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THE CASE FOR A TYPHOID VACCINE PROBE STUDY AND OVERVIEW OF DESIGN ELEMENTS

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Abstract

Recent advances in typhoid vaccine, and consideration of support from Gavi, the Vaccine Alliance, raise the possibility that some endemic countries will introduce typhoid vaccine into public immunization programs. This decision, however, is limited by lack of definitive information on disease burden. We propose use of a vaccine probe study approach. This approach would more clearly assess the total burden of typhoid across different syndromic groups and account for lack of access to care, poor diagnostics, incomplete laboratory testing, lack of mortality and intestinal perforation surveillance, and increasing antibiotic resistance. We propose a cluster randomized trial design using a mass immunization campaign among all age groups, with monitoring over a 4-year period of a variety of outcomes. The primary outcome would be the vaccine preventable disease incidence of prolonged fever hospitalization. Sample size calculations suggest that such a study would be feasible over a reasonable set of assumptions.

Keywords

clinical trial; disease burden; trial design; typhoid; typhoid vaccine; vaccine; vaccine probe study

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AUTHOR ROLES

BDG conceived of the study and wrote the first draft of the manuscript. MEH performed sample size calculations and provided critical input into the manuscript draft. IK provided data on fever incidence, assisted with interpretation, and provided critical input into the manuscript draft.

CONFLICTS OF INTEREST

BDG works for AMP, which receives or has received during the past 3 years grant-specific support from Crucell, GSK, Merck, Novartis, Pfizer, and sanofi pasteur; however, none of these grants relate to work on typhoid or typhoid vaccines. The other authors declare no conflicts relevant to the current manuscript.

BACKGROUND

Three recent vaccines have or are soon to be introduced in the majority of low income settings, including *Haemophilus influenzae* type b (Hib) conjugate vaccine, pneumococcal conjugate vaccine (PCV), and rotavirus vaccine. For each of these vaccines, introduction was delayed in part because of the difficulty in defining disease burden. For Hib and PCV, the most common severe manifestation was non-bacteremic pneumonia, which required for diagnosis trans-tracheal aspirates or lung punctures, both of which were impractical in low-income settings. For all three diseases, other issues existed including lack of access to health care facilities, poor laboratory capacity, and lack of collection of clinical specimens by health care providers.

A solution to this issue was the development of vaccine probe studies. Probe studies employ standard clinical trial designs, ideally a blinded community or individually randomized design. However, they differ conceptually from vaccine licensing studies in that they use a previously licensed vaccine of known efficacy (or hypothesized efficacy based on established correlates of immunity) to define characteristics of disease rather than of vaccine (1). The primary outcome of a vaccine probe study is the vaccine preventable disease incidence (also known as the vaccine attributable rate reduction), which is defined as the difference in incidence between control and intervention populations, or, mathematically equivalently, the control group incidence multiplied by vaccine efficacy. Because probe studies assess disease burden, they may provide a better assessment of a vaccine's public health value than vaccine efficacy or effectiveness. Additionally, such studies can provide a way to prioritize vaccines based on the preventable disease burden, which may be high even when vaccine efficacy is relatively low (2).

Vaccine probe studies have been successful in convincing policymakers of the importance of Hib (3), PCV (4) and rotavirus (5) vaccines and of providing anchoring data to help interpret less robust studies, such as surveillance of etiologically confirmed disease. As we describe, probe studies could provide a similar benefit for typhoid vaccine in low- and middle-income countries.

JUSTIFICATION

The expense and complexity of vaccine probe studies can be justified for numerous reasons, many of which apply to typhoid. As described above, probe studies seek to define characteristics of disease rather than vaccine. Currently, disease burden estimates are unknown. A recent systematic literature review estimated that in 2010, low- and middle-income countries experienced 11.9 million typhoid cases and 129,000 deaths (6), which differs from earlier estimates (7) and the 2013 global burden of disease study (7,8). Moreover, the latter study provides a range from 85,900 to 268,000, emphasizing the degree of uncertainty in this estimate. As with other studies, this most recent estimate has several limitations. It does not adjust for limited access to care in many of the countries with the highest typhoid burden.

