong coelution, baseline instabilities, and slight shifts in retention time leading to alignment problems, makes the whole data processing workflow a real challenge, particularly if in addition we have a limited supply of examples for the machine learning step. The GC-MS results obtained in this study revealed that, even though the use of experimental design and good analytical chemistry practices are essential, good validations techniques in the development of the models are key to avoid false discoveries in complex data. [65]

5.3.3 Guidelines

There were several studies on the use of national guidelines in their procedure. Zetola et al. (2017) reported that all cases included in the study were started on first-line anti-TB treatment (rifampicin, isoniazid, ethambutol and pyrazinamide) on the day of enrolment, in accordance to Botswana national TB guidelines. [35,64] According to study conducted by Coronel Teixeira R et al. (2023), the TB patients included in the study received anti-TB treatment according national guideline, National Guide for The Management of Tuberculosis: Health Services Local, District, Regional Health and Family Health Units: Asuncion: Ministry of Health Public and Social Welfare, Paraguay. Besides, Badola M et

al. (2023) also reported that all cases included in the study were treated in accordance with India national TB guidelines, on a first-line anti-TB treatment on the day of enrolment.[49,62]

All the practices regarding the test analysis including the pre-examination, examination, post-examination, safety and training of staff or operator shall follow international or local guidelines such as Malaysian Standard, MS ISO 15189:2014, Specific Technical Requirement for Accreditation of Medical Microbiology Laboratories (STR 2.5) and National Policy and Guidelines for Point of Care Testing by Ministry of Health Malaysia.

5.4 ECONOMIC IMPLICATION

There was no retrievable evidence on the cost-effectiveness or other economic analysis related to breath tests to diagnose Tuberculosis. However, based on the study conducted by Phillips M et al. (2012) which evaluated breath VOC biomarkers in subjects with active pulmonary TB, using an internet-linked rapid point-of-care breath test (**portable gas chromatograph** coupled to a surface acoustic wave (SAW) detector (ATD-GC-SAW)), reported that a **laboratory-based** automatic thermal desorption-gas chromatography- mass spectrometer (ATD-GC-MS) fully equipped for breath VOC analysis costs nearly $[redacted]

5.5 LIMITATIONS

We acknowledge some limitations in our review and these should be considered when interpreting the results. The selection of the studies was done by one reviewer. Although there was no restriction in language during the search, only the full text articles in English published in peer-reviewed journals were included in the report, which may have excluded some relevant articles and further limited our study numbers. Most of the studies included small sample sizes and the follow-up was relatively short, limited to approximately one year, which does not allow drawing any conclusions for a longer period of time.

6.0 CONCLUSION

A fair level of retrieved evidences has demonstrated that point-of-care (POC) breath test has moderate to good diagnostic accuracy in detecting TB in high incidence setting.

No safety issue has reported with the use of this device. No cost effectiveness study retrieved on this device.

