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SUPPLEMENT ARTICLE

A Blueprint to Address Research Gaps in the Development of Biomarkers for Pediatric Tuberculosis

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Childhood tuberculosis contributes significantly to the global tuberculosis disease burden but remains challenging to diagnose due to inadequate methods of pathogen detection in paucibacillary pediatric samples and lack of a child-specific host biomarker to identify disease. Accurately diagnosing tuberculosis in children is required to improve case detection, surveillance, healthcare delivery, and effective advocacy. In May 2014, the National Institutes of Health convened a workshop including researchers in the field to delineate priorities to address this research gap. This blueprint describes the consensus from the workshop, identifies critical research steps to advance this field, and aims to catalyze efforts toward harmonization and collaboration in this area.

Keywords. tuberculosis; children; diagnosis; biomarker; blueprint.

Childhood tuberculosis is estimated to account for 6% of the tuberculosis caseload globally, and for 4%–21% of the caseload in the 22 high-incidence countries that account for 80% of global tuberculosis cases [1]. Mathematical modeling suggests that only 35% of tuberculosis cases in children are detected [2]. Improving the accuracy of tuberculosis diagnosis in children is required to improve case detection and outcomes, surveillance, efficiency of healthcare delivery, future research, and effective advocacy.

However, attaining an accurate diagnosis in children in tuberculosis-endemic settings remains challenging. There is overlap of the clinical presentation of tuberculosis with other common childhood diseases such as pneumonia, human immunodeficiency virus (HIV)–associated lung disease, and severe malnutrition [3]. Clinical and chest radiographic features are often nonspecific and subject to variable interpretation [4]. Structured diagnostic scoring systems based on clinical and radiological findings and tuberculin skin testing show high variability in case yield, with poor agreement between scoring systems [5]. Microbiological confirmation is possible in children of all ages, but is rarely attempted due to perceived difficulties in obtaining respiratory specimens

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and because both culture [6] and automated real-time nucleic acid amplification tests are only positive in a proportion of children who have been clinically diagnosed with tuberculosis [7–9]. Current diagnostics that measure immunological responses following infection with *Mycobacterium tuberculosis* have uncertain sensitivity and are unable to distinguish active tuberculosis from latent tuberculosis [10]. The clinical distinction between latent and active tuberculosis is unlikely to be dichotomous, especially following recent infection—a common scenario in children.

In 2014, the National Institutes of Health (NIH) of the United States convened a group of panelists to develop a blueprint for the process of discovery and implementation of new diagnostic biomarkers for pediatric tuberculosis. In the 19th century, a "blueprint" was a reproduction of a technical drawing through a contact print process on light-sensitive sheets that allowed the rapid and accurate reproduction of documents but was unable to reproduce color or shades of gray. This article shares several similarities with the original blueprint method, inasmuch as it builds on existing efforts for pediatric diagnostic biomarker discovery, qualification, validation, and implementation, but does not dictate the exact approach in view of the rapidly changing technological, regulatory, and health implementation realities. The blueprint presented here outlines the issues facing the field of pediatric tuberculosis biomarker development and is aligned with the Stop TB Partnership and the World Health Organization (WHO) International Roadmap for Tuberculosis Research [11]. The blueprint covers the following critical steps:

- Defining and prioritizing which biomarkers are needed
- How to select the most appropriate markers for qualification and validation
- New and potential future technologies
- Study design considerations
- Collection and storage of suitable specimens
- Biorepository considerations
- Regulatory challenges

The target audience for this article includes researchers, healthcare providers, funding agencies, and regulatory bodies, in an effort to coordinate and streamline the challenging process of pediatric tuberculosis diagnostic biomarker discovery, validation, qualification, and implementation.