More problematically, the estimate of deaths relies on in-hospital case-fatality ratios, which may greatly underestimate total deaths. While outpatient antibiotic therapy may reduce

mortality and morbidity even in the absence of hospital care, the degree to which this occurs remains unknown, and may be blunted in settings where counterfeit antibiotics exist or antibiotic resistance is common (9,10). A WHO expert panel has noted the importance of estimating access to care when estimating typhoid burden (11). A potential clue to the underestimation of burden due to limited access to care is provided by a rotavirus vaccine probe study that found that vaccine prevented six-fold more cases of severe dehydration in the community than in the clinic (5). A 2014 review summarizes the situation in Africa as follows: “much is not known about typhoid fever in Africa; and appropriate technology to assess the actual burden of disease is not available” (12).

Laboratory diagnostics are imperfect for typhoid, particularly blood culture, which has been estimated as having a sensitivity of approximately 61% (6). A systematic review published during 2012 estimated that adjusting for imperfect sensitivity of blood cultures increased the estimated number of cases globally from 13.5 million to 26.9 million (7). However, other issues exist with laboratory diagnostics besides in-hospital sensitivity including that blood cultures may not be performed at all. For example, persons who present with critical illness may die or have antibiotics given before culture can be obtained, something that may occur more frequently among the youngest age groups. Laboratories in remote settings may not have diagnostic capacity. Where diagnostic capacity exists, it may be limited to certain periods (e.g., during weekdays).

Typhoid fever documentation requires a reasonable level of clinical suspicion, particularly where other common causes of fever exist such as malaria. One solution to this problem is to conduct a study in which all persons presenting with clinical fever receive diagnostic testing. This was the approach taken in an urban slum in Kenya, where community surveillance was followed by blood culture of all persons with fever or pneumonia (9). This approach documented higher typhoid incidence than previous African studies (13), on a par with burden in Southeast Asia (14). Nevertheless, the early case detection and small sample size prevented estimation of severe disease burden.

Similarly, a substantial contributor to typhoid burden is the complication of intestinal perforation (15). Case fatality ratios for this complication may be high (20% in Africa and over 10% in Asia) with prolonged hospitalization. Yet few studies have attempted to estimate the incidence of typhoid-associated intestinal perforation with concurrent long-term follow-up.

In theory, robust surveillance with high levels of health care access and adjustment for false negative results could provide a more straightforward means of burden estimation. However, in typhoid endemic areas health care access is frequently low and its level for a particular disease unknown. As a characteristic of the test, blood culture and other diagnostic sensitivity may be known but positive predictive value is not and remains dependent on variable levels of pre-treatment with antibiotics, duration and severity of illness before testing, and blood volume collected. Additionally, diagnostic tests often are not performed, particularly among patients who present with critical or atypical illness, and when performed may not be processed correctly. By contrast, most of these issues could be addressed with a

well-designed probe study. The remainder of this manuscript addresses some of the associated design issues.

ASSUMPTIONS

Several assumptions are made in the proposed study design described below. We assume that a vaccine will be licensed in the target age group and that the primary indication will be for prevention of laboratory confirmed typhoid disease. We assume that the vaccine has no more than minimal side effects, on a par with those for vaccines such as a pneumococcal and Hib conjugate vaccines.

GOAL, OBJECTIVES AND ENDPOINTS

Goal

To determine the burden of typhoid across various syndromic categories in suspected high burden areas.

Primary outcome

The vaccine-preventable burden of clinical typhoid presenting as hospitalization with prolonged fever of greater than 5 days.

Justification—In most settings with high typhoid burden, the main concern - other than mortality - is likely to be severe disease and high cost health care utilization. Focusing on fever with greater than 5 days duration should enrich the definition for typhoid and thus require a smaller sample size, similar to the use of a specific radiological endpoint for PCV trials (4). Mortality is not recommended as the primary outcome due to difficulty in identifying deaths and the frequent impact the presence of a study has in decreasing mortality rates through improving care (e.g., during the RTS,S malaria vaccine trials [16]).

Secondary outcomes

- Vaccine-preventable burden of clinical typhoid presenting as hospitalization with fever of any duration.
- Vaccine-preventable burden of clinical typhoid across all-cause hospitalizations.
- Vaccine-preventable burden of clinically diagnosed intestinal perforation
- Burden of clinical typhoid prevented through indirect protection.

Justification—The first two outcomes have greater sensitivity but less specificity than the primary outcome and thus may require unrealistic sample sizes. The third outcome assesses the primary severe complication of typhoid but requires additional diagnostic capacity.