7.0 REFERENCES

1. World Health Organization website. Available at: https://www.who.int/health-topics/tuberculosis#tab=tab_1 (Accessed on 22nd November 2023)
2. Global tuberculosis report 2020. Geneva: World Health Organization; 2020. Available at: https://apps.who.int/iris/ bitstream/handle/10665/336069/9789240013131-eng.pdf.
3. ynnycky E, Fine PE. The natural history of tuberculosis: the implications of age dependent risks of disease and the role of reinfection. Epidemiol Infect 1997 Oct; 119(2):183–201.
4. Cai J, Wang X, Ma A et al. Factors associated with patient and provider delays for tuberculosis diagnosis and treatment in Asia: a systematic review and meta-analysis. PLoS One 2015 Mar;10(3): e0120088.
5. Sreeramareddy CT, Panduru KV, Menten J et al. Time delays in diagnosis of pulmonary tuberculosis: a systematic review of literature. BMC Infect Dis 2009 Jun; 9:91.
6. WHO consolidated guidelines on tuberculosis. Module 2: screening – systematic screening for tuberculosis disease. Geneva: World Health Organization; 2021. Licence: CC BY-NC-SA 3.0 IGO.
7. High-priority target product profiles for new tuberculosis diagnostics: report of a consensus meeting, 28–29 April 2014, Geneva, Switzerland. Geneva: World Health Organization; 2014. Available at: https://apps.who.int/ iris/handle/10665/135617 (accessed on 19th February 2024)
8. World Health Organization (WHO). Global Tuberculosis Report 2023. Available at https://www.who.int/teams/global-tuberculosis-programme/tb-reports/global-tuberculosis-report-2023 (accessed on 19th February 2024)
9. Saktiawati AMI, Triyana K, Wahyuningtias SD et al. eNose-TB: A trial study protocol of electronic nose for tuberculosis screening in Indonesia. 2021. PLoS ONE 16(4): e0249689. https://doi.org/10.1371/journal.pone.0249689
10. Coronel Teixeira R, IJdema D, Gómez C et al. The electronic nose as a rule-out test for tuberculosis in an indigenous population. J Intern Med. 2021 Aug;290(2):386-391. doi: 10.1111/joim.13281. PMID: 33720468; PMCID: PMC8361912.
11. Ketchanji Mougang YC, Endale Mangamba LM, Capuano R et al. On-Field Test of Tuberculosis Diagnosis through Exhaled Breath Analysis with a Gas Sensor Array. Biosensors (Basel). 2023 May 22;13(5):570. doi: 10.3390/bios13050570. PMID: 37232931; PMCID: PMC10216756.
12. Saktiawati AMI, Stienstra Y, Subronto YW et al. Sensitivity and specificity of an electronic nose in diagnosing pulmonary tuberculosis among patients with suspected tuberculosis. PLoS One. 2019 Jun 13;14(6): e0217963. doi: 10.1371/journal.pone.0217963. PMID: 31194793; PMCID: PMC6563983.
13. Tuberculosis country profiles: Malaysia. Available at: https://www.who.int/tb/country/data/profiles/en/ (accessed on 1st March 2024).
14. Ministry of Health Malaysia: National strategic plan for tuberculosis control (2016-2020). Disease Control Division (TB/ Leprosy Sector) Ministry of Health Malaysia, Malaysia; 2016. Available at: https://www.moh.gov.my/index.php/dl/554756755a584a6961585268626939515a5778686269425464484a686447566e615773674c7a4977 (accessed on 23rd February 2024)
15. Hickmann KS, Fairchild G, Priedhorsky R et al. Forecasting the 2013-2014 influenza season using Wikipedia. PLoS Comput Biol. 2015, 11: e1004239. 10.1371/journal.pcbi.1004239

