WHO *Salmonella* Typhi/Paratyphi A Bivalent Vaccine Research and Development Technology ROADMAP

Priority activities for the research, development, testing, licensure, and use of vaccines against enteric fever.
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Executive Summary

This roadmap sets out the WHO Vision and strategic goals for the research and development of bivalent *Salmonella* Typhi/Paratyphi A vaccines against enteric fever for use in low- and middle-income countries (LMICs). It is designed for use by key stakeholders to outline activities that should be prioritized. The vision of this roadmap is to develop a safe, affordable, and broadly effective vaccine to protect children against the two etiological causes of enteric fever. The strategic goals are categorized into near-term, medium-term, and long-term objectives. In the near term, the focus is on demonstrating the safety, immunogenicity, and efficacy of a candidate bivalent vaccine. The medium-term goal is to achieve WHO prequalification for at least one bivalent vaccine for use in endemic LMICs. In the long term, the aim is to integrate the vaccine into national immunization programs, potentially as part of a combination vaccine.

To achieve these strategic goals, the roadmap outlines three main themes with specific actions. The first theme is addressing evidence gaps. This involves improving surveillance and burden of disease data, including antimicrobial resistance (AMR) surveillance, addressing diagnostic gaps with better laboratory capacity and point-of-care tests, and using modelling to predict the impact of vaccination and other interventions.

The second theme is accelerating vaccine development. This includes defining correlates of protection and appropriate trial designs, conducting immune interference studies and co-administration trials with existing vaccines, and establishing a clear regulatory pathway for licensure, including the use of controlled human infection models (CHIM).

The third theme is maximizing public health impact. This involves understanding the requirements for vaccine buy-in from stakeholders, demonstrating the cost-benefit value of a bivalent vaccine, enhancing national surveillance capacity, and ensuring effective communication and stakeholder engagement.

To support these activities, the roadmap emphasizes the need for establishing manufacturing capacity and workforce development in LMICs, creating sustainable financing mechanisms.
and incentives to ensure vaccine supply, enhancing national and regional surveillance networks, and effective communication and stakeholder engagement to ensure successful vaccine rollout and uptake.

By addressing evidence gaps, accelerating vaccine development, and maximizing public health impact, this strategic framework provides a comprehensive approach to developing and implementing effective vaccines in high-burden settings.
Acknowledgements

The Department of Immunization, Vaccines and Biologicals (IVB) at the WHO would like to thank the many individuals who contributed to the development of this document.

The draft R&D Roadmap for bivalent *Salmonella Typhi/Paratyphi A* vaccines was prepared by Ana Belén Ibarz-Pavon in the IVB department at WHO, with contributions from and review by the Technical Advisory Group for *Salmonella* Vaccines (TAG-SV):

Alejandro Cravioto, University of Mexico, Mexico; John Clemens, International Vaccine Institute, South Korea; John A. Crump, University of Otago, New Zealand; Melita Gordon, Malawi-Liverpool Wellcome Trust Clinical Research Programme, Malawi; Jacob John, Christian Medical College (Vellore), India; Andrew Pollard, University of Oxford, UK; Denise Garrett, Sabin Institute, USA; Karen Keddy, University of Pretoria, South Africa; Matthew Laurens, University of Maryland, USA; Xinxue Liu, University of Oxford, UK; Florian Marks, International Vaccine Institute, South Korea; Senjuti Saha, Child Research Foundation, Bangladesh.

WHO would like to thank Calman Maclennan, who participated in one or more of the consultations that informed the drafting of this document, and reviewed earlier drafts of the document:

Finally, WHO would like to acknowledge the contributions from WHO Headquarters: Adwoa Bentsi-Enchill, Katherine Emary and Annelies Wilder-Smith, Vaccine Product & Delivery Research, IVB, WHO, Switzerland.

Declarations of potential competing interests were received from all experts. WHO processes were used to assess declared interests and to manage any real or perceived conflicts of interest.

Funding statement:

The production of this document, was funded by the Bill and Melinda Gates Foundation.
Acronyms

AMR  antimicrobial resistance
CDC  centre for disease control and prevention
CHIM  controlled human infection model
ECVP  evidence considerations for vaccine policy development
EPI  expanded program on immunization
GAVI  global alliance for vaccines and immunisations (currently known and GAVI, the vaccine alliance)
LMIC  low- and middle-income country
MAPS  multiple antigens presenting systems
MIC  Minimum inhibitory concentration
MDR  multi-drug resistance
NTS  non-typhoidal salmonella
OMV  Outer membrane vesicles
R&D  research and development
SAGE  strategic advisory group of experts (on Immunization)
SEFI  surveillance for enteric fever in India
SEAP  surveillance for enteric fever in Asia project
STRAATA  strategic typhoid alliance across Africa and Asia
TCV  typhoid conjugate vaccine
TSAP  typhoid fever surveillance in Africa
TyVAC  typhoid vaccine accelerating consortium
WASH  water, sanitation, and hygiene
WHO  world health organization
Background On Vaccine R&D Technology Roadmaps

Vaccine R&D technology roadmaps produced by the World Health Organization (WHO) aim to provide a strategic framework outlining priority activities for global stakeholders - vaccine researchers, funders, industry, regulatory authorities, policy makers and the wider community - to accelerate availability of effective vaccines from development through licensure and policy recommendations and ultimately to reach the people in need with the greatest burden of disease, in particular in low- and middle-income countries.