CONSENSUS STATEMENT PREPARATION

Wide consultation was sought in the development of this blueprint with input from international experts from relevant clinical, basic science, public health and regulatory fields, and other stakeholders. Among the panelists specifically included were pediatric tuberculosis clinicians, tuberculosis researchers, HIV research network representatives, ethicists, representatives from research funding agencies, and individuals from nongovernmental, advocacy, and community research organizations. Panelists were invited to a workshop entitled "Pediatric Tuberculosis: Addressing Research Gaps in Diagnostic TB Biomarkers" organized by the NIH in Bethesda, Maryland, in May 2014. Prior to the workshop, conference calls were held to identify key questions for discussion at the workshop. A table summarizing existing pediatric (and major adult) specimen repositories was developed (Table 1). During the workshop, there were timed discussions, including break-out groups, with statement modification in real time. The statements were reviewed in the subsequent plenary sessions. Agreement was reached by consensus or by vote.

The consensus questions covered 3 key areas relating to pediatric tuberculosis biomarker research:

1. What are the criteria for the optimal pediatric tuberculosis biomarkers?
2. What are the challenges and sustainability issues for a pediatric tuberculosis specimen repository?
3. What are the custodianship, ownership, legal, regulatory, and policy issues relating to such repositories?

This document, which captures the consensus from the workshop, aims to generate further discussion about pediatric tuberculosis biomarker research, and to catalyze efforts toward harmonization and collaboration. An NIH-sponsored Pediatric TB Biomarker Working Group has been established to assist in moving this process forward.

THE CHARACTERISTICS OF AN "IDEAL" BIOMARKER FOR TUBERCULOSIS IN CHILDREN

A biomarker (or set of biomarkers) that could be used to develop an accurate test for tuberculosis in a child would ideally fulfill the following requirements:

- Measurable in a readily obtainable matrix such as blood (eg, by fingerstick), urine, stool, saliva, buccal mucosal transudates, or exhaled air given the challenges in obtaining respiratory specimens from infants and young children (<5 years).
- Identify *M. tuberculosis* with high sensitivity and specificity, independent of age, nutritional status, or HIV status as the cause of, or contributing factor to, the current illness in children presenting clinically with pulmonary or extrapulmonary tuberculosis.
- Distinguish between children with latent tuberculosis (including children with latent tuberculosis who have respiratory symptoms due to another pathogen) and those with active tuberculosis disease.
- Suitable for incorporation into a diagnostic platform that would provide a rapid, accurate result at, or close to, the point of care.

Table 1. Repositories From Existing Studies Evaluating Tuberculosis Biomarkers

Reference Sample Biorepository	UCT Pediatric TB	Kenya Pediatric TB	Pneumonia Etiology Research for Child Health TB	Grand Challenges in Global Health GC6-74 Biomarkers for TB	Aeras TB	European Union Action for Diseases of Poverty, Diagnostics Consortium–TB	Foundation for Innovative New Diagnostics–TB[a]	Consortium for Tuberculosis Biomarkers–TB
Enrollment type(s)	P	P	P	A, P	A, P	A, P	A	A
HIV status, Positive or negative	Both	Both	Both	Both	Both	Both	Both	Both
Clinical categories	Suspected TB cases (n = 1800)	Children <5 y of age with (1) Suspected TB (n = 300), further categorized as definite, probable, possible, unlikely, no TB; (2) asymptomatic children (n = 100)	(1) Children with severe and very severe pneumonia and (2) healthy community controls	(1) Newly diagnosed adult pulmonary TB cases, (2) HHCs: adults with TB disease and healthy controls, (3) adolescents (non-HHC) including progressors with TB disease	Vaccine trial cohorts	(1) Suspected TB cases and (2) TB contacts further categorized as TB disease (culture positive, probable or possible TB), non-TB diseases, or healthy LTBI	Suspected pulmonary TB further categorized as (1) TB disease including (i) Smear and culture positive; (ii) smear negative, culture positive; (2) clinical diagnosis only, with response to treatment (chest radiograph), (3) not TB	Newly diagnosed pulmonary TB. Culture-confirmed TB disease specimens from clinical trials: TB Alliance, CDC TB Trials Consortium, and AIDS Clinical Trials Group
Countries included	South Africa	Kenya	South Africa, Zambia, Kenya, The Gambia, Mali, Thailand, Bangladesh	South Africa, The Gambia, Malawi, Uganda, Ethiopia	Sub-Saharan Africa	South Africa, Kenya, Malawi	Bangladesh, Brazil, Moldova Peru, South Africa, Vietnam, Zimbabwe. TDR/WHO TB Specimen Bank samples: Bangladesh, Brazil, Canada, Colombia, Kenya, Peru, South Africa, Spain, The Gambia, Uganda, Vietnam	To date: South Africa, Kenya, Uganda
Subject follow-up duration	6 mo if TB treatment given, 2 mo if TB treatment not given	6 mo or until TB treatment completion. Evaluations at 0, 0.5, 2, 6 mo)	Up to 30 d after hospital discharge	24 mo (HHC evaluations at 0, 6, 18 mo, index case evaluations at 0, 12 mo).	Up to 24 mo	6 mo	2–3 mo	Samples collection at treatment initiation, weeks 2, 4, and 8, months 4, 6, and 12. Also at time of relapse or withdrawal from study