Site-specific outcomes

- Vaccine-preventable burden of severe clinical typhoid (using systematic case definitions) in the community.
- Vaccine-preventable burden of clinical typhoid presenting to outpatient settings

- Vaccine-preventable burden of typhoid mortality

Justification—Researchers in some settings with poor health care access may want to include measurement of community cases. For example, a rotavirus vaccine probe study found six-fold more preventable disease in the community than in a hospital setting (5). Conversely, in settings with better health care access and case management, the primary typhoid burden may occur in outpatient settings. Finally, mortality outcomes may be appropriate in settings with both substantial typhoid mortality and high quality mortality surveillance.

Ancillary outcomes

- Vaccine immunogenicity through a nested trial to document vaccine performance in the target population.
- Vaccine effectiveness against etiologically confirmed disease in the target population

Justification—A vaccine probe study using vaccine with unknown properties in the target population should have a nested immunogenicity study. In addition to short-term immunogenicity assessment, such a study should follow a subset of vaccinated children over the study duration to estimate duration of immunity. Because vaccines do not prevent all cases of target diseases, it is useful to measure vaccine effectiveness, which then can be used to adjust burden estimates and to determine the etiologic fraction of syndromic illness attributable to typhoid (defined as VE against syndromic disease divided by VE against etiologically-confirmed disease).

STUDY DESIGN

As with vaccine against other enteric diseases such as cholera (17), typhoid vaccine should have substantial indirect effects. This has not occurred in all trials, however. For example, a mass vaccination of all children age 2 years and older with ViPS demonstrated strong indirect protection at modest coverage (18) while vaccinating school-aged children with ViPS in Karachi did not demonstrate indirect protection (19). A recent modeling study indicated that indirect protection is likely to increase where transmission levels are lower and where chronic carriers have less of a role in transmission (20).

If indirect effects are strong (that is, vaccination of a relatively small proportion of the population leads to substantial reduction in transmission), then an individually randomized study may underestimate vaccine preventable disease burden since disease incidence in both vaccinated and unvaccinated groups will be low. For example, Hib vaccine may show indirect effects starting at 20% coverage (21). Consequently, we recommend a community or cluster randomized trial as has been done for previous probe studies using Hib (3) and pneumococcal (4) conjugate vaccines, as well as typhoid vaccine trials (22).

CLUSTER DEFINITION

The study should include a sufficient number of clusters to ensure that the cluster-randomized design does not have a large impact on study power. Cluster characteristics should consider the following:

- It is likely that clusters will be defined based on administrative borders, and depending on the study site may include village, hamlet, or neighborhood.
- Cluster definition should respect catchment areas for immunization to ensure that persons assigned to a particular cluster receive the appropriate study vaccine.
- Clusters chosen for inclusion should not have substantial migration between vaccinated and intervention clusters, as this could bring incidences in the two areas closer to equilibrium.
- Clusters should have enough separation to minimize contamination of intervention clusters by control populations, which would minimize indirect effects.
- Ideally, the contamination source for any particular cluster will be within the cluster and not extend beyond it. Specifically, investigators should attempt to ensure that intervention and control clusters do not have a common source of contamination, such as a common water supply.

TARGET AGE GROUPS

The goal of a probe study is not to design an effective public health immunization program, but to define more accurately disease burden among populations already known to have high incidence or mortality. For Hib and rotavirus vaccines, this included only young children. PCV probe studies also were limited to young children, with the expectation that infant immunization would provide indirect protection to older age groups. In the case of typhoid, within high-risk settings risk is distributed widely across different age groups: in the 2014 review, 21% of typhoid cases occurred before age 5 years, 40% from age 5–14 years, and 39% among older persons (6), although this was based on only four studies. Furthermore, data do not exist on whether typhoid vaccination given to one age group will provide indirect protection to unvaccinated age groups.

Consequently, the optimal approach would be similar to that employed for a study of oral cholera vaccine, namely vaccinating all persons with an on-label age indication (23). This would lead to the maximum disease reduction within intervention communities, and thus most accurately define disease incidence. It also would address the concern that chronic carriers might maintain transmission and disease occurrence among unvaccinated persons. For practical reasons, a probe study might be confined to a more limited population. For example, modeling studies have suggested that routine vaccination of 6 year old children plus a onetime mass campaign among persons 6–15 years of age would substantially reduce disease in India (20). Vaccine cost and the cost of a mass campaign, however, represent only a portion of the cost of a clinical trial, and thus the marginal benefit of vaccinating only a limited population may be small compared to the risk of underestimating overall disease

burden. Consequently, we support vaccination of and determination of burden within all age groups.