This roadmap describes the areas of work and specific activities that WHO consider as priorities to support the development and deployment of vaccines targeting enteric fever caused by *Salmonella Paratyphi* A, which may be addressed as a combined enteric fever vaccine against *Salmonella Typhi* and *S. Paratyphi A* serovars, or as a combination vaccine product that also includes *Salmonella* serovars commonly associated with invasive Non-Typhoidal Salmonella (iNTS). This document has been compiled with inputs from academic groups, industry, funding bodies, public health agencies and national stakeholders from endemic areas. The document is not product- or product-type specific. WHO encourages implementation of the roadmap, which will be updated as necessary if the vision, goal, or priority activities evolve as new information becomes available.
Introduction

The genus *Salmonella* encompasses a group of gram-negative bacteria belonging to the *Enterobacteriaceae* family. Within this genus, two primary species exist: *Salmonella bongori* and *Salmonella enterica*. *Salmonella enterica* can be further categorized into subspecies and into > 2,500 serovars. Of these, around 50 account for approximately 99% of clinical isolates in cases involving human and animal diseases (1). In regard to human disease, *Salmonella* serovars are divided into typhoidal and non-typhoidal, and collectively they are responsible for gastrointestinal and systemic infections that range from mild disease to life-threatening infections (2).

*Salmonella enterica* serovar Typhi and *Salmonella enterica* serovar Paratyphi A cause typhoid and paratyphoid fever, and are collectively referred to as enteric fever. *S. Paratyphi* presents three distinct pathovars: A, B, and C. While all three pathovars can cause paratyphoid fever, *S. Paratyphi* A is the most common aetiology, while *S. Paratyphi* C disease remains rare, and its presentation can be that of septicaemia with the formation of metastatic abscesses (3). Unlike other *Salmonellae*, serovars Typhi and Paratyphi A are restricted to human host, and disease spreads through a faecal-oral route through the ingestion of contaminated food and water, or through direct contact with infected individuals or carriers who excrete the bacterium in their faeces (4,5). The incubation period of the disease is typically 7-14 days, and it commonly presents as high fever, general malaise, and mild gastrointestinal symptoms, often making it indistinguishable from other common febrile illnesses (5,6). A confirmative diagnostic of enteric fever and the distinction between the two causative serovars is done by bone marrow culture, with blood culture and stool culture or rectal swab being accepted alternatives. Serological tests such as the Widal test can also be used to diagnose typhoid fever, albeit appropriate testing timing and consideration to their low performance can result in misleading findings. The lack of appropriate microbiology capacity in low-income settings, the inconsistent blood culture sensitivity, ranging from 30-90% (7,8), and the lack of well-performing point-of-care tests (9–11) often result in lack of etiological confirmation. Moreover, diagnostic efforts have been focused on the detection of *S. Typhi*, making *S. Paratyphi* A detection only available to laboratories with good microbiology and molecular testing capacity, which are scarce in Low – and Middle-Income Countries (LMICs), where enteric fever is prevalent (12). Misdiagnosis of enteric fever also results in inappropriate use of antibiotics and the consequent emergence of Antimicrobial Resistance (AMR). Overuse of antibiotics has led to the emergence and spread of *S. Typhi* resistant to first line antimicrobial treatment (13,14), including Multidrug Resistant (MDR) strains, and the more recent identification of Extensive Drug Resistant (XDR) strains in outbreaks in Pakistan and sporadic cases among returning travellers to the
US and Europe (15–18). AMR among *Salmonella Paratyphi* A is more heterogeneous, and MDR remains uncommon, albeit an increasing trend in recent years (16,19–23).

Despite *S. Typhi* being the most commonly identified serovar in enteric fever cases, *S. Paratyphi* A and, to a lesser extent, *S. Paratyphi* B also contribute significantly to the burden of disease. Both: typhoid and paratyphoid fever are associated with urban, highly populated settings, and poor water sanitation and hygiene practices, making LMICs particularly vulnerable (24–27). In 2019, 3.8 million cases of paratyphoid fever were diagnosed worldwide, leading to >23,000 deaths (28). However, it is likely the burden of paratyphoid fever is underestimated as, in the absence of laboratory confirmation and *S. Paratyphi*-specific PoC tests, infections might be wrongly attributed to *S. Typhi* (29,30).

Paratyphoid aetiology appears to affect more commonly older children and adults, and current data point to it being mostly confined to Asia and the Middle East, with sporadic endemic cases being detected in Europe, and rarely occurring in Africa (27,31,32). However, the distribution and burden of disease is likely to be underestimated, as there is evidence of substantial geospatial variability on the proportion of enteric fever due to *S. Paratyphi* A, hence, calling for the implementation of robust enteric fever surveillance strategies, particularly in LMICs (29,33). In contrast, typhoid fever infections affect mainly school-age children and young adults, although evidence points at a higher-than-anticipated incidence among young children in endemic settings (27,34–36), and even a disease burden comparable to that of found in other age-groups (19,37,38). While the highest prevalence of typhoid fever is found across Asia, there is evidence that typhoid burden is higher in sub-Saharan Africa than previously described, with several countries surpassing 100 cases per 100,000 population (25,24,26), further advocating for the need of improved national and regional surveillance, better diagnostics, and preventive measures targeting both aetiologies (29).