Table 1 continued.

Reference Sample Biorepository	UCT Pediatric TB	Kenya Pediatric TB	Pneumonia Etiology Research for Child Health TB	Grand Challenges in Global Health GC6-74 Biomarkers for TB	Aeras TB	European Union Action for Diseases of Poverty, Diagnostics Consortium–TB	Foundation for Innovative New Diagnostics–TB[a]	Consortium for Tuberculosis Biomarkers–TB
Type of samples	Whole-blood (EDTA tube), whole blood (PAXgene tube), serum, induced sputum, NP swab, urine, stool, selected extrapulmonary specimens	Whole blood (PAXgene tube), plasma, serum, QFT supernatant, nasopharyngeal and oropharyngeal (NP/OP) swabs, gastric aspirate, urine, stool	Whole blood, nasopharyngeal and oropharyngeal (NP/OP) swabs, induced sputum or gastric aspirate (if no sputum; cases only), pleural fluid (cases only), lung aspirates (cases only), urine, postmortem lung needle biopsy	Serum, plasma, PBMC, RNA, DNA. Most samples stored at field sites, selected samples at UCT central repository	PBMC, whole blood, plasma, serum, urine	Whole blood (PAXgene tubes), plasma, throat swabs	Plasma (EDTA), plasma (P800), serum, sputum, saliva, urine	Whole blood (PAXgene tube), whole blood (EDTA), whole blood in QFT tubes (nil, mitogen, TB antigen), sputum, spot urine
Diagnostic gold standard	MGIT culture of 2× induced sputum or Xpert of respiratory specimen.	MGIT culture and Xpert MTB/RIF (suspected TB cohort, per participant): NP aspirate (2), induced sputum (2), gastric aspirate (2), string test (2), urine (2), stool (2); MGIT culture on blood (1)	TB culture	TB culture (solid and liquid), concentrated Ziehl-Neelsen microscopy post-NALC-NaOH.	TB culture	TB culture	TB culture (solid or liquid), concentrated Ziehl-Neelsen microscopy post-NALC-NaOH	TB culture (solid or liquid)
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Table 1 continued.

Reference Sample Biorepository	UCT Pediatric TB	Kenya Pediatric TB	Pneumonia Etiology Research for Child Health TB	Grand Challenges in Global Health GC6-74 Biomarkers for TB	Aeras TB	European Union Action for Diseases of Poverty, Diagnostics Consortium–TB	Foundation for Innovative New Diagnostics–TB[a]	Consortium for Tuberculosis Biomarkers–TB
Website	TBD	TBD	http://www.jhsph.edu/research/centers-and-institutes/ivac/projects/perch/	http://www.biomarkers-for-tb.net/consortium/the-consortium	http://www.aeras.org/	TBD	http://www.finddiagnostics.org/programs/tb/find_activities/tb_specimen_bank.html	www.tbbiorepository.org

Blank fields indicate no data available.