16. World Health Organization. Treatment of tuberculosis guidelines 4th edition (internet). (cited 2016 Apr 4). Available from: http://www.who.int/tb/publications/2010/9789241547833/en/; 2010
17. World Health Organization. Global tuberculosis report 2018 (internet). Geneva; (cited 2018 Sep 27). Available from: http://apps.who.int/iris/bitstream/handle/10665/274453/9789241565646-eng.pdf?ua=1
18. Bruins M. Transferable odor differentiation models for infectious disease diagnostics. Rotterdam: Erasmus MC University Medical Center; 2014.
19. Saktiawati AMI, Putera DD, Setyawan A et al. Diagnosis of tuberculosis through breath test: A systematic review. EBioMedicine. 2019 Aug; 46:202-214. doi: 10.1016/j.ebiom.2019.07.056. Epub 2019 Aug 8. PMID: 31401197; PMCID: PMC6712009
20. Gardner JW, Bartlett PN. A brief history of electronic noses. Sens. Actuators B Chem. 1994, 18, 210–211
21. Persaud K, Dodd G. Analysis of discrimination mechanisms in the mammalian olfactory system using a model nose. Nature 1982. 299, 352–355.
22. Van de Goor R, Van Hooren M, Dingemans AM et al. Training and Validating a Portable Electronic Nose for Lung Cancer Screening. J. Thorac. Oncol. 2018. 13, 676–681.
23. Bos LDJ, Van Walree IC, Kolk AHJ et al. Alterations in exhaled breath metabolite-mixtures in two rat models of lipopolysaccharide-induced lung injury. J. Appl. Physiol. 2013, 115, 1487–1495.
24. De Vries R, Muller M et al. Prediction of response to anti-PD-1 therapy in patients with non-small-cell lung cancer by electronic nose analysis of exhaled breath. Ann. Oncol. 2019, 30, 1660–1666.
25. Licht J-C, Grasemann H. Potential of the Electronic Nose for the Detection of Respiratory Diseases with and without Infection. International Journal of Molecular Sciences 2020; 21(24):9416. https://doi.org/10. 3390/ijms21249416 PMID: 33321951
26. Haworth J, Pitcher C, Ferrandino G et al. Breathing new life into clinical testing and diagnostics: perspectives on volatile biomarkers from breath. Critical Reviews in Clinical Laboratory Sciences 2022; 1 (20):353–72. https://doi.org/10.1080/10408363.2022.2038075 PMID: 35188863
27. Nathavitharana RR, Yoon C, MacPherson P et al. Guidance for studies evaluating the accuracy of tuberculosis triage tests. J. Infect. Dis. 2019, 220, S116–S125.
28. Bijland LR, Bomers MK, Smulders YM. Smelling the diagnosis a review on the use of scent in diagnosing disease. Netherlands J Med 2013; 71:300–7.
29. Syhre M, Manning L, Phuanukoonnon S et al. The scent of Mycobacterium tuberculosis—part II breath. Tuberculosis (Edinb) 2009 Jul;89(4):263–6.
30. Syhre M, Chambers ST. The scent of Mycobacterium tuberculosis. Tuberculosis (Edinb) 2008 Jul;88(4):317–23.
31. Phillips M, Cataneo RN, Condos R et al. Volatile biomarkers of pulmonary tuberculosis in the breath. Tuberculosis 2007 Jan;87(1):44–52.
32. Phillips M, Basa-Dalay V, Blais J et al. Point-of-care breath test for biomarkers of active pulmonary tuberculosis. Tuberculosis 2012 Jul;92(4):314–20.
33. Nakhleh M, Jeries R, Gharra A et al. Detecting active pulmonary tuberculosis with a breath test using nanomaterial-based sensors. Eur Respir J 2014;43(5):1522–5.
34. McNerney R, Wondafrash BA, Amena K et al. Field test of a novel detection device for Mycobacterium tuberculosis antigen in cough. BMC Infect Dis 2010;10.
35. Zetola NM, Modongo C, Matsiri O et al. Diagnosis of pulmonary tuberculosis and assessment of treatment response through analyses of volatile compound patterns in exhaled breath samples. J Infect 2017; 74(4):367–76.
36. Bruins M, Rahim Z et al. Diagnosis of active tuberculosis by e-nose analysis of exhaled air. Tuberculosis 2013 Mar;93(2): 232–8.