Access to clean water, sanitation and hygiene practices (WASH) have been proven effective in reducing the burden of enteric fever (39). However, such initiatives are hard to implement in LMICs as they require high, long-standing investment, political accountability and commitment, and proper community engagement and information interventions driven by multisectoral collaborations, and accompanied by local, sustainable capacity building in order to make an impact in disease prevention (40–43). Hence, vaccination still remains a desirable intervention for the prevention of enteric fever (44). Currently available vaccines are directed against typhoid fever and, to date, there are no licensed vaccines against *S. Paratyphi* A and other disease-associated *Salmonella* serovars. The WHO recommended the inclusion of typhoid vaccines into the vaccine programmatic schedules of high-burden countries in 2008 (45) and, in 2018 the recommendation was updated to recommend the use...
of a typhoid conjugate vaccines (TCV) (46). These vaccines have been proven safe and highly effective in the prevention of typhoid fever from early infancy, and induce long-term T-cell mediated immunity in high-burden settings in Africa and Asia (47–50). Moreover, TCV can be safely administered with other scheduled vaccines (49,51).

While the inclusion of a paratyphoid vaccine is desirable for the comprehensive control of enteric fever, it is unlikely a monovalent paratyphoid vaccine would be an attractive product. A bivalent typhoid-paratyphoid vaccine could prove a highly valuable public health tool for the control of enteric fever in high-burden settings (52,53). Moreover, the feasibility of a vaccine containing iNTS-associated serovars S. Typhimurium and S. Enteriditis in addition to enteric fever-causing serovars is currently under expert discussion (52). There are currently several S. Paratyphi A-containing vaccine candidates at different stages of clinical development that use different immunizing strategies to deliver the O-antigen, a surface-exposed polysaccharide known to induce a protective antibody response, to the immune system (54), and while monovalent S. Paratyphi A vaccine candidates are still investigated, developers have shifted towards bivalent combinations. Such vaccines use different strategies to induce an immune response. Live-attenuated S. Typhi and S. Paratyphi A strains (54,55), genetically-engineered live-attenuated typhoid strains expressing the O:2 lipopolysaccharide and flagellin H antigens from S. Paratyphi A (56) or, alternatively S. Paratyphi A Outer Membrane Vesicles (OMV) expressing the typhoid Vi-polysaccharide (57), and conjugate formulations containing O:2 and Vi antigen conjugated to a protein carrier (54,58) are some of the vaccine constructs currently under pre-clinical and early clinical stages of development. A bivalent typhoid-paratyphoid vaccine, whether a live-attenuated, and OMV, or a conjugate construct, would allow for a comprehensive, cost-effective control of enteric fever in a single product that can be incorporated into the Expanded Programme on immunization (EPI), and delivered concomitantly with other vaccines.

WHO Vision and Strategic Goals

The WHO immunization agenda 2030 (IA2030) is a global stakeholder strategy that aims at i) reducing mortality and morbidity caused by vaccine-preventable diseases, and ii) decrease disease burden by increasing access and uptake of novel vaccines (59). Under this framework, the WHO vision is vaccination with a product that is protective against both enteric fever-causing Salmonella serovars: S. Typhi and S. Paratyphi A, as a stepping step towards the ultimate goal of an affordable, and broadly effective vaccine to protect children against all invasive disease caused by Salmonella enterica. Current licensed vaccines are only protective against S. Typhi and, given the existing knowledge gaps on the distribution and contribution of paratyphoid fever to the burden of enteric fever, together with the
prospect of a possible serovar replacement following the introduction of typhoid-only vaccines into the programmatic schedule, a bivalent S. Typhi/Paratyphi A is highly desirable. There are currently multiple possible combinations of vaccines to protect against Salmonella disease, ranging from the existing, currently licensed monovalent vaccines targeting serovar S. Typhi, to bivalent and trivalent combinations against enteric fever and iNTS to, ultimately, a quadrivalent Salmonella vaccine containing the four serovars most commonly associated with invasive disease (i.e., S. Typhi, S. Paratyphi A, S. Typhimurium, and S. Enteritidis). A bivalent vaccine against enteric fever serovars is a logic step to fulfil the ultimate vision of a pan-Salmonella vaccine, considering current knowledge gaps in disease epidemiology, the development status of various vaccine candidates, and the complex regulatory pathways for a multivalent product. The R&D roadmap to a bivalent Salmonella Paratyphi A-containing vaccine addresses the strategic needs for the development of a bivalent S. Typhi/Paratyphi A vaccine for the comprehensive control of enteric fever in endemic countries, and paves the way for further development of Salmonella combination vaccines.

Vision

Development of a safe, affordable, and broadly effective vaccine to protect children against invasive disease caused by Salmonella enterica for use in LMICs

Strategic Goals

Near Term: to demonstrate the safety, immunogenicity, and efficacy of a candidate bivalent enteric fever vaccine against Salmonella serovars Typhi and Paratyphi A that shows immunological protection against S. Typhi non-inferior to currently licensed TCVs, and demonstrates immunogenicity in a controlled human infection model (CHIM) study, which is confirmed in post-licensure effectiveness studies.

Medium Term: to have at least one WHO-prequalified bivalent enteric fever vaccine to be used in endemic LMICs.