Target groups for outcome monitoring would differ slightly from those for vaccination and would include the entire population regardless of age. Specifically, it would be important to assess typhoid burden young infants, as well as other risk groups that may be excluded from immunization such as pregnant women. The main exception to this principle would be exclusion from primary analysis of persons unlikely to respond to vaccine due to immunosuppression, to avoid biasing disease burden estimates toward the null. The most important consideration is likely to be human immunodeficiency virus (HIV) infection, since HIV infection may reduce response to polysaccharide-protein conjugate vaccines (24). Consequently, HIV testing of patients presenting with fever would be an important consideration in areas of high infection prevalence.

SETTINGS

Typhoid is an enteric disease with complications associated with lack of health care access, i.e., it is primarily a disease of poverty. Consequently, probe studies should occur in low-income settings. Because the main typhoid burden exists in Southeast Asia and sub-Saharan Africa, studies should emphasize these locations. Important rural and urban differences may exist as well (6,9), and thus it will be important to include sites with both characteristics.

Study sites should have accurate data on populations (i.e., denominator data) and outcomes (i.e., numerator data). This requires having a study site with a population of known size that seeks care at a known and limited number of health care centers. This will usually eliminate the largest urban centers from consideration.

Demographic health surveillance (DHS) sites have some advantages, namely a population with extensive background data, known population, and known health seeking behavior. However, for practical reasons they have relatively small populations under surveillance. Moreover, frequent and intense community follow-up may alter risk for illness (e.g., populations under surveillance may have better adherence to sanitation principles) and severity (e.g., populations under surveillance may have increased health seeking behavior [9]) making them unrepresentative of the surrounding population. For these reasons, DHS sites may be inappropriate for probe studies.

The goal of targeting endemic areas is to provide data on vaccine utility in areas most likely to use vaccine, including through support from Gavi, the Vaccine Alliance. Consequently, we recognize the study results may not represent results globally. Conceptually, this does not differ from probe studies of Hib vaccine (e.g., results from Indonesia [3] differed from other areas of the world) or rotavirus (e.g., the large unmeasured community burden in Kenya [5] has not been replicated elsewhere). Additionally, conducting studies in several different health care and epidemiological contexts in Asia and Africa should frame the overall burden in endemic areas. Areas of potential high public health importance but with little available typhoid data (e.g., rural India) could be considered but as an initial step should document the existence of disease through traditional surveillance.

STUDY DURATION AND FREQUENCY OF FOLLOW-UP

Total population enrolled is the product of persons enrolled per year by study years, and study power can be addressed through increases in either of these factors. However, increasing the study area to shorten study duration may create logistical issues for study staff, and provides no margin of error for correcting issues identified early during study implementation. Additionally, a multi-year study can adapt to factors external to the study that may alter epidemiology, such as improvements or declines in water and sanitation, climatic events, or natural variations in disease incidence based on waxing and waning population immunity. For these reasons, we recommend a study of at least four years duration.

Community follow-up needs to balance identification of the majority of ill persons that fail to present to a health care center with not altering disease outcomes and thus reducing study representativeness. The precise balance for particular diseases is not well-defined, but based on a trial with rotavirus vaccine, monthly home visits may be appropriate (5).

VACCINE

Efficacy and effectiveness

Again, because the goal is not to define a practical immunization schedule, probe studies should use vaccines with the highest efficacy (assuming appropriate safety), even if issues exist with schedule or presentation. This approach will maximize the estimate of preventable disease burden. Most currently licensed vaccines do not provide direct protection for children age less than 2 years. An exception is Typbar typhoid conjugate vaccine produced by Bharat Biotech in India, which is licensed for use starting at age 6 months (25). Other polysaccharide-protein conjugate vaccines may provide higher efficacy and protect younger children (26,27). Additionally, polysaccharide vaccine may have relatively limited efficacy in some settings. For example, efficacy in India was 61% but less than 50% among older persons (18) while efficacy in Pakistan was 31% among children age 2–16 years (19).