37. Coronel Teixeira R, Rodríguez M, Jiménez de Romero N et al. The potential of a portable, point-of-care electronic nose to diagnose tuberculosis. J Infect 2017;75(5):441–7.
38. Sahota AS, Gowda R, Arasaradnam RP et al. A simple breath test for tuberculosis using ion mobility: a pilot study. Tuberculosis 2016; 99:143–6 Internet. Available from https://doi.org/10.1016/j.tube.2016.05.005.
39. Mgode GF, Weetjens BJ, Nawrath T et al. Mycobacterium tuberculosis volatiles for diagnosis of tuberculosis by Cricetomys rats. Tuberculosis 2012;92(6):535–42 Internet. Available from https://doi.org/10.1016/j.tube.2012. 07.006.
40. Phillips M, Basa-Dalay V, Bothamley G et al. Breath biomarkers of active pulmonary tuberculosis. Tuberculosis (Edinb) 2010 Mar;90(2):145–51.
41. Beccaria M, Mellors TR, Petion JS et al. Preliminary investigation of human exhaled breath for tuberculosis diagnosis by multidimensional gas chromatography – Time of flight mass spectrometry and machine learning. J Chromatogr B Anal Technol Biomed Life Sci 2018:1074–5 Internet. (December 2017):46–50. Available from: https://doi.org/10.1016/j.jchromb.2018. 01.004.
42. Ahmed WM, Lawal O, Nijsen TM, et al. Exhaled volatile organic compounds of infection: a systematic review. ACS Infect Dis. 2017; 3:695–710.
43. Van de Kant KD, van der Sande LJ, Jobsis Q, et al. Clinical use of exhaled volatile organic compounds in pulmonary diseases: a systematic review. Respir Res. 2012; 13:117.
44. Moufid M, Bouchikhi B et al. Assessment of outdoor odor emissions from polluted sites using simultaneous thermal desorption-gas chromatography-mass spectrometry (TD-GC-MS), electronic nose in conjunction with advanced multivariate statistical approaches. Atmospheric Environment. Volume 256. 2021, ISSN 1352-2310. https://doi.org/10.1016/j.atmosenv.2021.118449. Available at: https://www.sciencedirect.com/science/article/abs/pii/S1352231021002703. Accessed online on 12th March 2024.
45. [redacted] Available at: https://teknoscan.com/2021/05/16/teknoscan-tsi-3000-breath-analyser-for-covid-19/. Accessed online on 12th March 2024.
46. Sakumura Y, Koyama Y, Tokutake H et al. Diagnosis by Volatile Organic Compounds in Exhaled Breath from Lung Cancer Patients Using Support Vector Machine Algorithm. Sensors (Basel). 2017 Feb 4;17(2):287. doi: 10.3390/s17020287. PMID: 28165388; PMCID: PMC5335963**.**
47. Oliveira LF, Mallafré-Muro C, Giner J et al. Breath analysis using electronic nose and gas chromatography-mass spectrometry: A pilot study on bronchial infections in bronchiectasis. Clin Chim Acta. 2022 Feb 1;526:6-13. doi: 10.1016/j.cca.2021.12.019. Epub 2021 Dec 23. PMID: 34953821.
48. Li, Y, Wei X, Zhou Y et al. Research progress of electronic nose technology in exhaled breath disease analysis. *Microsyst Nanoeng* 9, 129 (2023). https://doi.org/10.1038/s41378-023-00594-0.
49. Badola M, Agrawal A, Roy D et al. Volatile Organic Compound Identification-Based Tuberculosis Screening among TB Suspects: A Diagnostic Accuracy Study. Adv Respir Med. 2023 Jul 12;91(4):301-309. doi: 10.3390/arm91040024. PMID: 37489387; PMCID: PMC10366871.
50. Van Der Schee MP, Paff T, Brinkman P et al. Breathomics in lung disease. *Chest.* 2015; 147:224–231. doi: 10.1378/chest.14-0781.
51. Chen D, Bryden et al. Detection of Tuberculosis by The Analysis of Exhaled Breath Particles with High-resolution Mass Spectrometry. *Sci Rep* 10, 7647 (2020). https://doi.org/10.1038/s41598-020-64637-6.
52. [redacted] Available online at: https://www.enose-company.com/wp-content/uploads/2021/04/manual_EN-1.pdf. Accessed online on 14th March 2024.