Long Term: programmatic inclusion of vaccine(s) for the prevention of enteric fever in infants and young children caused by typhoidal Salmonella serovars (which might be as part of a combination vaccine with additional Salmonella serovars and/or with other antigens) to be chosen on the basis of the clinical needs by region.
The long-term goal emphasizes the overarching vision of combination vaccines against multiple serovars of *Salmonella* that cause invasive disease, and the near-term strategic goal to develop an immunogenic enteric fever vaccine that elicits an immune response against *S. Paratyphi* A and confers protection against *S. Typhi* non-inferior to existing, prequalified TCVs, which can be prioritized for further evaluation in later phase clinical trials.
Priority Activities

Vision

Development of a safe, affordable, and broadly effective vaccine to protect children against invasive disease caused by *Salmonella enterica* for use in low- and middle-income countries

Strategic goals

1. Near term: to demonstrate immunogenicity, safety, and efficacy of a candidate bivalent enteric fever vaccine against serovars Typhi and Paratyphi A that protects against *S. Paratyphi* A infection and elicits protection against *S. Typhi* non-inferior to currently licensed monovalent vaccines
2. Medium term: Licensure of at least one bivalent enteric fever vaccine to be used in LMICs

Addressing evidence gaps

1. Improve epidemiology and burden of disease data, including AMR surveillance
2. Addressing the diagnostic gap
3. Modelling

Accelerating vaccine development

1. Define correlates of protection
2. Define the appropriate trial design and clinical endpoints
3. Immune interference studies for Typhi/Paratyphi A vaccines, and co-administration with existing vaccines
4. Define the regulatory approach and pathway to licensure

Maximizing public health impact

1. Understanding the requirements and needs for vaccine buy-in
2. Demonstrate the cost-benefit value of a bivalent enteric fever vaccine

Key Capacities

- National and regional surveillance networks
- Manufacturing and workforce capacity building
- Effective communication and stakeholder engagement
- Funding & sustainability

- Academia
- Clinicians
- Industry
- Vaccine developers
- Community
- Global health agencies
- LMIC decision makers
- Funders
- National regulators
Theme 1: Addressing Evidence Gaps

The development and subsequent introduction of TCV in several high burden countries in prompted WHO to recommend the implementation of laboratory-confirmed health facility-based surveillance to determine disease burden and antimicrobial susceptibility patterns of strains circulating prior and post vaccine implementation, assess vaccine impact, and monitor possible serovar replacement patterns (60). The Typhoid Fever Surveillance in Africa (TSAP) was established in 2009 as a passive laboratory-based surveillance for bloodstream infections across ten countries in Africa, aiming at generating data on the incidence of typhoid fever and iNTS disease in Africa, and informing targeted interventions to prevent *Salmonella* disease (61). Several regional surveillance programs were established in 2016 up under the auspice of non-profit research and academic institutions, and run until 2019/20 depending on the program and site. These initiatives, namely the Severe Typhoid in Africa (SETA) program, the Surveillance for Enteric Fever in Asia Project (SEAP) and Surveillance for Enteric Fever in India (SEFI), and the Strategic Typhoid Alliance Across Africa and Asia (STRAATA) (62–64) generated invaluable data to inform the 2018 recommendation from the Strategic Advisory Group in Immunisations (SAGE) for the use of the TCV in endemic settings (46). All studies implemented passive, health facility-based, blood culture-confirmed surveillance among febrile patients to generate crude and age-stratified incidence rates, complemented by healthcare utilization surveys to adjust rates based on catchment area population for participating health facilities. As well as disease burden for typhoid, these studies are providing information on circulating strains characteristics, data on AMR patterns, and generating a bacterial population collection to be used in future genomic surveillance, population structure and evolutionary patterns investigations, and to inform mathematical modelling studies (62). While data on paratyphoid fever are being collected through typhoid surveillance initiatives, these are yet to be reported in the published literature. Access to these data is crucial for vaccine developers and policy maker in order to make decisions in regards to vaccine demand and supply needs, to provide evidence to support the introduction or switch from TCV to a bivalent *S. Typhi/Paratyphi* A vaccine, and to establish a baseline that can be used to evaluate vaccine impact through post-licensure vaccine effectiveness studies, which will likely be a requirement for vaccine licensure and WHO prequalification in the absence of phase 3 efficacy trial data. These data will also be crucial for modelling studies to evaluate the impact and cost-effectiveness of different vaccination strategies against enteric fever in settings with different disease epidemiology.
1.1 Improve Surveillance and Burden of Disease Data

While ongoing surveillance initiatives might shed light on the burden and distribution of paratyphoid fever and its contribution to the overall burden of enteric fever, knowledge gaps still remain in regard to the relative contribution from pathovars A, B, and C, which is not addressed by current surveillance initiatives (65,66). Uncertainties still remain in regards to the true incidence of enteric fever and, particularly, of paratyphoid fever in young infants, and complications derived from the disease (36,67,68). Ongoing surveillance embedded in national surveillance systems and beyond research initiatives is needed in order to inform of long-term stochastic changes on disease patterns before and after vaccination. Such changes might occur also as a result of the implementation of other preventive interventions, and from improvements in water, sanitation and hygiene (WASH) measures. Serovar distribution presents geographic and temporal variations, which are likely to be further driven by mass-vaccination with the non-comprehensive TCV vaccine (69,70). Early detection of serovar replacement will be crucial in the post-vaccine implementation era and, particularly, in the context of various combinations of multi-serovar vaccines against both: enteric fever and iNTS, being under consideration. Good quality burden of disease data will be crucial for decision-making in regards of bivalent vaccine use, and to allow vaccine impact evaluations in the post-implementation era, which will provide policy makers with the necessary information for decision-making.