Duration of immunity

Vaccine should provide protective immunity for the duration of the trial, using whatever schedule is necessary to achieve this goal. Protection beyond this is a public health concern but not a concern for documenting disease burden using a probe approach. ViPS has a three-year cumulative vaccine efficacy of 55% (22) while the conjugate Vi-rEPA conferred 87% protection during the second year (22,28). If duration of immunity is a concern, and the trial persists longer than 3 years, a second round of immunization may be needed.

Schedule

The schedule should be that which provides the highest level of protection within ethical, safety, and financial bounds, as well as limitations imposed by choice of control vaccine (see below). It is likely that a probe study would start with a mass catch-up campaign, following by routine infant immunization with a conjugate vaccine.

Coverage

The trial should seek to obtain the highest vaccine coverage possible to reduce transmission in the community as a whole. High vaccine coverage will limit assessment of indirect protection among target age groups. However, this is not a primary goal of the study. Moreover, indirect protection can be assessed among subgroups excluded from vaccination, such as young infants, the elderly, and pregnant women.

Control vaccine

The control vaccine should have no impact on syndromes associated with typhoid (primarily febrile illness) and should have a schedule compatible with typhoid vaccine. As used in malaria vaccine trials (16), serogroup C meningococcal conjugate vaccine represents one option given the global occurrence of this serogroup. Rabies vaccine represents another option, and may have appeal in rabies-endemic areas, which often overlap areas of typhoid endemicity (29). Recent dengue vaccine trials used placebo injections (30) and this represents a third option. While inactivated poliovirus vaccine (IPV) has the advantage that it likely will be licensed and introduced in all countries globally as part of global polio eradication efforts, in a polio-free world it offers little additional advantage over placebo.

Another option is to conduct a randomized, open-label trial with no control injection. The disadvantage of this approach is that knowledge of vaccination status may alter care-seeking behavior differentially among vaccinated and non-vaccinated populations, leading to differential bias of disease incidence estimations within the two groups. Because vaccine preventable disease incidence equals the product of vaccine effectiveness and control group incidence, this may then have an unknown impact on estimations of the primary study outcome. Consequently, we prefer an controlled, blinded study design.

SAMPLE SIZE

Since the underlying vaccine preventable disease incidence will not be known a priori, conceptually, the sample size for a probe study can be approached by identifying the level of disease considered of public health importance. In this respect, previous examples exist. For example, the Gambian trial found that PCV prevented 180 cases of severe clinical pneumonia per 100,000 child-years of follow-up, with vaccine effectiveness against severe disease of 12% (4).

An ancillary issue is the degree to which a probe study finds disease not identified through laboratory confirmation. In Indonesia, the probe approach found a vaccine preventable incidence of hospitalized Hib meningitis four- to five-fold higher than that identified with laboratory confirmation (3). A rotavirus vaccine trial using a probe approach found six-fold more preventable disease in the community than presenting to the clinic (5).

An approach to reducing sample size requirements is to identify syndromic outcomes enriched for the target etiology. For example, Hib vaccine and PCV studies relied on pneumonia with radiologic confirmation of an alveolar consolidation or pleural effusion. For typhoid, prolonged fever as the primary outcome could serve this purpose. However, the all-age incidence of hospitalizations for febrile illness of greater than 5 days duration is poorly

defined in low-income settings. One of the current authors, during re-analysis of an earlier typhoid vaccine trial (19) found an annual incidence of 1000 per 100,000 patients of all ages. Other studies have estimated hospitalization for fever of any duration. For example, a study in Kenya found an annual incidence of 6825 per 100,000 persons age 5 years or older for conditions unrelated to injury or obstetrics, with an incidence of 1193 per 100,000 for fever alone, most of whom were hospitalized (31).

Finally, one can start with confirmed typhoid incidence and assume that true burden is some multiple of this value. In two cluster-randomized trials, the incidences of confirmed typhoid were estimated as approximately 200 to 300 per 100,000 person-years for ages 2 years and older in India (18) and ages 2–16 years in Pakistan (19).

Using the above data and methods, for base-case analysis, we estimated an incidence of hospitalization for prolonged febrile illness of 1000 per 100,000 population of which 25% would be prevented by typhoid vaccine. This would yield an underlying typhoid burden similar to that for documented cases in Pakistan and India and thus represents a conservative estimate. Nevertheless, it assumes a vaccine preventable disease incidence on a par with PCV in the Gambia.