53. Mohamed N, Van De Goor R, El-Sheikh M et al. Feasibility of a Portable Electronic Nose for Detection of Oral Squamous Cell Carcinoma in Sudan. *Healthcare*. 2021; 9(5):534. https://doi.org/10.3390/healthcare9050534.
54. Mohamed EI, Mohamed MA, Moustafa MH et al. Qualitative analysis of biological tuberculosis samples by an electronic nose based artificial neural network. Int J Tuberc Lung Dis 2017;21(7):810–7.
55. Rattray NJ, Hamrang Z, Trivedi DK et al. Taking your breath away: Metabolomics breathes life in to personalized medicine. Trends Biotechnol. 2014, 32, 538–548.
56. Beale DJ, Jones OAH, Karpe AV et al. A Review of Analytical Techniques and Their Application in Disease Diagnosis in Breathomics and Salivaomics Research. Int. J. Mol. Sci. 2016, 18, 24.
57. Ministry of health Malaysia. Guidelines on Prevention and Management of Tuberculosis for Health care Workers in Ministry of Health Malaysia. Available at: https://www.moh.gov.my/moh/images/gallery/Garispanduan/Guidelines%20On%20Prevention%20And%20Management%20of%20Tuberculosis%20For%20HCWs%20In%20MOH.pdf. Accessed online on 14th March 2024.
58. ROBIS: Risk of Bias in Systematic Reviews. Available at https://www.bristol.ac.uk/population-health-sciences/projects/robis/. Accessed on 27th February 2024.
59. Critical Appraisal Skills programme. Available at https://casp-uk.net/casp-tools-checklists/. Accessed on 06th March 2024
60. Harris RP, Helfand M, Woolf SH et al. Current Methods of the U.S. Preventive Services Task Force: A review of the process. Am J Prev Med. 2001; 20(30): 21-35.
61. Ministry of Health, Malaysia. Diagnosis Makmal Bagi Penyakit TB. Available at: http://www.myhealth.gov.my/diagnosis-makmal-bagi-penyakit-tb/. Accessed online on 18th March 2024.
62. Coronel Teixeira R, Gómez L, González E et al. The accuracy of an electronic nose to diagnose tuberculosis in patients referred to an expert centre. PLoS One. 2023 Feb 7;18(2): e0276045. doi: 10.1371/journal.pone.0276045.
63. Botswana National Tuberculosis Programme Report. 2009.
64. Oliveira LF, Mallafré-Muro C et al. Breath analysis using electronic nose and gas chromatography-mass spectrometry: A pilot study on bronchial infections in bronchiectasis. Clin Chim Acta. 2022 Feb 1; 526:6-13. doi: 10.1016/j.cca.2021.12.019. Epub 2021 Dec 23. PMID: 34953821.
65. Scheme, N.T. Ministry of Health and Family Welfare, Government of India. Available online: https://reports.nikshay.in/
66. Fu L, Feng Y, Ren T et al. Detecting latent tuberculosis infection with a breath test using mass spectrometer: A pilot cross-sectional study. Biosci Trends. 2023 Mar 11;17(1):73-77. doi: 10.5582/bst.2022.01476
67. Haddaway NR, Page MJ et al. PRISMA2020: An R package and Shiny app for producing PRISMA 2020-compliant flow diagrams, with interactivity for optimised digital transparency and Open Synthesis Campbell Systematic Reviews, 18, e1230. https://doi.org/10.1002/cl2.1230.
68. Munekata PES, Finardi S, de Souza CK et al. Applications of Electronic Nose, Electronic Eye and Electronic Tongue in Quality, Safety and Shelf Life of Meat and Meat Products: A Review. *Sensors*. 2023; 23(2):672. https://doi.org/10.3390/s23020672
69. World Health Organisation, Global Tuberculosis Report. Incidence of Tuberculosis (per 100, 1000 people). Available at https://data.worldbank.org/indicator/SH.TBS.INCD?locations=MY. Accessed on 14th June 2024.

8.0 APPENDIX

APPENDIX 1: LITERATURE SEARCH STRATEGY

Ovid MEDLINE(R) and Epub Ahead of Print, In-Process, In-Data-Review & Other Non-Indexed Citations, Daily and Versions <1946 to January 08, 2024>

1. TUBERCULOSIS/ (114993)
2. (mycobacterium tuberculosis adj2 infection*).tw. (5449)
3. Koch* disease.tw. (11)
4. tuberculos?s.tw. (215870)
5. 1 or 2 or 3 or 4 (248204)
6. BREATH TESTS/ (16427)
7. (breath adj1 test*).tw. (8438)
8. (breathalyzer adj1 test*).tw. (48)
9. 6 or 7 or 8 (20258)
10. 10 5 and 9 (81)

Other Databases	
EBM Reviews - Cochrane Database of Systematic Reviews <2005 to March 6, 2024>	Similar MeSH, keywords, limits used as per MEDLINE search
EBM Reviews - Database of Abstracts of Reviews of Effects <1st Quarter 2016>	
EBM Reviews - Cochrane Central Register of Controlled Trials <February 2024>	
PubMed	Similar MeSH, keywords, limits used as per MEDLINE search
INAHTA	
US FDA	

APPENDIX 2: HIERARCHY OF EVIDENCE FOR EFFECTIVENESS

DESIGNATION OF LEVELS OF EVIDENCE

I	Evidence obtained from at least one properly designed randomized controlled trial.
II-I	Evidence obtained from well-designed controlled trials without randomization.
II-2	Evidence obtained from well-designed cohort or case-control analytic studies, preferably from more than one centre or research group.
II-3	Evidence obtained from multiple time series with or without the intervention. Dramatic results in uncontrolled experiments (such as the results of the introduction of penicillin treatment in the 1940s) could also be regarded as this type of evidence.
III	Opinions or respected authorities, based on clinical experience; descriptive studies and case reports; or reports of expert committees.

SOURCE: US/CANADIAN PREVENTIVE SERVICES TASK FORCE (Harris 2001)

APPENDIX 3: EVIDENCE TABLE

Only available upon request.

e ISBN 978-967-2887-79-9

MaHTAS
(online)