1.1.1 Surveillance of Antimicrobial Susceptibility and Impact of Vaccination

As well as disease trends and age-disaggregated data, disease surveillance needs to record AMR data, as changes occur as a result of antibiotic use, or from the spread of particular strains. The growing threat of AMR worldwide, and the impact of vaccines as a tool to control their spread has gained momentum in recent years, following the introduction of conjugate vaccines capable of reducing transmission as well as providing protection against the disease (71–73). The emergence of S. Typhi resistant to first-line antibiotics has been reported since the 1950’s (74), and multidrug resistance is documented since the 1980’s (14,75). More recently, extensive drug resistance emerged in Pakistan, and spread worldwide through international travelers (16,18,50,76). S. Paratyphi A presents a different AMR pattern, with MDR being less common; however increasing Minimum Inhibitory Concentrations (MICs) are steadily raising, and non-susceptibility is increasingly observed (16,19–22). Mathematical models suggest the introduction of TCV in high-burden settings can significantly contribute to reduce the burden of AMR typhoid infections by preventing transmission of resistant and non-resistant strains among symptomatic and asymptomatic, chronic carriers, hence increasing the cost-effectiveness of the intervention by reducing healthcare-associated costs (77,78).
However, AMR emergence is driven by antimicrobial prescription patterns and usage, and any changes to these derived from vaccination will require dedicated investigations. Available evidence shows that the current TCV vaccine is equally effective against AMR strains (50), and while typhoid vaccination has not shown any substantial changes in antimicrobial prescription frequency and AMR patterns (79), adding a \( S. \) Paratyphi A component to the vaccine might result in a measure in high burden settings (15,80). Maximizing the impact of vaccination on AMR will require a coordinated, comprehensive intervention that extends beyond enteric fever vaccines and requires the implementation of global and national AMR action plans; however, the role and the value of vaccines as an additional tool to control AMR need to be taken into consideration (81–83).

### 1.2 Addressing the Diagnostic Gap

Confirmation of a diagnostic of enteric fever and its causative aetiology serves three distinct, yet related purposes: i) diagnostic of acute disease cases for the purpose of appropriate clinical management and antimicrobial usage; ii) detection of convalescent and asymptomatic carriers for that might result of exposed contacts, and iii) assessment of disease burden (84). Hence, fit-for-purpose diagnostic strategies that can address the three aforementioned need are essential for a holistic control of enteric fever, each of them perhaps requiring different sample types, diagnostic platform, and laboratory testing methods.

The clinical presentation of enteric fever ranges from a low-grade fever and malaise accompanied by gastrointestinal symptoms, to severe invasive disease that can result in intestine perforation and peritonitis (85). Distinguishing the disease from other febrile illnesses prevalent in endemic countries is not possible without laboratory diagnostics and often results in the prescription of broad-spectrum antimicrobial, which contributes to the spread of AMR. Moreover, typhoid and paratyphoid fever are clinically indistinguishable (30), making laboratory confirmation indispensable to fully elucidate the contribution of each pathogen to enteric fever burden, understand transmission patterns, and for decision-making in regards to the use of a \( S. \) Paratyphi A-containing vaccine. Yet another diagnostic gap is the distinction among the \( S. \) Paratyphi A, B, and C. Vaccination with a bivalent product would be expected to impact on the burden of disease caused by \( S. \) Paratyphi A and, to some extend B, both of which are a cause of enteric fever, and while the distinction among pathovars might not impact vaccine development and policy, is important for surveillance purposes to detect a hypothetical pathovar replacement. This diagnostic gap can only be addressed by increasing laboratory capacity, including genomic analysis capacity to allow the distinction among the three \( S. \) Paratyphi serovars (86,87). Clinical algorithms and decision-making tools without confirmatory tests provide only modest
discrimination power among a small number of febrile conditions, and cannot make the distinction between \textit{S. Typhi} and \textit{S. Paratyphi A} aetiologies \cite{88,89}. Microbiological culture and isolation of the aetiological agent from bone marrow, blood, stool, or anatomical lesions is the only conclusive way to diagnose typhoid and paratyphoid fever \cite{90}. While bone marrow culture is considered the "gold standard", and has demonstrated a sensitivity >80%, the challenges presented by the invasive procedure required to obtain the sample mean that such test is seldom performed, and its implementation as a routine test, particularly in LMICs with limited testing and microbiology capacity, is unrealistic. Blood culture, while still presenting challenges in settings with limited capacity, is an acceptable alternative. However, blood culture sensitivity is low and highly dependent on sampling technique, blood volume, and timely sample processing \cite{91–95}. Stool culture and rectal swabs, while capable of rendering positive results, they require cautious interpretation, as they are unable to distinguish acute illness from asymptomatic shedding \cite{95}. Building laboratory capacity by ensuring adequate infrastructure and capacitated personnel through capacity building programs, quality control and assurance systems, and staff retention initiatives, ensuring a good laboratory network with sample referral infrastructure and timely reporting of findings remains crucial. However, there is a need to develop alternative culture systems and incorporate molecular diagnostic techniques to routine laboratory procedures that ensure confirmatory can be used for clinical management as well as for surveillance purposes \cite{96,97}.

Rapid Diagnostic Tests (RDTs) for the diagnostic of febrile illnesses present an attractive alternative for triage and outbreak detection, as well as facilitating rapid decision-making for vaccine deployment \cite{98,99}. Current commercially available RDTs for the diagnostic of enteric fever are mostly only able to detect \textit{S. Typhi}, their performance is highly heterogeneous and, in all cases, their sensitivity and specificity values fall below the required level for them to be recommended for diagnostic use \cite{11,99}. While the development of RDTs able to accurately confirm enteric fever and distinguish between the two aetiological agents might not be technically feasible, alternative approaches ensuring reliable, fast, near PoC diagnostics is recommended.