In sum, the following parameters were used:

1. Annual incidence of prolonged fever hospitalizations per 100,000: 1000 base-case, 500 lower bound, 2000 upper bound.
2. Vaccine efficacy against fever: 25% base-case, 15% lower bound
3. Lower bound of the 95% CI for vaccine efficacy: base-case of 0%.
4. Study power: 80%
5. Loss to follow-up: 15%.
6. Number of people per cluster: base-case 100, lower bound 50, upper bound 500.
7. Coefficient of variation: base-case 0.3, upper bound 0.5.
8. Ratio of vaccinated to unvaccinated persons: 1 to 1.
9. Study duration: 4 years.

Illustrative sample size calculations for the primary outcome were based on previously published work (32,33) and are shown in Table 1. Base-case calculations yielded sample size estimates similar to probe studies of other vaccines (3–5). However, estimates were highly dependent on the parameters used, varying from 4,150 to 113,500 persons per arm. As expected, estimated vaccine effectiveness and background incidence had major impact on sample size estimates. Little difference in estimates was seen for cluster sizes of 50 versus 100, but this variable became important at a cluster size of 500. Similarly, when cluster size was larger and the number of clusters smaller, the coefficient of variation became important. These data emphasize the importance of having pre-study data on vaccine effectiveness, background incidence, and a realistic number and size of clusters.

ETHICS

Randomized trials of new vaccines are ethically acceptable because the vaccine's ability to prevent disease is unknown. Vaccine probe studies of an effective vaccine may be ethically acceptable because the community disease burden is unknown and may not be high enough to justify a public health immunization program; this latter situation existed in Indonesia at the time a Hib vaccine probe study was conducted (11). However, where an effective vaccine exists and disease burden is known or suspected to be high, even if imprecisely defined, implementation of a randomized trial becomes more problematic. Currently, countries have been reticent to introduce typhoid vaccine into public immunization programs. Additionally, Gavi has not committed financial support for typhoid vaccine introduction. While several reasons for this exist, one key reason is uncertainty regarding whether sufficient disease burden exists.

CONCLUSIONS

A typhoid vaccine probe study should be feasible under most scenarios. Such a trial could address many of the current limitations in estimating overall typhoid burden. This in turn would provide the data needed by national governments and immunization advisory bodies to determine the relative value of typhoid vaccine in public immunization programs. For eligible countries, these data will help inform the decision of Gavi to support typhoid vaccine use. Finally, a vaccine probe study can provide valuable anchoring data for interpretation of studies with less robust design.

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References

1. Feikin DR, Scott JAG, Gessner BD. Use of vaccines as probes to define disease burden. *Lancet*. 2014; 383:1762–70. [PubMed: 24553294]
2. Gessner BD, Feikin DR. Vaccine preventable disease incidence as a complement to vaccine efficacy for setting vaccine policy. *Vaccine*. 2014; 32:3133–8. [PubMed: 24731817]
3. Gessner BD, Sutanto A, Linehan M, et al. Incidences of vaccine-preventable *Haemophilus influenzae* type b pneumonia and meningitis in Indonesian children: hamlet-randomised vaccine-probe trial. *Lancet*. 2005; 365:43–52. [PubMed: 15643700]
4. Cutts FT, Zaman SM, Enwere G, et al. Efficacy of nine-valent pneumococcal conjugate vaccine against pneumonia and invasive pneumococcal disease in The Gambia: randomised, double-blind, placebo-controlled trial. *Lancet*. 2005; 365:1139–46. [PubMed: 15794968]
5. Feikin DR, Laserson KF, Ojwando J, Nyambane G, Ssempijja V, Audi a, et al. Efficacy of pentavalent rotavirus vaccine in a high HIV prevalence population in Kenya. *Vaccine*. 2012; 30S:A52–60. [PubMed: 22520137]
6. Mogasale V, Maskery B, Ochiai RL, Lee JS, Mogasale VV, Ramani E, et al. Burden of typhoid fever in low-income and middle-income countries: a systematic, literature-based update with risk-factor adjustment. *Lancet Glob Health*. 2014; 2:e570–80. [PubMed: 25304633]