Abbreviations: A, adult; HHC, household contact; EDTA, ethylenediaminetetraacetic acid; HIV, human immunodeficiency virus; LTBI, latent tuberculosis; MGIT, mycobacteria growth indicator tube; NALC, N-acetyl-L-cysteine; NaOH, sodium hydroxide; NP, nasopharyngeal; OP, oropharyngeal; P, pediatric; PBMC, peripheral blood mononuclear cell; PI, principal investigator; QFT, Quantiferon-TB; SATVI, South African Tuberculosis Vaccine Initiative; TBD, to be determined; TDR/WHO, World Health Organization Special Programme for Research and Training in Tropical Diseases; UCT, University of Cape Town.

[a] Also includes the TDR/WHO specimen samples. The WHO Tuberculosis Strain Bank has been transferred to the Institute of Tropical Medicine, Antwerp, Belgium.

PRIORITIES FOR PEDIATRIC TUBERCULOSIS BIOMARKER DEVELOPMENT/VALIDATION

There is a substantial gap between the requirements for an ideal pediatric tuberculosis biomarker and currently available biomarkers. To close this gap, the following priority areas for research were identified:

1. Biomarkers for the diagnosis of active tuberculosis disease among symptomatic children (both HIV-infected and uninfected) should be considered top priority.
2. The optimal biomarker should enable diagnosis of active tuberculosis disease in all children; however, the highest priority should be given to young children aged 0–5 years who are known to have the greatest burden of disease [12], are at highest risk for developing severe disease, with associated morbidity and mortality, and are the age group in which diagnostic confirmation of disease is most challenging.
3. Both pulmonary and extrapulmonary tuberculosis should be targeted for new diagnostics, but the priority should be given to pulmonary tuberculosis given the disease burden and diagnostic challenges highlighted above.

NEW AND POTENTIAL FUTURE TECHNOLOGIES

There has been progress in identifying host and pathogen biomarkers with diagnostic potential as well as in the development, optimization, and integration of new and current technologies. This has resulted in rapid population of the diagnostic pipeline for tuberculosis [13]. Improvements in current nucleic acid amplification (NAAT) technologies include the development of highly sensitive next-generation NAAT platforms [14] and new portable, battery-operated NAAT platforms with built-in communication technologies to speed up the communication of results [15, 16], which could be suitable for low-resource settings. Other promising pathogen detection methods include molecular detection of small fragments of tuberculosis-specific transrenal DNA [17, 18] and fluorogenic enzymatic tests for the specific detection of BlaC [19]. The latter is a highly conserved and specific class A β-lactamase naturally expressed and secreted by *M. tuberculosis*. These new technologies are particularly important because they utilize readily available specimen types or amplify signal from organisms that are scarce in specimens due to the paucibacillary nature of pediatric tuberculosis.

Other diagnostic approaches not relying on pathogen detection are also emerging. For example, microRNAs (miRNAs) have been shown to modulate the pathogenesis of tuberculosis infection, disease, and treatment response, and such studies have already been conducted in children. Studies evaluating the potential for diagnostics based on miRNA profiles in serum, peripheral blood immune cells, and sputum have

reported that specific miRNAs show the potential to discriminate infected from healthy individuals, and active from latent infection, and to be useful for monitoring response to treatment [20]. Similarly, pediatric studies have demonstrated that genomewide host transcriptional RNA signatures in blood can distinguish tuberculosis from other diseases and from latent tuberculosis infection, and that risk scores based on gene expression may be useful for ruling out tuberculosis [21]. It has also been recently shown that a T-cell activation marker present on circulating *M. tuberculosis*–specific T cells can discriminate active from latent infection in children [22]. Given the rapid emergence of new technologies, there is a need to better define the strategy for biomarker selection and validation.

KEY DESIGN FEATURES FOR BIOMARKER STUDIES

Diagnostic studies in children should be standardized with regard to the key elements of study design such as eligibility criteria, radiological and microbiological assessments, specimen collection and storage, data collection and analysis, clinical care, and clinical case definitions. The Pediatric TB Biomarker Working Group is currently developing consensus guidelines in this regard. Case definitions of disease should preferably follow the NIH consensus definitions for diagnostic study categories for intrathoracic tuberculosis [23], which have recently been revised to define 3 categories of tuberculosis disease: (1) confirmed tuberculosis (microbiologically confirmed); (2) unconfirmed tuberculosis (formerly possible or probable tuberculosis); (3) unlikely tuberculosis [24].