1.3 Modelling

In the absence of long-term data, disease transmission models are used to predict the long-term evolution of an infectious disease through the population, and to predict the effect and impact of one or more control interventions while accounting for confounders and effect modifiers that cannot always be accounted for in experimental studies, yet might shape the outcome \cite{100–102}. In order for mathematical modelling to be relevant for infection prevention and control, these need to
be able to answer questions that are relevant for policy-makers in a timely manner, while making use of the available, good quality data. In situations where a clinical trial might not be possible, such as low disease incidence that requires a large trial, difficulty to draw conclusions on population subgroups, or the need to extrapolate outcomes on the long term, modelling can further the validity of experimental trials (103). Modelling studies have been conducted on the transmission of typhoid fever (104), to estimate disease incidence in the absence of complete data (105–107), to evaluate and the impact of vaccination with TCV and other control measures on disease dynamics (108–111), to estimate the cost-effectiveness of TCV vaccination (112–115), and the impact of vaccination on antimicrobial resistance (116,117). Models have further ascertained that typhoid fever incidence is underestimated in endemic settings, and demonstrated the benefits of vaccination with TCV on disease and outbreak control, and on reducing the risk of AMR typhoid spread. However, there are currently no modelling studies on the potential impact of a S. Paratyphi A containing vaccine on enteric fever incidence or on AMR, and no models assessing the impact of a bivalent S. Typhi/Paratyphi A vaccine in endemic settings. There is currently sufficient information available to justify the use of a bivalent S. Typhi/Paratyphi A vaccine in endemic settings, however, if such the use of such vaccine requires additional financial resources, modelling-based cost-effective and cost-benefit analyses will be required to justify the additional costs to national immunization programs.

**Theme 2: Accelerating Vaccine Development**

Several S. Paratyphi A vaccine candidates, alone or in combination with S. Typhi Vi-antigen are currently under development (54,118). Evaluation of such vaccines will prove challenging, as there are currently no animal models to investigate the immunobiology of the disease and, due to the low and variable attack rates of paratyphoid fever, the need for sample sizes on the magnitude of six figures make efficacy trials unviable (119,120). Vaccine development has been hampered by the belief that the contribution of paratyphoid fever to the total burden of enteric fever is negligible; hence, efforts to date were vastly focused on the development and implementation of a vaccine against typhoid fever, which is reflected in the scarcity of S. Paratyphi A vaccine candidates currently under development, and mostly all still in early pre-clinical phase. Given that classic approaches to measuring vaccine efficacy are unlikely to be feasible, there is need to define correlates of protection or a surrogate, and to link vaccine-induced serum antibody responses to vaccine to protection against paratyphoid fever. Likewise, there is a need to ascertain serum antibody levels against the O:2 lipopolysaccharide and bactericidal activity as appropriate markers of protection, and to consider whether mucosal immunity should be considered when evaluating vaccine-induced protection (118).

In the absence of correlates of protection, controlled human infection models (CHIM) have already
shed some light on the immunological responses generated against the O-antigen of S. Paratyphi (121,122). In light that phase 3 efficacy studies are unlikely to be conducted, and in the absence of correlates of protection, establishing consensus on the approved regulatory pathway becomes critical to incentivise and accelerate vaccine development and programmatic uptake.

2.1 Define of Correlates of Protection

There is currently a scarcity of data on the host-response to S. Paratyphi A infection, the mechanisms and duration of protective immunity, and the role of continuous exposure to subclinical infections in high-burden settings (123). There is a need to get a better understanding on the mechanisms that lead to the development of humoral and cellular immunity following infection with S. Paratyphi A, and the role of individual factors (e.g. epithelium barrier, natural flora) and asymptomatic carriage in the development of clinical infection and development of a durable immune response. There is also a need to identify a clear surrogate or correlate of protection to link immunogenicity to efficacy against the S. Paratyphi O-antigen. There is evidence that anti-O:2 antibodies protect from re-infection, and that bactericidal activity correlates with the anti O:2 antibody titres (124,125). However, the role of bactericidal activity as a mechanism of antibody-mediated protection remains unclear. In the absence of phase 3 efficacy data, post-licensure phase 4 studies could be used to elucidate the immunological mechanisms of protection elicited by exposure to the pathogen, and to validate the correlates of protection derived from the CHIM model.

2.2 Define the Appropriate Trial Design and Clinical Endpoints

Given that it is unlikely that phase 3 efficacy randomized controlled trials (RDTs) that would allow an efficacy evaluation of the S. Paratyphi A component of any vaccine will be feasible, there is an urgent need for consensus on the most appropriate efficacy trial design. Human challenge models have already been developed, and findings suggest that microbiological confirmation is not always correlated to clinical symptoms. Hence, as well as microbiological confirmation, multiple secondary endpoints including proportion of cases diagnosed by clinical, microbiological or both should be considered when evaluating vaccine efficacy in such studies (126). Further investigations using CHIM will shed light in several existing knowledge gaps, such as host-pathogen interaction and the development of disease, immune protection, and duration of immune response, the identification of appropriate biomarkers of infection, correlates of protection, and vaccine efficacy against homologous and heterologous strain challenge (121). Given the impossibility of using conventional approaches to
vaccine development, future investigations might require innovative approaches such as adaptative
trial design and qualitative decision-making (118,127).