7. Buckle GC, Walker CLF, Black RE. Typhoid fever and paratyphoid fever: systematic review to estimate global morbidity and mortality for 2010. *J Glob Health*. 2012; 2:010401. [PubMed: 23198130]
8. GBD 2013 Mortality and Causes of Death Collaborators. Global, regional, and national age-sex specific all-cause and cause-specific mortality for 240 causes of death 1990–2013: a systematic analysis for the Global Burden of Disease Study 2013. *Lancet*. 2015; 385:117–71. [PubMed: 25530442]
9. Breiman RF, Cosmas L, Njuguna H, Audi a, Olack B, Ochieng JB, et al. Population-based incidence of typhoid fever in an urban informal settlement and a rural area in Kenya: implications for typhoid vaccine use in Africa. *PLoS One*. 2012; 7:e0029119.
10. Marks F, Adu-Sarkodie Y, Hunger F, Sarpong N, Ekuban S, Agyekum A, et al. Typhoid fever among children, Ghana. *Emerg Infect Dis*. 2010; 16:1796–7. [PubMed: 21029549]
11. World Health Organization. Immunization and Vaccine related Implementation Research Advisory Committee (IVIR-AC): summary of conclusions and recommendations 17–19 September 2014 meeting. *Wkly Epidemiol Rep*. 2015; 90:1–8.
12. Wain J, Hendriksen RS, Mikoleit ML, Keddy KH, Ochiai RL. Typhoid fever. *Lancet*. 2014 Epub.
13. Kothari A, Pruthi A, Chugh TD. The burden of enteric fever. *J Infect Dev Countries*. 2008; 2:253–9.
14. Sinha A, Sazawal S, Kumar R, Sood S, Reddalah VP, Singh B, et al. Typhoid fever in children aged less than 5 years. *Lancet*. 1999; 354:734–7. [PubMed: 10475185]
15. Mogasale V, Desai SN, Mogasale VV, Park JK, Ochiai RL, Wierzbza. Case fatality rate and length of hospital stay among patients with typhoid intestinal perforation in developing countries: a systematic literature review. *PLoS One*. 2014; 9:e93784. [PubMed: 24743649]
16. The RTSS Clinical Trials Partnership. First results of phase 3 trial of RTS,S/AS01 malaria vaccine in African children. *N Engl J Med*. 2011; 365:1863–75. [PubMed: 22007715]
17. Khatib AM, Ali M, von Seidlein L, Kim DR, Hashim R, Reyburn R, et al. Effectiveness of an oral cholera vaccine in Zanzibar: findings from a mass vaccination campaign and observational cohort study. *Lancet Infect Dis*. 2012; 12:837–44. [PubMed: 22954655]
18. Sur D, Ochiai RL, Bhattacharya SK, Ganguly NK, Ali M, et al. A cluster-randomized effectiveness trial of Vi typhoid vaccine in India. *N Engl J Med*. 2009; 361:335–44. [PubMed: 19625715]
19. Khan MI, Soofi SB, Ochiai RL, Habib MA, Sahito SM, et al. Effectiveness of Vi capsular polysaccharide typhoid vaccine among children: a cluster randomized trial in Karachi Pakistan. *Vaccine*. 2012; 30:5389–95. [PubMed: 22721899]
20. Pitzer VE, Bowles CC, Baker S, Kang G, Balaji V, Farrar JJ, et al. Dynamics of typhoid in South Asia: a mathematical modeling study. *PLoS Negl Trop Dis*. 2014; 8:e2642. [PubMed: 24416466]
21. Moulton LH, Chung SE, Croll J, Reid R, Weatherholtz RC, Santosham M. Estimation of the indirect effect of *Haemophilus influenzae* type b conjugate vaccine in an American Indian population. *Int J Epidemiol*. 2000; 29:753–6. [PubMed: 10922355]
22. Anwar E, Goldberg E, Fraser A, Acosta CJ, Paul M, Leibovici L. Vaccines for preventing typhoid fever (review). *Cochrane Database Syst Rev*. 2014; 1:CD001261. [PubMed: 24385413]
23. Sur D, Lopez AL, Kanungo S, Paisley A, Manna B, Ali M, et al. Efficacy and safety of a modified killed-whole-cell oral cholera vaccine in India: an interim analysis of a cluster-randomised, double-blind, placebo-controlled trial. *Lancet*. 2009; 374:1694–702. [PubMed: 19819004]
24. Madhi SA, Levine OS, Hajjeh R, Mansoor OD, Cherian T. Vaccines to prevent pneumonia and improve child survival. *Bull World Health Organ*. 2008; 86:365–72. [PubMed: 18545739]
25. Bharat Biotech. [last accessed March 16, 2015] Typbar TCV package insert. Website : <http://www.bharatbiotech.com/pdf/Typbar-TCV%20Pack%20Insert.pdf>
26. Thiem VD, Lin FY, Canh DG, Canh G, Son NH, et al. The Vi conjugate typhoid vaccine is safe, elicits protective levels of IgG anti-Vi, and is compatible with routine infant vaccines. *Clin Vaccine Immunol*. 2011; 18:730–5. [PubMed: 21411598]
27. Bhutta Z, Capeding MR, Bavdekar A, Marchetti E, Ariff S, Soofi SB. Immunogenicity and safety of the Vi-CRM197 conjugate vaccine against typhoid fever in adults, children, and infants in south and southeast Asia: results from two randomized, observer-blind, age de-escalation, phase 2 trials. *Lancet Infect Dis*. 2014; 14:119–29. [PubMed: 24290843]