Ideally, prospective cohort studies with adequate follow-up to monitor response to therapy should be undertaken to enable accurate, standardized collection of data and specimens. However, leveraging existing cohorts and biorepositories from well-characterized cohorts (eg, Table 1) will also be of considerable value. Children with a wide range of symptoms and clinical manifestations should be included in studies to cover the full spectrum of tuberculosis disease. Studies should be done in different epidemiological settings that include a broad range of frequent nontuberculosis conditions (eg, malnutrition, HIV-associated infections, bacterial pneumonia, malaria) as differential diagnoses. Detailed historical, clinical, laboratory, and radiological characteristics should be collected to enable standardized case definitions. A standardized data dictionary and specimen collection template would be useful to enable meta-analysis of data from different studies, cohorts, and repositories, and should be developed by pediatric tuberculosis researchers.

Study design should include follow-up of all children, with and without tuberculosis infection or disease, not only to allow collection of sequential specimens, but also to strengthen the case definitions for tuberculosis, by evaluating response to therapy in children treated or not treated for tuberculosis. The suggested follow-up times for children treated for tuberculosis disease include visits at 2 weeks, 2 months, and 6 months or treatment completion and a visit at 2 months for children not treated for tuberculosis disease.

COLLECTION AND STORAGE OF SUITABLE SPECIMENS

A key component of study design is the choice of specimen type and collection method. Specimens should be collected bearing in mind that both pathogen and host biomarker detection strategies may be used, including, for example, proteomic, metabolomic, and gene expression profiling. In children with pulmonary tuberculosis, *M. tuberculosis* has been isolated from a variety of respiratory and alimentary tract specimens, and potential diagnostic biomarkers can also be detected in blood products and urine [7, 8, 21, 25–28]. Selection of the most suitable specimen types for a pediatric tuberculosis diagnostic biomarker study should take into account the issues of feasibility, acceptability, effectiveness, and cost of the collection procedure under programmatic conditions. In general, this would include blood (eg, serum, whole blood for RNA), respiratory specimens (eg, induced or spontaneously expectorated sputum, nasopharyngeal aspirate or swab, gastric aspirate/lavage, string test), urine, and other extrapulmonary specimens (when clinically indicated). To facilitate comparison of results between biomarker studies and thereby increase their usefulness, it is important to standardize specimen collection, handling, processing, and storage across studies.

There are many considerations to take into account to assure standardization. Collection of specimen types should be well documented, including anatomical location (eg, nasopharyngeal/oropharyngeal/laryngopharyngeal); specimen content (eg, "pure" aspirate vs diluted lavage/wash); collection vehicle (eg, swab tip vs suction catheter); swab type (eg, cotton vs synthetic tip); additives used (eg, antimicrobial agent, nucleic acid stabilizer, buffer); and volume accepted and processed, as volume can significantly impact assay sensitivity. Handling, processing, and storage of specimens vary by type, and considerations for standardization include pH (eg, neutralization based on initial pH); transport time, especially for nonsterile specimens; centrifugation parameters; and refrigeration and storage temperature. Ideally, standard operating procedures for the collection of each specimen type should be collaboratively developed, piloted and refined, and consistently implemented.

Stored specimens may be used not only for tuberculosis biomarker discovery and detection, but, as improved diagnostics for other respiratory pathogens become available, may also be tested for such pathogens to improve the specificity of clinical case definitions. Research protocols, reviewed by institutional

review boards, need to incorporate specific consent for storage and future testing of samples.

DEVELOPING A SUSTAINABLE BIOREPOSITORY

The biomarker discovery and validation process would be facilitated by the availability of biorepositories containing well-characterized and appropriate pediatric specimens. Given the substantial upfront effort and cost involved in carefully characterizing a symptomatic cohort by clinical criteria and in confirming a case of tuberculosis in a child, it is particularly important to maximize the benefit associated with this initial investment by establishing repositories for pediatric specimens. At present, tuberculosis-specimen biorepositories are focused on samples from adults, or are in the hands of individual investigators (Table 1), with little coordination and standardization. A key outcome of the workshop was an initiative to develop a shared pediatric repository.