2.3 Immune Interference Studies for *Salmonella* Typhi/Paratyphi A Vaccines, and Co-
Administration with Existing Vaccines

There is currently evidence that the co-administration of TCV with other EPI vaccinations
administered after nine months of age; namely, measles-containing vaccines and meningococcal A
conjugate vaccine, does not have a negative impact in the development of protection conferred by
both vaccines, nor result in increased adverse events (128,129). However, immune interference studies
will be required to ascertain whether a bivalent enteric fever vaccine could impair the immune
response to either serovar, or to other co-administered vaccines. CHIM studies presenting homologous
and heterologous challenge with *S.* Typhi and *S.* Paratyphi A strains indicate that primary infection has
a partial protective effect against re-infection with the same serovar, but cross-protection is unlikely
(126). While CHIM findings suggest that it is unlikely the combination of Vi and O:2 lipopolysaccharides
will reduce immune response across serovars, it is still important to demonstrate that the immune
response induced by a bivalent vaccine is non-inferior to currently licensed typhoid vaccines.

2.4 Define the Regulatory Approach and Pathway to Licensure

Vaccine development and trial design should ensure that available data meet the necessary
regulatory requirements to inform national regulatory authorities’ decision, and provide policymakers
with the evidence needed to inform policy recommendations at national level and beyond. However,
clarity is needed from national regulators and WHO PQ team to ensure any upcoming bivalent products
are developed and evaluated to meet their requirements. The current paratyphoid fever disease
epidemiology makes classic phase 3 efficacy trials financially unfeasible; hence CHIM studies are likely
to play a big role in generating the necessary information that might be used for decision-making and
licensure on the basis of immunogenicity data from phase 2, demonstration of protection on a CHIM,
and field studies to confirm vaccine effectiveness. A bivalent vaccine will need to demonstrate that it
confers immunogenicity against typhoid fever at least non-inferior to that conferred by TCV; for the *S.*
Paratyphi A component, the immune response in endemic settings should be shown to be superior to
that observed in convalescent individuals. Evidence of protection against infection would also be
demonstrated in CHIM, with a minimum acceptable vaccine efficacy level of 50%.
Discussion to agree on the basis for vaccine licensure will need to initiate at the earliest possible stage, in preparation for a vaccine product that is ready for the market. This will require consultation with regulatory bodies and authorities to commence at early stages of vaccine development. In the absence of efficacy data, a conditional approval might be agreed with regulatory bodies of high-burden countries, where a post-marketing phase IV assessment could be conducted as a condition for a full regulatory approval and global roll out. In-country vaccine registration, especially on the basis of a conditional approval, will require decisive steps from WHO to include the vaccine in the high priority listing to ensure prompt prequalification, and provide the necessary evidence to a SAGE recommendation. The WHO’s Evidence Considerations for Vaccine Policy (ECVP) framework for collating available evidence might be a good instrument to collate the necessary evidence in preparation for regulatory and policy recommendations, all in view of informing of the potential public health value of a future pan-salmonella vaccine, incorporating iNTS causing serovars (130).
Theme 3: Maximizing Public Health Impact

In light of existing epidemiology data on the burden of enteric fever in Asia, vaccine development efforts have historically been focused on a product against typhoid fever that could confer long-term protection. As a result, research and development efforts from investigators and manufacturers alike were directed at S. Typhi, and culminated with the licensure and subsequent introduction of TCV in several endemic countries. However, surveillance efforts resulting from the need for post-licensure evaluation studies have brought attention to the increasing contribution of S. Paratyphi A to the burden of enteric fever, and renewed interest in a potential vaccine that could comprehensively protect against both aetiologies.

One of the major roadblocks to TCV advocacy for decision-making has been the lack of reliable burden of disease data. Likewise, the incorporation of a S. Paratyphi A component to the vaccine, which might result in additional requirements such as adjustments to costs, adapted vaccination strategies, and updating information materials for the target population will require robust data to justify the additional efforts. However, a bivalent vaccine might become an attractive product in additional markets outside S. Paratyphi A endemic settings as a mean to prevent a possible serovar replacement, which would favour a market price adjustment to a cost comparable to that of current S. Typhi vaccines. In such scenario, complete, accurate data on disease burden and trends of paratyphoid fever incidence prior to and following TCV introduction become crucial to advocate with (i) developers and researchers to continue to fill in the existing gap in regards to safety, immunogenicity, and efficacy of the S. Paratyphi A component of the vaccine and the non-inferiority of the S. Typhi component in regards to existing products, (ii) clarity on the requirement of post-licensure studies as a possible condition to licensure in the absence of clinical efficacy data, (iii) stakeholders and governments for the implementation/switch towards a bivalent product, (iv) manufacturers, funders, and donors to ensure sustainable, steady supply of vaccine at affordable cost and effective implementation strategies.

3.1 Understanding the Requirements for Vaccine Buy-In

There is a general consensus among researchers and policy makers alike that a standalone S. Paratyphi A vaccine is unlikely to be an attractive product to neither policy makers nor vaccine manufacturers. Hence, product development efforts need to focus on development of a bivalent product conferring against enteric fever as a whole. Vaccine implementation strategies are likely to require consideration to the fact that S. Paratyphi A infections appear to peak at a slightly older age
than those caused by *S. Typhi*, and will require further data on age-stratified surveillance. Overall, a bivalent vaccine is likely to be required to demonstrate non-inferiority to existing typhoid vaccines in terms of protection, affordability, and long-term sustainability. Initial implementation is likely to be constrained to markets in Asia, where incidence of paratyphoid fever has demonstrated to be a cause of concern. Additional markets might open if serovar replacement following TCV vaccination is confirmed, or as a preventive measure to a possible *S. Paratyphi A* introduction through travel.