28. Lin FY, Ho VA, Khiem HB, Trach DD, Bay PV, Thanh TC, et al. The efficacy of a *Salmonella* typhi Vi conjugate vaccine in two-to-five-year-old children. *N Engl J Med*. 2001; 344:1263–9. [PubMed: 11320385]
29. Fooks AR, Banyard AC, Horton DL, Johson N, McElhinney LMm, Jackson AC. Current status of rabies and prospects for elimination. *Lancet*. 2014; 384:1389–99. [PubMed: 24828901]
30. Villar L, Dayan GH, Arredondo-Garcia JL, et al. Efficacy of a tetravalent dengue vaccine in Latin America. *N Engl J Med*. 2015; 372:113–23. [PubMed: 25365753]
31. Feikin DR, Jagero G, Aura B, Bigogo GM, Oundo J, Beall BW, et al. High rate of pneumococcal bacteremia in a prospective cohort of older children and adults in an area of high HIV prevalence in rural western Kenya. *BMC Infect Dis*. 2010; 10:186. [PubMed: 20573224]
32. Hayes RJ, Bennett S. Simple sample size calculation for cluster-randomized trials. *Int J Epidemiol*. 1999; 28:3193–6.
33. Hayes, RJ.; Moulton, LH. *Cluster Randomised Trials: A Practical Approach*. Chapman and Hall/CRC; Boca Raton, FL: 2009.

KEY POINTS

1. Typhoid disease burden remains poorly defined, preventing decisions on vaccine use.
2. Vaccine probe studies have catalyzed use of other new vaccines including Hib and pneumococcal conjugate vaccines.
3. A typhoid vaccine probe study would address many of the current questions regarding typhoid disease burden.
4. Such a study would be feasible over a reasonable set of assumptions.

Table 1

Sample size calculations for a hypothetical four-year typhoid vaccine probe study with number of clusters per arm (and total persons per arm) reported for varying levels of annual prolonged fever hospitalization incidence per 100,000 population (I), vaccine effectiveness (VE), cluster size, and coefficient of variation (CoV). Values reported are for each arm of the trial with a 1:1 ratio among vaccinated and unvaccinated populations. We assumed a dropout rate of 15% and a lower bound of the 95% confidence interval for VE of 0%.

		VE = 25%			VE=15%		
		I=500	I=1000	I=2000	I=500	I=1000	I=2000
50 per cluster	CoV = 0.3	277 (13850)	148 (7400)	83 (4150)	813 (40650)	434 (21700)	245 (12250)
	CoV = 0.5	308 (15400)	179 (8950)	115 (5750)	909 (45450)	530 (26500)	341 (17050)
100 per cluster	CoV = 0.3	148 (14800)	105 (10500)	51 (5100)	434 (43400)	245 (24500)	150 (15000)
	CoV = 0.5	179 (17900)	115 (11500)	82 (8200)	530 (53000)	341 (34100)	246 (24600)
500 per cluster	CoV = 0.3	44 (22000)	32 (16000)	25 (12500)	131 (65500)	93 (46500)	74 (37000)
	CoV = 0.5	76 (38000)	63 (31500)	56 (28000)	227 (113500)	189 (94500)	170 (85000)