Key questions in designing a repository are: What is the scientific objective of the work and what are the potential biomarker targets of interest? Scientific objectives, for example, could include discovery of new diagnostics for tuberculosis disease or of markers of treatment response and will guide decisions about whether to focus more on specimen collection at baseline vs longitudinal specimen collection [29]. Biomarker targets of interest will guide decisions about which specimens to collect, how to store specimens, and whether initial processing is required prior to biobanking. Practical questions to be addressed in repository planning are: (1) How many aliquots of each specimen to biobank and in what volume, based on considerations of known vs unknown potential applications, cost, and clinical limitations (eg, blood volume)? (2) Which specimens should be prioritized in case it is not possible to collect all specimens from every participant? and (3) What are the time points for specimen collection during longitudinal sampling?

The technical differences between methods used to collect, transport, process, and store specimens in studies may be contributing factors to variation in the published accuracy of diagnostic markers [30]. Data collection and process harmonization are therefore critical for interpretability and comparability of results. Detailed data on the specifics of specimen collection are necessary to determine what potential targets are likely preserved in a sample. Similarly, linkage to clinical metadata is needed to identify the potential suitability of specific samples for studies and the interpretation of study findings.

A large challenge to the integrity of repositories is the requirement for meticulous record-keeping. Potential solutions to record-keeping challenges include the use of barcode labeling, electronic databases to indicate the location of each sample and to link sample data to clinical metadata, and segregation of samples by specimen type and aliquot number to facilitate easy access at a later time.

As a part of first steps, it was determined at the workshop that a Data Sharing Framework needed to be created with agreed-upon standard procedures and pediatric tuberculosis nomenclature, and a Pediatric TB Biomarkers Working Group is being organized by NIH, including the authors of this manuscript, other tuberculosis experts, and microbiologists. This group is currently developing standard nomenclature and operating procedures for pediatric specimen collection.

The establishment, maintenance, and custodianship of a biorepository will require dedicated funding. Funding of a biorepository requires careful planning as costs for specimen collection, handling, and processing for long-term storage, and for data management and storage, are often not included in the initial budget of research studies, and specific funding sources are currently limited. Appropriate funders or partners will need to be identified and key partnerships should be sought with existing networks, such as the International Maternal, Pediatric, Adolescent AIDS Clinical Trial Network (IMPAACT) and foundations with experience in running biorepositories, such as the Foundation for Innovative New Diagnostics (FIND), as well as potential funders such as the NIH and the Bill & Melinda Gates Foundation. Considering that prior experience has shown that the creation and maintenance of such repositories is a large and expensive undertaking, entities such as those listed above could pool resources and each fund parts of the repository creation and maintenance. Funders may be more amenable to provide resources when existing specimens are pooled together in a single repository, as opposed to generating de novo repositories or funding individual repositories. A repository team would be needed to plan the establishment of the repository in detail. This blueprint can be seen as part of the lobbying exercise to facilitate such funding and may stimulate the participation of other collaborators. Funding would have to be for a period of at least 5 years, with clear milestones and deliverables, culminating in a sample release phase during which researchers could apply for sample release to facilitate their test development work. A long-term plan for sustainability should be developed, which may include contributions from researchers wishing to access samples.

To encourage investigators to submit specimens to biorepositories, funders could consider making submission of samples a component of funding opportunities or incentivizing submission through the provision of supplementary funding.