There is a need to conduct a stakeholder analysis early on to understand and define who needs to be involved, at which stage, and at what level. Furthermore, there is still limited insight on the willingness of countries to adopt a bivalent vaccine, and their views on what data and vaccine implementation conditions they would require to make a favourable programmatic decision. This is particularly relevant in the African region, where TCVs have or are in the process to be introduced, but paratyphoid fever is rare and, therefore, a decision to adopt a bivalent vaccine will require ethical and financial considerations as much as scientific evidence. It is important that key stakeholders participate in shaping the clinical and regulatory strategies, and have a say on programmatic implementation strategies, acceptable pricing levels and investments, and in the development of policy guidance and vaccine recommendations. The existing TyVac consortium might be a good platform for advocacy, and to use the lessons learned from TCV development and implementation to help shape vaccine development, advocacy, and introduction strategies.

### 3.2 Demonstrate the Cost-Benefit Value of a Bivalent Enteric Fever Vaccine

A key consideration for vaccine implementation will be the demonstration of the cost-effectiveness and cost-benefit value, and the development of long-term investment plan to ensure long-term sustainability of vaccine supply and demand. Surveillance strategies such as SETI, SEAP, and STRAATA have made considerations to embed cost-effectiveness evaluation studies. However, these initiatives will need to look at enteric fever as a whole, without consideration to the relative contribution of *S. Typhi* and *S. Paratyphi A* to the model. Current available data show low impact of a monovalent *S. Paratyphi A* vaccine on morbidity and mortality or contention of AMR; this is particularly driven by low paratyphoid disease incidence and mortality rates. There is a need for future economic analysis models to take into consideration indirect measurements such as out-of-pocket expense, loss of productivity, and transportation costs. Likewise, AMR impact needs to be accounted for, with the understanding that data on economic impact of AMR on healthcare burden and costs are currently limited due to the lack empirical evidence, which might be a limitation for their incorporation into modelling approaches (131).
Key Capacities

Establish Manufacturing Capacity and Workforce Capacity Building in LMICs

A sustainable alternative with a long-term vision would be working towards instituting regional vaccine development and manufacturing capacity that could potentially take over vaccine production for the local market. Current S. Paratyphi containing vaccines utilise different platforms for antigen delivery (i.e. attenuated strains, protein-carrier conjugation, genetically-modified OMVs, multiple antigen presenting systems (MAPs)) which could be applied to other pathogens. The lack of local capacity for health products and vaccines became a significant constraint to LMICs during the COVID19 pandemic, and it is currently a strategic priority for pandemic preparedness to the Africa CDC (132).

Establish Sustainable Financing Mechanisms and Incentives to Ensure Vaccine Supply

Due to the limited projection of vaccine usage being mostly confined to LMICs in Asia, financial incentives and minimum market coverage assurance will be necessary to incentivise vaccine developers and manufacturers. Vaccine introduction will be primarily driven by disease burden, public health competing priorities, and the availability of support from international bodies such as GAVI. Initial market assurance, financial incentives, and generation of additional data from alternative sources such as environmental surveillance and serosurveys will be crucial to provide additional supporting information to support countries’ decision on implementation and continuous vaccine demand.

Enhanced National Surveillance Capacity

Lack of accurate burden of disease data has hampered the decision-making process for the implementation of the TCV (133). The implementation and development of a national enteric fever surveillance system that builds upon current ongoing surveillance initiatives established to inform decision-making for the implementation of the TCV is paramount to understand disease epidemiology prior to making an informed decision on vaccine implementation, to identify any changes in the disease epidemiology brought upon by the programmatic use of TCV, including serovar replacement or emergence of S. Paratyphi A in settings where it is currently not widespread, and to provide evidence to support the implementation or switch to a bivalent product once these are in the market (62,63).

Moreover, surveillance capacity, once established, can be expanded to cover other Salmonella infections, hence, further supporting evidence generation efforts towards a tetravalent Salmonella
combination vaccine, and contributing to the long-term goal of a single vaccine against invasive
Salmonella disease (52).

Effective Communication and Stakeholder Engagement

The successful development and implementation of a S. Paratyphi A-containing vaccine in the
context of an existing vaccine against typhoid fever will require extensive, ongoing engagement and
open communication among stakeholders. Communication across stakeholders has been identified as
a key barrier for the effective rollout of the TCV, resulting in lower-than-desired vaccine coverage (133).
Vaccine rollout and catch-up campaigns require coordination at several levels of the health and social
system, including private practices and the department of education if vaccination is to be distributed
through the school system. Misinformation regarding the vaccine itself can quickly spread rumours
and vaccine hesitancy, and an ineffective communication strategy to the population might result in
missing targets, of generate confusion when vaccination is to be implemented concomitantly with
other health interventions. In the context of an ongoing TCV vaccination strategy, the shift towards a
bivalent vaccine will require context-appropriate health promotion strategies and communication
materials ahead of any vaccine implementation. Questions regarding the reasons behind the
replacement of an existing, effective vaccine with a newer product, and the implications of such change
are likely to arise in the community, and among healthcare workers and decision-makers alike. Gaining
understanding of and addressing these issues alongside vaccine development and ahead of vaccine
marketing and deployment will ensure acceptability among the communities, and maximize uptake.
References


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