REGULATORY CHALLENGES FOR PEDIATRIC BIOREPOSITORIES

Custodianship of a biorepository involves the provision of careful oversight and management of samples from the point of specimen collection all the way to the research use of biospecimens and

the linked data. A plan for selection of an appropriate custodian, with explicit details on the responsibilities, should be developed prior to the launch of a biobanking initiative. Policies need to be in place to safeguard the quality of samples being entered into the repository and their long-term storage. Additionally, policies are needed for the appropriate use of biospecimens and data, while assuring the privacy and confidentiality of participants and their data. Research data standards, such as those provided by the Clinical Data Interchange Standards Consortium [31], along with a data-sharing framework and agreed-upon pediatric tuberculosis nomenclature, should be established as part of the custodianship plan. Local and national regulatory bodies may have restrictions on types of specimens that may be collected as well as the international exchange of samples. Such regulations vary by country, by specimen type (eg, host DNA), and by target patient population (eg, pediatric patients and pregnant women). A well-delineated custodianship plan can help address some of these issues by providing to the local and national regulatory bodies the agreed-upon principles and protocols that govern the biorepository. The National Cancer Institute Best Practices for Biospecimen Resources [32] notes that the "custodian is the trusted intermediary and caretaker of biospecimens and associated data, and the custodian's caretaking responsibilities should align with applicable ethical and policy standards," adding that the ideal custodian should be someone other than research investigators or sponsor(s) of the biospecimen resource (eg, a biospecimen resource manager) to eliminate potential conflicts of interest.

As part of a governance plan for the biorepository, an organizational structure should be delineated that includes a biorepository team responsible for planning and managing the repository as well as a sample access committee that governs access to samples. These could be modeled on the framework established by CTB2, a project of the TB Alliance, the CDC's TB Trials Consortium, and the AIDS Clinical Trials Group of the US National Institute of Allergy and Infectious Diseases (NIAID), NIH. This project is collecting high-quality patient specimens in late-stage tuberculosis drug clinical trials where they are linked to detailed, yet anonymized clinical documentation to enable discovery and qualification of biomarkers for clinical development of improved tuberculosis treatments for both drug-sensitive and multidrug-resistant tuberculosis (see http://www.tbbiorepository.org/). The sample access committee should include scientists knowledgeable about the research that may arise from the specimens, experienced curators of biorepositories, and experts in epidemiology, biostatistics, and informatics, among other technical consultants. Patient advocates and research participants, where feasible, should be included as members. The governance plan should describe (1) the methods for safeguarding the integrity of the samples and associated data, providing protocols used for biospecimen collection and storage; (2) clear procedures for requesting access to samples and data by investigators; (3) the review process for sample and data requests, assuring that the composition of a review committee that evaluates access requests is aligned with the stated mission/goals of the biobank; (4) policies for the dissemination of results from research that uses the samples; and (5) responsible fiscal planning for the long-term storage of specimens and data, as well as plans for securing future funds to maintain the repository until samples are depleted. Early planning for the custodianship, governance, and sample access mechanisms will help mitigate potential legal, ethical, and regulatory complications that can arise in the future operations of the biorepository.

CONCLUSIONS: CHARTING THE WAY FORWARD

Continued advocacy, collaboration, and communication are needed to ensure the key elements of the blueprint are adopted and implemented. Future pediatric tuberculosis biomarker efforts will require focus on standardization in terms of case definitions, specimen collection methods, and clinical data collection, as well as the adaptability to evaluate new potential biomarkers and technologies. Early inclusion of donors into the discussion should emphasize the need of funding not only for diagnostic development and pediatric cohorts, but also for building and maintaining specimen repositories as part of a larger research network. Developers of new tuberculosis diagnostics should be involved at early stages to consider ways to integrate new biomarkers into already existing, adapted, or new platforms.

Given the limited number of pediatric specimens in existing centralized repositories, there is a clear need to continue to enroll children in prospective cohort studies that collect standardized high-quality data and samples and to optimize available resources. Harmonization of specimen collection methods and clinical data collection has begun with the formation of a Pediatric TB Biomarker Working Group and a coordinating committee from the attendees of this workshop, with the goal of having unified standards for evaluating biomarkers from existing and future repositories. Building on newly discovered promising biomarkers and on new technologies with a focus on the clinical challenges in children and the priority areas as identified in this blueprint can serve as the initial stages of tuberculosis diagnostics development. Successful identification of a child-friendly tuberculosis diagnostic biomarker will require input, collaboration, and coordination from many stakeholders, from concept development of the study design to, ultimately, the development of a point-of-care test appropriate for use in those regions with the highest burden of pediatric tuberculosis.

Notes

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